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Interactive Effects of *Ascophyllum nodosum* Seaweed Extract and Silicon on Growth, Fruit Yield and Quality, and Water Productivity of Tomato under Water Stress

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Abstract

Agricultural crops including tomato (Solanum lycopersicum L.) are severely affected by drought, which is a critical abiotic stress. Biostimulants, such as Ascophyllum nodosum seaweed extract (ASE), and silicon (Si) are independently used in alleviating drought stress and in enhancing growth and productivity of agronomic and horticultural crops. The present study was conducted to assess the combined effects of ASE and Si on growth, fruit yield, fruit quality, and water productivity of tomato under water stress. Five doses of ASE $(0, 1.25, 2.5, 3.75, \text{ and } 5 \text{ mL L}^{-1})$ were applied in combination with 60 kg ha⁻¹ soluble Si in the form of monosilicic acid (as soil incorporation regardless of ASE doses) along with a control (where no ASE or Si was applied) under three soil moisture regimes of 50%, 75%, and 100% field capacity (FC). Data on growth, fruit yield, water productivity, fruit quality, and physio-biochemical parameters of tomato were collected. The results revealed that severe water stress of 50% FC negatively affected growth, physiological traits, and fruit yield of tomato (43–80% lower yield across ASE doses) compared with those at 100% FC, whereas fruit quality parameters (total soluble solids, fruit firmness, color index, and fruit pH) increased with reduced soil moisture regime. Application of ASE at 3.75 and 5 mL L^{-1} in combination with soluble Si at 60 kg ha⁻¹ resulted in statistically similar fruit yields under a sufficient soil moisture level of 100% FC and a moderate soil moisture level of 75% FC, respectively. A consistent trend of higher fruit yield and water productivity was observed for plants supplemented with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ soluble Si regardless of soil moisture regimes. Similarly, individual Si supplementation at 60 kg ha⁻¹ was also effective and caused 207% increase in fruit yield even at severe water stress of 50% FC compared with the control. However, a combined application of ASE and Si had more promising results than the sole application of Si. Exogenous soil application of ASE at 5 mL L^{-1} along with soluble Si at 60 kg ha⁻¹ holds promise for tomato production under moderate to sufficient soil moisture availability.

Keywords Beneficial element · Biostimulant · Drought · Monosilicic acid · Solanum lycopersicum L

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1 Introduction

Tomato (*Solanum lycopersicum* L.) is an important vegetable crop that occupies the second largest vegetable-producing area of the world [1]. It is extensively farmed and consumed across the world for its rich source of minerals, vitamins, and bioactive compounds that include a significant amount of vitamin C (ascorbic acid), vitamin E (tocopherol), carotenoids (β -carotene and lycopene), and phenolic compounds responsible for protecting human body from free radicals and tumor cells [2–5]. Tomato is most commonly grown in tropical, subtropical, and temperate climatic zones; however, water scarcity, one of the most critical abiotic stresses, can reduce marketable fruit yields by up to 80% [6]. Its vulnerability to water stress necessitates management strategies



aiming at maintaining satisfactory levels of production under water-scarce environments. The use of different types of exogenous protectants, such as osmoprotectants (proline, glycine betaine, trehalose), phytohormones (salicylic acid, jasmonic acid, gibberellic acid, brassinosteroids), antioxidants (ascorbic acid, glutathione, tocopherols), signaling molecules (hydrogen peroxide, nitric oxide), polyamines (putrescine, spermidine, spermine), and trace elements (silicon [Si], selenium), has been found beneficial in protecting plants against water stress [7].

Drought is a serious environmental stress that severely limits growth and production of all major agronomic and horticultural crops [8–11]. Drought frequency is projected to increase significantly in many parts of the world because of global warming and climate change [12]. Drought stress has adverse effects on plant growth and development by impairing cell division, cell enlargement, and cell differentiation, as well as genetic, physiological, ecological, and morphological processes and their complex relationships [13]. All these events have a negative impact on morphological and physio-biochemical processes in plants, including stomatal conductance, carbon dioxide diffusion, membrane electron transport, carboxylation efficiency, transpiration, respiration, water loss, water use efficiency, photosynthesis, and membrane functions. Drought stress causes oxidative stress by increasing the generation of reactive oxygen species, such as superoxide anion radicals, singlet oxygen, hydrogen peroxide, and hydroxyl radicals [13]. This oxidative stress damages cells and their components and slows down plant's physio-biochemical life processes through increased peroxidation of membrane lipids and degradation of proteins and nucleic acids [13], resulting in cell death [14, 15]. Plants have a variety of stress tolerance and/or avoidance mechanisms, of which the most essential are osmotic adjustment and effective antioxidant system. The increased synthesis of proline, glycine betaine, and other metabolites with structural characteristics to maintain homeostasis and improve plant functioning under drought stress indicates osmotic adjustment [16]. The capacity of plants to strengthen their stress defense mechanisms is also dependent on their ability to synthesize secondary metabolites with high antioxidant activity, such as phenolic compounds [17]. Exogenous administration of biostimulants is one possible approach for improving plant tolerance to drought stress, but other strategies, such as plant breeding and genetic engineering, have also been shown to be effective in reducing the effects of drought stress in plants [18–20].

Seaweed is a rich source of growth-promoting hormones, nutrients, vitamins, and amino acids, and its extract has been reported to enhance plant's defense system against stress [21, 22]. Among different groups of seaweeds (brown, red, and green), brown seaweeds are the most extensively utilized in agriculture as biofertilizers

[23–25], and Ascophyllum nodosum (L.) Le Jolis is the most studied species [26]. Numerous types of seaweed extracts are presently available for commercial agriculture, most notably for vegetable production [27]. It has been demonstrated that Ascophyllum nodosum seaweed extract (ASE) improves the drought tolerance capacity of container-grown sweet orange [Citrus sinensis (L.) Osbeck] plants [28]. Different plant species respond differently to seaweed concentrations, application techniques, and application doses [25, 28]. In addition, ASE influences the physical, chemical, and biological qualities of soil, which affect plant growth and development. The application of ASE improves soil health by increasing the ability of the soil to retain more moisture and by stimulating the growth of beneficial soil microorganisms. Brown seaweeds (ASE) have a high concentration of polyuronides, including alginates and fucoidans. Various betaines and betaine-like substances are present in ASE [23, 25, 29]. Betaines operate as a compatible solute in plants, alleviating osmotic stress caused by drought and salinity [23, 25]. Other beneficial roles associated with ASE application include increasing plant leaf chlorophyll content [30], which might be attributed to a reduction in chlorophyll breakdown [31]. The betaines present in ASE have been linked to increased yields owing to increased chlorophyll content in the leaves of several agricultural crops [30, 31].

The exogenous application of Si is also a promising approach to improve yield and quality of different crops [32]. Plant growth and development might encounter various challenges due to an insufficient availability of Si in the soil. This makes it a 'quasi-essential' element for plants. The positive function of Si against several abiotic stresses, such as water stress [8, 10, 11, 33–40], salt stress [41], radiation stress [35], freezing stress [42], and heavy metal toxicity [43], is well documented. Silicon-treated plants exhibit increased stomatal conductance and transpiration rate, leaf relative water content (LRWC), and root and whole-plant hydraulic conductivity [44]. Exogenous application of Si under water stress helps plants maintain certain physiological processes, such as keeping a higher LRWC [45], improving plant water relations with changes in osmotic pressure [33, 35, 46, 47], and adjusting proline levels [35].

Although the individual effect of ASE and Si on growth and productivity of various crops has been well documented under water stress, no literature is available evaluating their interactive role on tomato grown under water stress. It was hypothesized that the combined application of ASE and Si would exert a synergistic effect and would help maintain growth and productivity of tomato under water stress. The objective of the present study was to assess the interactive effects (synergistic) of ASE and Si on tomato growth, fruit yield, water productivity, and fruit quality under water stress.



2 Materials and Methods

2.1 Experimental Setup and Growth Condition

A pot experiment was conducted from July to November 2021 at the Department of Food, Agriculture and Bioresources, Asian Institute of Technology (latitude 14°04′53" N and longitude 100°36′33" E, 2.27 m above mean sea level), Klong Luang, Pathum Thani, Thailand. Seeds of Season 9, a hybrid Thai tomato cultivar, were grown in plastic pots under a naturally-lighted polyhouse. The relative humidity and temperature inside the polyhouse fluctuated between 70 and 75% and 28 °C and 34 °C, respectively, during the entire study period. The soil, which is classified as Bangkok clay soil containing 61% clay, 17% silt, 22% sand, 2.5% organic matter, and having an acidic pH of 5.2 (1:1 water), was collected from the experimental field (0-20 cm depth) of the Department. The soil was sun dried for five days, followed by crushing the large particles and removing the undesirable materials, such as coarse fragments, stones, pebbles, plant roots, and debris, and then 15 kg of the thoroughly-crushed soil was filled into each plastic pot (dimension: 30 cm height × 36 cm top diameter × 28 cm bottom diameter). The commercial seaweed extract used in the experiment was an alkaline hydrolysis product from A. nodosum (Amino Seaweed, SV Group, Bangkok, Thailand). According to the seaweed extract product specification (Amino Seaweed, SV Group, Bangkok, Thailand), ASE possesses the following physicochemical properties: 44% organic matter, > 13% alginic acid, 2.1% total N, 15.9% K, 2.7% P, and a small amount of amino acid, vitamins, and minerals. Silicon was applied as monosilicic acid (obtained from a local fertilizer shop marketed by Thai Green Agro Co. Ltd. with a Si content of 20%) at 60 kg ha⁻¹, which is equivalent to 300 kg ha⁻¹ monosilicic acid [8, 10]. Other fertilizers were applied at $112.5 \text{ kg N ha}^{-1}$, $50 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$, and $100 \text{ kg K}_2\text{O ha}^{-1}$ as recommended by the Department of Agriculture, Royal Thai Government for growing tomato. The respective fertilizer dosage was urea at 244.6 kg ha⁻¹ (1.83 g pot⁻¹), triple superphosphate at 108.7 kg ha⁻¹ (0.81 g pot⁻¹), and potassium chloride at 166.7 kg ha⁻¹ (1.24 g pot⁻¹). A total of 50% urea, 100% triple superphosphate, and 100% potassium chloride was applied as a basal dose thoroughly mixed with soil seven days prior to transplanting seedlings into the main pot. The remaining 50% urea was applied 30 days after transplanting. Following transplanting, all pots were watered daily for two weeks to ensure optimal seedling establishment. Later, an artificial water stress was imposed based on soil moisture content by withholding irrigation until the appropriate level of soil moisture was reached. Enough trellises were utilized to support the plants and fruits throughout the season of heavy bearing. Disease and insect pests were controlled according to the standard methods as recommended by the Department of Agriculture, Royal Thai Government.

2.2 Experimental Design and Treatments

A factorial experiment containing four replications using five ASE doses (0, 1.25, 2.5, 3.75, and 5 mL L^{-1}) applied in combination with 60 kg ha⁻¹ soluble Si as soil amendment along with a control (where no ASE or Si was applied) and three soil moisture regimes (50%, 75%, and 100% field capacity-FC) was laid out in a completely randomized design. Soil moisture content was determined using the gravimetric method [48]. Soil moisture contents at 100%, 75%, and 50% FC were determined as 46%, 35%, and 23%, respectively. For germination, seeds were placed in small trays with sterilized peat moss substrate. One healthy and strong seedling was transplanted into each pot at the twoleaf stage (21-day old) and each pot with one seedling was considered as one treatment replication. The plants were climate-hardened for two weeks before the required doses of ASE were sprayed uniformly on soil at the five-leaf stage (35 days after sowing) in the early morning [49]. The application of ASE was made to the soil once a week for six weeks using 100 mL of solution in each pot [50]. The control plants received no ASE and Si application. The application of Si was made to each pot (excluding the control) at 0.45 g pot⁻¹ (equivalent to 2.25 g pot⁻¹ monosilicic acid) one day before transplanting. Throughout the crop growth period, a portable soil moisture meter (SM150 Soil Moisture Sensor; SM150, Delta-T Devices Ltd., Cambridge, UK) was utilized to measure soil moisture on a daily basis. When the soil moisture level in each pot declined below the intended level of FC, the soil moisture level in each pot was corrected to the appropriate level by irrigating the pots.

2.3 Data Collection

2.3.1 Growth, Fruit Yield Parameters, and Water Productivity

Data on plant height (cm) and leaf area (cm² plant⁻¹) were collected at maturity stage (90 days after sowing). A measuring tape was used to measure plant height from the soil surface to the tip of the farthest leaf. Individual plant leaf area was calculated nondestructively from leaf width and leaf length following the method of Blanco and Folegatti [51]. After fruit harvest, fresh shoot and root samples were ovendried at 72 °C until a consistent weight was obtained, and shoot dry matter (g plant⁻¹) and root dry matter (g plant⁻¹) were measured. At harvest, data on fruit yield (g plant⁻¹) and number of fruits per plant were gathered. According to Ullah



et al. [52] and Maneepitak et al. [53], water productivity (kg m⁻³) was estimated by dividing fruit yield (kg) by total irrigation water input (m⁻³).

2.3.2 Fruit Quality Parameters

At harvest, measurements of fruit length (cm) and fruit width (cm) were taken. A pH meter (Model FiveGo F2, Mettler-Toledo GmbH, Im Langacher, Greifensee, Switzerland) was used to determine fruit pH. Total soluble solids (TSS) content of juice was determined using a refractometer (Model HI96801, Hanna Instruments, Woonsocket, RI, USA) after the fruits were homogenized in a blender. The color of the fruit surface was measured with a Colorimeter (ColorFlex, Model 45/0, HunterLab, Reston, VA, USA) and the color space coordinates *L*, *a*, and *b* of the Commission Internationale de l'Eclairage (CIE) were recorded. According to Hobson et al. [54] and Chen et al. [55], fruit color index was determined using the following equation:

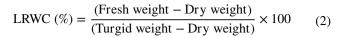
Color index =
$$\frac{2000a}{L\sqrt{(a^2 + b^2)}}$$
 (1)

where L denotes the lightness, a denotes the coloration intensity ranging from – greenness to + redness, and b denotes the coloration intensity ranging from – blueness to + yellowness.

A Fruit Texture Analyzer (GUSS, Model No. GS25, SA) was used to determine fruit firmness. For this measurement, a stainless-steel cylindrical probe with a flat end having a diameter of 8 mm was utilized. At a speed of 5 mm s⁻¹, the probe was inserted into tomatoes to a depth of 3 mm (the same spot for each sample). The highest penetrating force was utilized to determine fruit firmness, and the data were collected from five whole tomatoes (two opposing spots in each tomato) under each treatment combination.

2.3.3 Physio-biochemical Parameters

Leaf greenness (relative chlorophyll content) was assessed nondestructively from completely-grown young leaves using a handheld chlorophyll meter (SPAD-502 Plus; Minolta Co. Ltd., Osaka, Japan). Individual leaves were cut from the midsection of each plant, preserved in plastic bags, and fresh weight was measured immediately after sampling. After that, the leaves were sliced into short segments (5 cm), immersed in distilled water on Petri dishes for 24 h in the dark, and the turgor weight of the samples was determined. The completely turgid leaf samples were oven-dried at 70 °C until they reached a consistent weight, and the dry weight was measured. Leaf relative water content (LRWC) was calculated using the following equation (Jones and Turner [56]):



With slight adjustments, electrolyte leakage was measured as described by Camejo et al. [57]. For each treatment, the plant's fully-expanded leaf was utilized. For 60 min at 25 °C, samples were put in capped vials containing 10 mL of deionized water. By measuring the electrical conductivity (EC) of water after 60 min (EC₁) and after breaking cell membranes by heating the samples at 100 °C for 30 min (EC₂), the percent electrolyte leakage of the sample was calculated. A conductivity meter (Model Eutech CON 150, Thermo Scientific, Eutech Instruments, Singapore) was used to determine the solution's EC. The following equation was used to determine electrolyte leakage:

Electrolyte leakage (%) =
$$\frac{EC_1}{EC_2} \times 100$$
 (3)

Cell membrane stability was calculated by measuring the conductivity of leachates caused by damaged plasma membranes as described by Shanahan et al. [58]. One gram of leaf material (10 mm pieces) was placed in glass vials with 10 mL of distilled water and shaken for 24 h at 10 °C. After heating the sample at 25 °C, the initial conductivity (C_1) was measured using a conductivity meter (Model Eutech CON 150, Thermo Scientific, Eutech Instruments, Singapore). The samples were then kept in a boiling water bath (100 °C) for 10 min, cooled to 25 °C, and the final conductivity (C_2) was measured. The following equation was used to determine membrane stability index:

Membrane stability index (%) =
$$(1 - \frac{C_1}{C_2}) \times 100$$
 (4)

A portable photosynthesis system (LI-6400XT, Li-COR, Lincoln, NE, USA) was used to measure leaf gas exchange parameters, such as net photosynthetic rate (P_n), stomatal conductance (g_s), and transpiration rate (E), of the upper leaf between 10.30 and 11.30 am at the fruiting stage. There was about $370 \pm 20 \, \mu \text{mol mol}^{-1}$ of CO_2 in the air when the assimilation chamber was set up. The ambient temperature was 28 ± 1 °C. Artificial lighting from a red- blue 6400-02B LED light supply that could distribute continuous light at 1,000 μ mol m⁻¹ s⁻¹ photosynthetic photon flux density was applied during the measurements (Cha-um et al. [59]).

The amount of free proline (mg g⁻¹ fresh weight) was measured following Bates et al. [60] quick colorimetric technique. Proline was extracted from 250 mg of fresh leaf sample in 10 mL of 3% sulfosalicylic acid. The mixtures were then centrifuged for 10 min at 3,000 rpm. In a test tube, 2 mL of the supernatant was combined with 2 mL of newly-made acid ninhydrin solution. The tubes were placed in a water bath at 90 °C for 30 min and the reaction was



stopped in an ice bath. A total of 5 mL toluene was added to each reaction mixture and vortex-mixed for 15 s. To allow the toluene and aqueous phases to separate, the tubes were left in the dark for at least 20 min at room temperature. The absorbance of each toluene phase was properly collected into a clean test tube and measured at 520 nm. Free proline content in each sample was determined from a standard curve prepared using analytical grade proline.

2.4 Statistical Analysis

The data of all the parameters (growth, fruit yield, total water input, water productivity, fruit quality, physiological, and biochemical) were collected from four biological replicates for each treatment combination. The data were subjected to two-way analysis of variance (ANOVA) and were statistically analyzed utilizing Statistix 8 (Analytical Software, Tallahassee, FL, USA) software program. Differences between means were compared by the least significant difference (LSD) test at $P \le 0.05$.

Table 1 Significance levels in two-way ANOVA of the interactive effects of Ascophyllum nodosum seaweed extract (ASE) applied in combination with silicon (Si) and soil moisture regime on growth, fruit yield parameters, total water input, water productivity, fruit quality parameters, and physiobiochemical parameters of tomato

Items	ASE+Si	Soil moisture level (M)	(ASE+Si)×M
Growth parameters			
Plant height (cm)	**	**	**
Leaf area (cm ² plant ⁻¹)	**	**	**
Shoot dry matter (g plant ⁻¹)	**	**	**
Root dry matter (g plant ⁻¹)	**	**	**
Fruit yield parameters, total water input, water prod	ductivity		
Fruit yield (g plant ⁻¹)	**	**	**
Number of fruits per plant	**	**	**
Total water input (m ³ plant ⁻¹)	**	**	ns
Water productivity (kg m ⁻³)	**	**	*
Fruit quality parameters			
Fruit length (cm)	**	**	ns
Fruit width (cm)	**	**	ns
Total soluble solids (°Brix)	**	**	ns
Fruit firmness	**	**	ns
Color index	**	**	ns
Fruit pH	**	**	**
Physio-biochemical parameters			
Leaf greenness (SPAD value)	**	**	**
Leaf relative water content (%)	**	**	**
Electrolyte leakage (%)	**	**	**
Membrane stability index (%)	**	**	**
Net photosynthetic rate (μmol CO ₂ m ⁻² s ⁻¹)	**	**	**
Stomatal conductance (mmol H ₂ O m ⁻² s ⁻¹)	**	**	**
Transpiration rate (mmol H ₂ O m ⁻² s ⁻¹)	**	**	**
Free proline content (mg g ⁻¹ fresh weight)	**	**	**

^{**, *,} and ns indicate $P \le 0.01$, $P \le 0.05$, and not significant, respectively

3 Results

3.1 Growth Parameters

The interactive effect of ASE applied in combination with Si and soil moisture regime was significant for plant height, leaf area, shoot dry matter, and root dry matter (Table 1). The overall performance of tomato plants supplemented with 5 mL L⁻¹ ASE in combination with 60 kg ha⁻¹ Si was better than all other ASE doses. The control plants and plants supplemented with 60 kg ha⁻¹ Si without ASE had largely similar performance, especially at lower soil moisture regimes of 50% and 75% FC in terms of plant height, shoot dry matter, and root dry matter (Table 2). Plant height, leaf area, shoot dry matter, and root dry matter were reduced by 29–37%, 49–70%, 37–48%, and 46–61% across soil moisture regimes for the control plants compared with those parameters of plants grown with the combined application of 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si. Decreasing soil moisture level resulted in a significant reduction in all growth parameters



Table 2 Effects of *Ascophyllum nodosum* seaweed extract (ASE) (0, 1.25, 2.5, 3.75, and 5 mL L⁻¹) applied in combination with silicon (Si) (60 kg Si ha⁻¹) and soil moisture regime on growth parameters of tomato

Factor	Plant height (cm)	Leaf area (cm ² plant ⁻¹)	Shoot dry matter (g plant ⁻¹)	Root dry matter (g plant ⁻¹)
$\overline{\text{ASE (mL L}^{-1}) + \text{Si (kg ha}^{-1})}$				
$ASE_0 + Si_0$	78.4 ± 4.11 f	$17,916 \pm 1684e$	$18.7 \pm 0.89e$	$2.1 \pm 0.24 \mathrm{f}$
$ASE_0 + Si_{60}$	$83.7 \pm 4.41e$	$23,058 \pm 1868d$	$20.4 \pm 0.90d$	$2.4 \pm 0.21e$
$ASE_{1.25} + Si_{60}$	$90.9 \pm 4.11d$	$29,276 \pm 2288c$	$27.4 \pm 1.11c$	$3.2 \pm 0.33d$
$ASE_{2.5} + Si_{60}$	$97.0 \pm 4.53c$	$31,606 \pm 2681c$	$28.8 \pm 1.79c$	$3.6 \pm 0.27c$
$ASE_{3.75} + Si_{60}$	$101.8 \pm 4.37b$	$37,187 \pm 3859b$	31.2 ± 1.84 b	4.0 ± 0.34 b
$ASE_5 + Si_{60}$	$117.0 \pm 7.01a$	$48,429 \pm 5697a$	$34.0 \pm 2.21a$	$4.6 \pm 0.39a$
Soil moisture regime (M)				
M ₅₀ -50% field capacity	$74.1 \pm 2.05c$	$17,943 \pm 978c$	$20.3 \pm 0.80c$	$2.1\pm0.14c$
M ₇₅ -75% field capacity	103.2 ± 3.60 b	$35,533 \pm 2827b$	28.9 ± 1.33 b	$3.5 \pm 0.22b$
M ₁₀₀ -100% field capacity	$107.1 \pm 3.05a$	$40,260 \pm 2724a$	$30.9 \pm 1.50a$	$4.4\pm0.22a$
$(ASE+Si)\times M$				
$(ASE_0 + Si_0) \times M_{50}$	$60.2 \pm 0.85i$	$11,239 \pm 524 \text{ m}$	$14.6 \pm 0.36 \text{ k}$	$1.1 \pm 0.02 \text{ k}$
$(ASE_0 + Si_0) \times M_{75}$	$82.2 \pm 0.85 \text{ fg}$	$17,725 \pm 275$ kl	$20.2 \pm 0.52j$	$2.1 \pm 0.02i$
$(ASE_0 + Si_0) \times M_{100}$	92.7 ± 1.11 de	$24,785 \pm 495 \text{ h}$	$21.1 \pm 0.27 ij$	$3.1 \pm 0.03 \text{ fg}$
$(ASE_0 + Si_{60}) \times M_{50}$	$64.2 \pm 1.70i$	$15,340 \pm 1245 \text{ lm}$	$16.6 \pm 0.54 \text{ k}$	$1.5\pm0.05\mathrm{jk}$
$(ASE_0 + Si_{60}) \times M_{75}$	$89.5 \pm 2.47ef$	$24,402 \pm 1395$ hi	21.9 ± 0.63 hij	$2.6 \pm 0.15 \text{ h}$
$(ASE_0 + Si_{60}) \times M_{100}$	$97.2 \pm 2.56d$	$29,432 \pm 954 \text{ g}$	22.6 ± 1.11 hij	$3.1 \pm 0.04 \text{ fg}$
$(ASE_{1.25} + Si_{60}) \times M_{50}$	$73.2 \pm 1.18 \text{ h}$	$19,070 \pm 640$ jkl	$22.9 \pm 1.11 \text{hi}$	$1.8\pm0.06ij$
$(ASE_{1.25} + Si_{60}) \times M_{75}$	94.2 ± 1.49 de	$32,907 \pm 1459 \text{ fg}$	$28.2 \pm 0.87 \text{ g}$	$3.4 \pm 0.15 \mathrm{ef}$
$(ASE_{1.25} + Si_{60}) \times M_{100}$	105.2 ± 2.50 bc	$35,851 \pm 1248ef$	$31.0 \pm 0.44 f$	$4.4\pm0.08c$
$(ASE_{2.5} + Si_{60}) \times M_{50}$	78.7 ± 1.11 gh	$19,567 \pm 916$ jk	$20.8 \pm 0.77 ij$	$2.6 \pm 0.12 \text{ h}$
$(ASE_{2.5} + Si_{60}) \times M_{75}$	100.0 ± 4.77 b	$35,449 \pm 1041$ f	$31.7 \pm 0.87ef$	$3.7 \pm 0.16 \mathrm{de}$
$(ASE_{2.5} + Si_{60}) \times M_{100}$	112.2 ± 3.19 cd	$39,802 \pm 1232$ de	33.8 ± 1.11 de	$4.7 \pm 0.14 \mathrm{c}$
$(ASE_{3.75} + Si_{60}) \times M_{50}$	$83.5 \pm 3.23 \text{ fg}$	$20,356 \pm 3380$ ijk	$22.9 \pm 0.92 hi$	2.7 ± 0.13 gh
$(ASE_{3.75} + Si_{60}) \times M_{75}$	$109.7 \pm 4.82b$	$43,295 \pm 580d$	$34.3 \pm 1.12 \text{ cd}$	$3.9 \pm 0.16d$
$(ASE_{3.75} + Si_{60}) \times M_{100}$	112.2 ± 2.66 b	$47,910 \pm 2618c$	36.3 ± 0.70 bc	5.3 ± 0.36 b
$(ASE_5 + Si_{60}) \times M_{50}$	$84.7 \pm 2.29 \text{ fg}$	$22,088 \pm 1819$ hij	$24.2 \pm 1.32 \text{ h}$	$2.8\pm0.06\mathrm{gh}$
$(ASE_5 + Si_{60}) \times M_{75}$	$131.0 \pm 2.58a$	$59,418 \pm 1631b$	37.3 ± 0.90 b	$5.3 \pm 0.12ab$
$(ASE_5 + Si_{60}) \times M_{100}$	$135.2 \pm 2.25a$	$63,780 \pm 995a$	$40.6 \pm 1.07a$	$5.7 \pm 0.15a$

Within each parameter means within a column followed by the same letters are not significantly different by least significant difference test at $P \le 0.05$; data are means of four replications \pm standard errors

(plant height, leaf area, shoot dry matter, and root dry matter) regardless of ASE doses.

3.2 Fruit Yield Parameters, Total Water Input, and Water Productivity

The interactive effect of ASE applied in combination with Si and soil moisture regime was significant for fruit yield, number of fruits per plant, and water productivity, whereas the main effects were significant for total water input (Table 1). A progressive increase in fruit yield was evident with increasing ASE dose regardless of soil moisture regimes (Table 3). A sixfold increase in fruit yield of plants supplemented with 5 mL L⁻¹ ASE in combination with 60 kg ha⁻¹ Si was evident compared with the control plants at 50% FC. The corresponding increase at 75% and

100% FC was 238% and 134%, respectively. Individual Si supplementation with 60 kg ha⁻¹ was also effective and caused an increase of as high as 207% in fruit yield compared with the control plants at severe water stress of 50% FC. Fruit yield was reduced by 43-80% when soil moisture level was reduced from 100 to 50% FC across ASE doses. Interestingly, the interaction effect indicated that there was no significant difference in fruit yield between moderate water stress of 75% FC with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si and well-watered condition of 100% FC with 3.75 mL L⁻¹ ASE and 60 kg ha⁻¹ Si, indicating beneficial effect of ASE and Si in alleviating the negative effects of water stress on plants and, therefore, increasing water productivity. Number of fruits per plant followed a nearly identical trend to that of fruit yield (Table 3). It was increased by 125%, 158%, and 107% in plants supplemented with 5 mL



Table 3 Effects of *Ascophyllum nodosum* seaweed extract (ASE) (0, 1.25, 2.5, 3.75, and 5 mL L⁻¹) applied in combination with silicon (Si) (60 kg Si ha⁻¹) and soil moisture regime on fruit yield parameters, total water input, and water productivity of tomato

Factor	Fruit yield (g plant ⁻¹)	Number of fruits per plant	Total water input (m ³ plant ⁻¹)	Water produc- tivity (kg m ⁻³)
$\overline{ASE (mL L^{-1}) + Si (kg ha^{-1})}$				
$ASE_0 + Si_0$	$251.4 \pm 41.84f$	12.7 ± 1.13 f	0.043 ± 0.001 d	$5.4 \pm 0.66 f$
$ASE_0 + Si_{60}$	$390.4 \pm 42.99e$	$14.6 \pm 0.89e$	$0.049 \pm 0.003c$	$7.7 \pm 0.51e$
$ASE_{1.25} + Si_{60}$	$485.3 \pm 45.40d$	16.5 ± 0.94 d	0.051 ± 0.004 bc	$9.3 \pm 0.50d$
$ASE_{2.5} + Si_{60}$	$554.8 \pm 46.28c$	$19.1 \pm 0.86c$	0.052 ± 0.003 bc	10.5 ± 0.47 c
$ASE_{3.75} + Si_{60}$	$621.9 \pm 42.68b$	$22.0 \pm 1.18b$	0.054 ± 0.004 ab	11.5 ± 0.35 b
$ASE_5 + Si_{60}$	$775.5 \pm 63.40a$	$28.9 \pm 2.47a$	$0.056 \pm 0.005a$	$13.8 \pm 0.58a$
Soil moisture regime (M)				
M ₅₀ -50% field capacity	$329.0 \pm 28.91c$	$13.9 \pm 0.78c$	$0.042 \pm 0.001c$	$7.9 \pm 0.60c$
M ₇₅ -75% field capacity	$514.3 \pm 41.23b$	20.5 ± 1.49 b	0.053 ± 0.001 b	9.8 ± 0.68 b
M ₁₀₀ -100% field capacity	$696.4 \pm 37.65a$	$22.5 \pm 1.37a$	$0.062 \pm 0.001a$	$11.4 \pm 0.48a$
$(ASE+Si)\times M$				
$(ASE_0 + Si_0) \times M_{50}$	$84.7 \pm 3.86 \text{ m}$	8.0 ± 0.41 j	0.032 ± 0.001	$2.7 \pm 0.16 \text{ k}$
$(ASE_0 + Si_0) \times M_{75}$	246.0 ± 7.141	$13.0 \pm 0.40 \text{hi}$	0.047 ± 0.002	5.4 ± 0.13 j
$(ASE_0 + Si_0) \times M_{100}$	423.5 ± 6.76 hi	17.0 ± 0.43 efg	0.054 ± 0.002	$8.1 \pm 0.21 \text{ h}$
$(ASE_0 + Si_{60}) \times M_{50}$	259.7 ± 13.17 kl	$11.5 \pm 1.19i$	0.039 ± 0.001	6.5 ± 0.35 ij
$(ASE_0 + Si_{60}) \times M_{75}$	$327.0 \pm 21.13j$	14.7 ± 0.75 gh	0.050 ± 0.002	6.5 ± 0.43 ij
$(ASE_0 + Si_{60}) \times M_{100}$	$584.4 \pm 11.67 f$	$17.5 \pm 0.87 \mathrm{efg}$	0.059 ± 0.002	$9.9 \pm 0.25 efg$
$(ASE_{1.25} + Si_{60}) \times M_{50}$	315.3 ± 25.87 jk	$13.0 \pm 0.82 \text{hi}$	0.041 ± 0.001	$7.7 \pm 0.74 \text{hi}$
$(ASE_{1.25} + Si_{60}) \times M_{75}$	474.6 ± 31.91 gh	$17.0 \pm 1.08 efg$	0.051 ± 0.002	$9.3 \pm 0.69 \text{ fg}$
$(ASE_{1.25} + Si_{60}) \times M_{100}$	666.1 ± 20.27 de	19.5 ± 0.87 cde	0.062 ± 0.003	10.8 ± 0.34 cde
$(ASE_{2.5} + Si_{60}) \times M_{50}$	$363.4 \pm 18.22ij$	$15.7 \pm 0.75 \text{fgh}$	0.041 ± 0.001	8.7 ± 0.54 gh
$(ASE_{2.5} + Si_{60}) \times M_{75}$	$574.6 \pm 19.72 f$	$20.5 \pm 1.19 \text{ cd}$	0.053 ± 0.002	10.9 ± 0.44 cde
$(ASE_{2.5} + Si_{60}) \times M_{100}$	726.4 ± 26.06 cd	$21.0 \pm 0.71c$	0.061 ± 0.003	11.9 ± 0.41 bc
$(ASE_{3.75} + Si_{60}) \times M_{50}$	446.3 ± 12.88 gh	17.2 ± 0.63 efg	0.043 ± 0.001	$10.3 \pm 0.39 \text{def}$
$(ASE_{3.75} + Si_{60}) \times M_{75}$	633.4 ± 22.84 ef	24.0 ± 1.08 b	0.054 ± 0.002	11.6 ± 0.37 bc
$(ASE_{3.75} + Si_{60}) \times M_{100}$	786.0 ± 7.58 bc	24.7 ± 1.55 b	0.063 ± 0.003	12.5 ± 0.46 b
$(ASE_5 + Si_{60}) \times M_{50}$	$504.4 \pm 17.54 \text{ g}$	$18.0 \pm 0.71 \text{def}$	0.045 ± 0.001	11.3 ± 0.37 cd
$(ASE_5 + Si_{60}) \times M_{75}$	830.3 ± 18.03 b	$33.5 \pm 1.71a$	0.056 ± 0.003	$14.8 \pm 0.64a$
$(ASE_5 + Si_{60}) \times M_{100}$	$991.8 \pm 49.92a$	$35.2 \pm 1.89a$	0.065 ± 0.003	$15.2 \pm 0.24a$

Within each parameter means within a column followed by the same letters are not significantly different by least significant difference test at $P \le 0.05$; data are means of four replications \pm standard errors

L⁻¹ ASE and 60 kg ha⁻¹ Si compared with the control at 50%, 75%, and 100% FC, respectively. Only Si supplementation resulted in 44% increase in number of fruits per plant compared with the control at 50% FC. A clear benefit of ASE and Si supplementation was observed where the control plants produced lower number of fruits per plant at 100% FC compared with the plants grown with 5 mL L⁻¹ ASE in combination with 60 kg ha⁻¹ Si at 50% FC, which indicates the potential of ASE and Si as drought mitigating agents. Nevertheless, a decrease in soil moisture level was equally detrimental for all plants and caused a significant reduction in number of fruits per plant (25–53%) at 50% FC compared with that at 100% FC within an ASE dose. Total water input gradually increased with increasing ASE dose and increasing soil moisture regime as indicated by

the significant main effect of ASE and soil moisture regime (Table 3). The interaction between ASE and soil moisture regime indicates a gradual increase in water productivity with increasing ASE dose irrespective of soil moisture regimes (Table 3). The control plants had the lowest water productivity at all soil moisture regimes, which was 76%, 64%, and 47% lower than plants supplemented with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si dose at 50%, 75%, and 100% FC, respectively. Plants supplemented with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si had higher water productivity at 50% FC than water productivity of the control plants at 100% FC. Within an ASE and Si dose, water productivity exhibited an increase in the range of 21–200% at 100% FC compared with 50% FC across the treatment combination. Individual Si supplementation was also effective and caused 141%



increase in water productivity at 50% FC compared with that of the control plants.

3.3 Fruit Quality Parameters

The interactive effect of ASE applied in combination with Si and soil moisture regime was significant for fruit pH, whereas the main effects were significant for fruit length, fruit width, TSS content, fruit firmness, and color index (Table 1). A gradual improvement in fruit quality was observed with increasing ASE dose, while exactly the opposite was true for decreasing soil moisture regime as indicated by the significant main effect of ASE and soil

moisture regime (Table 4). Fruit length, fruit width, TSS content, fruit firmness, and color index were increased by 59%, 62%, 21%, 42%, and 73%, respectively, at 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si dose compared with the control. Decreasing soil moisture level from 100 to 50% FC caused 13% decrease in fruit length and fruit width, while TSS content, fruit firmness, and color index were increased by 35%, 14%, and 58%, respectively, for the same soil moisture regimes. Fruit pH was acidic regardless of ASE doses and soil moisture regimes; however, acidity was slightly decreased with increasing ASE dose at all soil moisture levels as indicated by a significant interactive effect between ASE dose and soil moisture regime for fruit pH (Table 4).

Table 4 Effects of *Ascophyllum nodosum* seaweed extract (ASE) $(0, 1.25, 2.5, 3.75, \text{ and } 5 \text{ mL L}^{-1})$ applied in combination with silicon (Si) $(60 \text{ kg Si ha}^{-1})$ and soil moisture regime on fruit quality parameters of tomato

Factor	Fruit length (cm)	Fruit width (cm)	Total soluble solids (°Brix)	Fruit firmness	Color index	Fruit pH
$\overline{\text{ASE (mL L}^{-1}) + \text{Si (kg ha}^{-1})}$						
$ASE_0 + Si_0$	$3.2 \pm 0.08e$	$2.6 \pm 0.09e$	$7.5 \pm 0.34e$	1.2 ± 0.03 d	45.3 ± 3.86 f	$4.10 \pm 0.02e$
$ASE_0 + Si_{60}$	$3.8 \pm 0.14d$	$3.2 \pm 0.10d$	$8.1 \pm 0.35d$	$1.3 \pm 0.05 \text{ cd}$	$51.5 \pm 3.82e$	$4.16 \pm 0.02d$
$ASE_{1.25} + Si_{60}$	$4.3 \pm 0.09c$	$3.6 \pm 0.11c$	8.3 ± 0.33 cd	$1.4 \pm 0.05c$	$58.1 \pm 3.70d$	$4.22 \pm 0.02c$
$ASE_{2.5} + Si_{60}$	$4.6 \pm 0.12b$	3.8 ± 0.10 bc	$8.6 \pm 0.30 \text{bc}$	1.6 ± 0.04 b	$66.3 \pm 3.83c$	4.27 ± 0.01 b
$ASE_{3.75} + Si_{60}$	$4.9 \pm 0.11ab$	4.0 ± 0.09 ab	8.8 ± 0.30 b	1.6 ± 0.05 ab	71.3 ± 3.74 b	$4.29 \pm 0.02b$
$ASE_5 + Si_{60}$	$5.1 \pm 0.11a$	$4.2 \pm 0.08a$	$9.1 \pm 0.31a$	$1.7 \pm 0.05a$	$78.5 \pm 4.11a$	$4.35 \pm 0.03a$
Soil moisture regime (M)						
M ₅₀ -50% field capacity	4.1 ± 0.14 b	$3.3 \pm 0.12c$	$9.7 \pm 0.14a$	$1.6 \pm 0.05a$	$78.2 \pm 2.70a$	$4.32 \pm 0.02a$
M ₇₅ -75% field capacity	4.2 ± 0.15 b	$3.6 \pm 0.13b$	8.4 ± 0.09 b	1.5 ± 0.04 b	57.9 ± 2.10 b	$4.26 \pm 0.02b$
M ₁₀₀ -100% field capacity	$4.7 \pm 0.14a$	$3.8 \pm 0.11a$	$7.2 \pm 0.15c$	1.4 ± 0.04 b	$49.4 \pm 2.74c$	$4.14 \pm 0.02c$
$(ASE + Si) \times M$						
$(ASE_0 + Si_0) \times M_{50}$	2.9 ± 0.04	2.2 ± 0.02	8.7 ± 0.15	1.4 ± 0.01	62.0 ± 0.71	$4.17 \pm 0.02ij$
$(ASE_0 + Si_0) \times M_{75}$	3.1 ± 0.06	2.5 ± 0.05	7.8 ± 0.05	1.2 ± 0.01	42.5 ± 1.00	$4.12 \pm 0.01 \text{ k}$
$(ASE_0 + Si_0) \times M_{100}$	3.5 ± 0.11	3.0 ± 0.03	6.0 ± 0.03	1.1 ± 0.01	31.3 ± 1.13	4.02 ± 0.011
$(ASE_0 + Si_{60}) \times M_{50}$	3.5 ± 0.16	2.9 ± 0.10	9.4 ± 0.19	1.5 ± 0.12	67.3 ± 2.48	$4.18 \pm 0.03 \text{hi}$
$(ASE_0 + Si_{60}) \times M_{75}$	3.6 ± 0.23	3.3 ± 0.05	8.1 ± 0.14	1.3 ± 0.05	48.6 ± 2.12	4.21 ± 0.02 fgh
$(ASE_0 + Si_{60}) \times M_{100}$	4.4 ± 0.13	3.4 ± 0.22	6.7 ± 0.12	1.2 ± 0.03	38.7 ± 3.10	$4.08 \pm 0.03 \text{ k}$
$(ASE_{1.25} + Si_{60}) \times M_{50}$	4.0 ± 0.14	3.3 ± 0.12	9.6 ± 0.19	1.6 ± 0.13	71.8 ± 3.34	$4.30 \pm 0.02c$
$(ASE_{1.25} + Si_{60}) \times M_{75}$	4.3 ± 0.04	3.7 ± 0.23	8.2 ± 0.18	1.4 ± 0.08	57.6 ± 1.80	$4.25 \pm 0.01 \text{def}$
$(ASE_{1.25} + Si_{60}) \times M_{100}$	4.6 ± 0.13	3.8 ± 0.17	7.0 ± 0.05	1.3 ± 0.02	45.0 ± 4.02	$4.12 \pm 0.01 jk$
$(ASE_{2.5} + Si_{60}) \times M_{50}$	4.4 ± 0.10	3.5 ± 0.09	9.8 ± 0.23	1.7 ± 0.08	82.5 ± 2.72	$4.36 \pm 0.01b$
$(ASE_{2.5} + Si_{60}) \times M_{75}$	4.6 ± 0.13	3.9 ± 0.23	8.4 ± 0.19	1.5 ± 0.04	62.8 ± 0.25	4.28 ± 0.01 cde
$(ASE_{2.5} + Si_{60}) \times M_{100}$	4.9 ± 0.28	4.1 ± 0.09	7.5 ± 0.14	1.5 ± 0.05	53.6 ± 2.81	4.17 ± 0.02 hij
$(ASE_{3.75} + Si_{60}) \times M_{50}$	4.7 ± 0.06	3.8 ± 0.05	10.0 ± 0.29	1.8 ± 0.08	88.3 ± 2.04	$4.39 \pm 0.01b$
$(ASE_{3.75} + Si_{60}) \times M_{75}$	4.8 ± 0.18	4.0 ± 0.24	8.6 ± 0.19	1.6 ± 0.02	64.4 ± 1.78	4.29 ± 0.01 cd
$(ASE_{3.75} + Si_{60}) \times M_{100}$	5.2 ± 0.24	4.2 ± 0.09	7.8 ± 0.11	1.5 ± 0.05	61.3 ± 0.82	4.20 ± 0.02 ghi
$(ASE_5 + Si_{60}) \times M_{50}$	4.8 ± 0.05	4.0 ± 0.09	10.4 ± 0.26	1.8 ± 0.12	97.3 ± 1.60	$4.48\pm0.02a$
$(ASE_5 + Si_{60}) \times M_{75}$	5.1 ± 0.20	4.2 ± 0.17	8.9 ± 0.19	1.8 ± 0.06	71.4 ± 0.46	$4.38 \pm 0.01b$
$(ASE_5 + Si_{60}) \times M_{100}$	5.4 ± 0.21	4.3 ± 0.09	8.1 ± 0.15	1.6 ± 0.09	66.7 ± 1.34	4.24 ± 0.02 efg

Within each parameter means within a column followed by the same letters are not significantly different by least significant difference test at $P \le 0.05$; data are means of four replications \pm standard errors



3.4 Physio-biochemical Parameters

The interactive effect of ASE applied in combination with Si and soil moisture regime was significant for leaf greenness (SPAD value), LRWC, electrolyte leakage, membrane stability index, P_n, g_s, E, and free proline content (Table 1). The 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si dose had an overall better performance in terms of all physio-biochemical parameters across soil moisture regimes, while decreasing soil moisture level was equally detrimental for all evaluated parameters (Table 5). Leaf greenness of plants supplemented with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si dose was 58% higher than the control plants at 50% FC. A progressive increase in LRWC was observed with increasing ASE dose irrespective of soil moisture regimes, which was maximized at 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si with 53%, 55%, and 40% increase

compared with that of the control at 50%, 75%, and 100% FC, respectively. Decreasing soil moisture level from 100 to 50% FC caused 25-39% decrease in LRWC across treatment combinations. Electrolyte leakage of the control plants was significantly higher at all soil moisture regimes and was gradually decreased with increasing ASE dose. It was significantly increased with decreasing soil moisture regime. The control plants had 64%, 81%, and 49% higher electrolyte leakage compared with plants supplemented with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si at 50%, 75%, and 100% FC, respectively. A decrease of 17-25% in electrolyte leakage was recorded at well-watered condition of 100% FC compared with that at severe water stress of 50% FC across treatment combinations. Membrane stability index followed exactly the opposite trend to that of electrolyte leakage and it was 144% higher for plants supplemented with 5 mL L⁻¹ ASE

Table 5 Effects of *Ascophyllum nodosum* seaweed extract (ASE) (0, 1.25, 2.5, 3.75, and 5 mL L⁻¹) applied in combination with silicon (Si) (60 kg Si ha⁻¹) and soil moisture regime on physiological parameters of tomato

Factor	Leaf greenness (SPAD value)	Leaf relative water content (%)	Electrolyte leakage (%)	Membrane stability index (%)
$\overline{ASE (mL L^{-1}) + Si (kg ha^{-1})}$				
$ASE_0 + Si_0$	$45.1 \pm 2.01e$	$56.0 \pm 3.36e$	$40.4 \pm 1.46a$	$20.4 \pm 1.98 f$
$ASE_0 + Si_{60}$	$50.6 \pm 2.39d$	61.6 ± 3.09 d	36.0 ± 1.26 b	$25.2 \pm 2.43e$
$ASE_{1.25} + Si_{60}$	$58.0 \pm 1.47c$	$66.5 \pm 3.41c$	$32.7 \pm 1.41c$	$28.3 \pm 1.62d$
$ASE_{2.5} + Si_{60}$	59.5 ± 1.49 bc	$67.5 \pm 2.43c$	$30.0 \pm 1.02d$	$30.0 \pm 1.97c$
$ASE_{3.75} + Si_{60}$	61.1 ± 1.71 b	$71.1 \pm 2.88b$	$27.9 \pm 0.72e$	32.6 ± 1.75 b
$ASE_5 + Si_{60}$	$64.6 \pm 1.94a$	$83.2 \pm 4.45a$	24.6 ± 0.76 f	$35.7 \pm 1.59a$
Soil moisture regime (M)				
M ₅₀ -50% field capacity	$48.9 \pm 1.68c$	$54.1 \pm 1.56c$	$35.7 \pm 1.29a$	20.2 ± 1.24 c
M ₇₅ -75% field capacity	59.3 ± 1.41 b	69.6 ± 2.25 b	$32.4 \pm 1.28b$	31.4 ± 1.09 b
M ₁₀₀ -100% field capacity	$61.3 \pm 1.41a$	$79.3 \pm 1.92a$	$27.7 \pm 0.83c$	$34.4 \pm 0.92a$
$(ASE + Si) \times M$				
$(ASE_0 + Si_0) \times M_{50}$	$36.1 \pm 0.30i$	41.4 ± 0.52 n	$45.3 \pm 0.57a$	11.7 ± 0.461
$(ASE_0 + Si_0) \times M_{75}$	47.3 ± 0.41 g	58.4 ± 0.71 jk	41.8 ± 0.86 b	22.4 ± 0.67 j
$(ASE_0 + Si_0) \times M_{100}$	51.8 ± 0.61 f	$68.1 \pm 1.12 \text{ fg}$	$34.1 \pm 0.56d$	$27.2 \pm 0.58 \text{ h}$
$(ASE_0 + Si_{60}) \times M_{50}$	$40.2 \pm 0.56 \text{ h}$	$49.3 \pm 2.23 \text{ m}$	40.3 ± 0.95 b	$14.1 \pm 0.87 \text{ k}$
$(ASE_0 + Si_{60}) \times M_{75}$	$53.6 \pm 1.34ef$	$62.0 \pm 1.02 \text{hij}$	$36.6 \pm 1.05c$	29.2 ± 0.93 gh
$(ASE_0 + Si_{60}) \times M_{100}$	$58.1 \pm 1.66d$	73.5 ± 1.33 de	$30.9 \pm 0.64 \text{ fg}$	32.2 ± 0.93 ef
$(ASE_{1.25} + Si_{60}) \times M_{50}$	$52.9 \pm 1.02 f$	54.0 ± 1.061	$37.8 \pm 0.67c$	21.0 ± 0.69 j
$(ASE_{1.25} + Si_{60}) \times M_{75}$	62.1 ± 1.07 bc	64.6 ± 1.10 gh	$32.9 \pm 1.36 def$	$30.5 \pm 0.74 \text{ fg}$
$(ASE_{1.25} + Si_{60}) \times M_{100}$	$59.0 \pm 2.64 \text{ cd}$	$80.8 \pm 1.98c$	27.5 ± 1.37 ij	33.3 ± 0.32 de
$(ASE_{2.5} + Si_{60}) \times M_{50}$	$53.1 \pm 1.36f$	57.0 ± 0.79 kl	33.2 ± 0.57 de	21.1 ± 1.14 j
$(ASE_{2.5} + Si_{60}) \times M_{75}$	62.6 ± 0.77 bc	$70.0 \pm 0.80 ef$	31.4 ± 0.51 efg	33.1 ± 0.81 de
$(ASE_{2.5} + Si_{60}) \times M_{100}$	62.8 ± 1.36 b	$75.5 \pm 1.84d$	25.5 ± 0.42 jk	35.7 ± 0.73 bc
$(ASE_{3.75} + Si_{60}) \times M_{50}$	53.7 ± 1.75 ef	59.5 ± 1.07 ijk	30.1 ± 0.86 gh	$24.9 \pm 0.59i$
$(ASE_{3.75} + Si_{60}) \times M_{75}$	64.7 ± 0.57 b	$71.5 \pm 1.31 \text{def}$	$28.6 \pm 0.47 \text{hi}$	34.8 ± 0.57 cd
$(ASE_{3.75} + Si_{60}) \times M_{100}$	64.8 ± 1.24 b	$82.3 \pm 1.10c$	25.1 ± 0.59 kl	37.9 ± 1.56 ab
$(ASE_5 + Si_{60}) \times M_{50}$	57.1 ± 1.63 de	$63.5 \pm 1.14 \text{hi}$	27.7 ± 0.70 ij	28.5 ± 0.90 gh
$(ASE_5 + Si_{60}) \times M_{75}$	65.3 ± 1.09 b	$90.6 \pm 3.13b$	23.1 ± 0.791	$38.6 \pm 0.70a$
$(ASE_5 + Si_{60}) \times M_{100}$	$71.5 \pm 1.69a$	$95.6 \pm 2.90a$	22.9 ± 0.521	$40.0 \pm 0.24a$

Within each parameter means within a column followed by the same letters are not significantly different by least significant difference test at $P \le 0.05$; data are means of four replications \pm standard errors



and 60 kg ha⁻¹ Si dose compared with that of the control at 50% FC. Membrane stability index exhibited a significant reduction at lower soil moisture regime even with the highest ASE dose; however, there was no difference in membrane stability index between 100 and 75% FC at this ASE dose. Plants supplemented with only Si without any ASE also had higher (21%) membrane stability index even at 50% FC compared with the control.

There was a steady increase in P_n , g_s , and E with increasing ASE dose, which were maximized at 5 mL L^{-1} ASE and 60 kg ha⁻¹ Si dose (Table 6). An increase of 185%, 267%, and 158% in P_n , g_s , and E were evident at 5 mL L^{-1} ASE and 60 kg ha⁻¹ Si dose compared with 0 mL L^{-1} ASE and 0 kg ha⁻¹ Si at 50% FC. Only Si supplementation resulted in

no significant increase in these parameters at 50% FC, nevertheless the same parameters were significantly increased at 75% FC. Decreasing soil moisture level from 100 to 50% FC caused 35--47%, 50--67%, and 51--74% decrease in P_n , g_s , and E, respectively.

Free proline content was generally higher in plants subjected to lower soil moisture level regardless of ASE and Si treatments (Table 6). However, it was the highest in plants supplemented with 5 mL L⁻¹ ASE and 60 kg ha⁻¹ Si dose at 50% FC, which was 46% higher than the control at the corresponding soil moisture regime. An increase in the range of 32–53% in free proline content at 50% FC was evident compared with that at 100% FC across treatment combinations. Only Si supplementation caused 6%, 13%, and 13%

Table 6 Effects of *Ascophyllum nodosum* seaweed extract (ASE) (0, 1.25, 2.5, 3.75, and 5 mL L⁻¹) applied in combination with silicon (Si) (60 kg Si ha⁻¹) and soil moisture regime on leaf gas exchange parameters and free proline content of tomato

Factor	Net photosynthetic rate (μmol CO ₂ m ⁻² s ⁻¹)	Stomatal conductance (mmol H ₂ O m ⁻² s ⁻¹)	Transpiration rate (mmol $H_2O m^{-2} s^{-1}$)	Free proline content (mg g ⁻¹ fresh weight)
$\overline{ASE (mL L^{-1}) + Si (kg ha^{-1})}$				
$ASE_0 + Si_0$	$5.0 \pm 0.37 f$	0.05 ± 0.01 f	$1.20 \pm 0.19e$	$0.057 \pm 0.001e$
$ASE_0 + Si_{60}$	$6.0 \pm 0.38e$	$0.08 \pm 0.02e$	$1.43 \pm 0.22d$	$0.064 \pm 0.002d$
$ASE_{1.25} + Si_{60}$	8.8 ± 0.57 d	$0.12 \pm 0.02d$	$1.76 \pm 0.21c$	$0.075 \pm 0.002c$
$ASE_{2.5} + Si_{60}$	$10.1 \pm 0.59c$	$0.13 \pm 0.01c$	1.79 ± 0.16 bc	0.080 ± 0.003 b
$ASE_{3.75} + Si_{60}$	12.1 ± 0.69 b	$0.15 \pm 0.01b$	1.97 ± 0.20 b	$0.084 \pm 0.003a$
$ASE_5 + Si_{60}$	$14.5 \pm 1.15a$	$0.18 \pm 0.02a$	$2.72 \pm 0.31a$	$0.086 \pm 0.004a$
Soil moisture regime (M)				
M ₅₀ -50% field capacity	$6.8 \pm 0.49c$	$0.07 \pm 0.01c$	$0.96 \pm 0.07c$	$0.091 \pm 0.001a$
M ₇₅ -75% field capacity	10.0 ± 0.79 b	$0.13 \pm 0.01b$	$1.88 \pm 0.14b$	$0.071 \pm 0.001b$
M ₁₀₀ -100% field capacity	$11.3 \pm 0.86a$	$0.16 \pm 0.01a$	$2.59 \pm 0.12a$	$0.063 \pm 0.001c$
$(ASE + Si) \times M$				
$(ASE_0 + Si_0) \times M_{50}$	$3.3 \pm 0.15 \text{ k}$	$0.03 \pm 0.01i$	0.52 ± 0.01 j	0.072 ± 0.002 f
$(ASE_0 + Si_0) \times M_{75}$	5.3 ± 0.21 ij	0.06 ± 0.01 gh	1.06 ± 0.01 gh	$0.054 \pm 0.001 \text{ h}$
$(ASE_0 + Si_0) \times M_{100}$	$6.2 \pm 0.12 hi$	$0.08 \pm 0.02e$	2.01 ± 0.01 ef	$0.047 \pm 0.002i$
$(ASE_0 + Si_{60}) \times M_{50}$	4.3 ± 0.26 jk	$0.04 \pm 0.02 hi$	$0.61 \pm 0.03ij$	$0.076 \pm 0.002e$
$(ASE_0 + Si_{60}) \times M_{75}$	6.7 ± 0.28 gh	0.08 ± 0.01 ef	1.36 ± 0.03 g	0.061 ± 0.001 g
$(ASE_0 + Si_{60}) \times M_{100}$	6.9 ± 0.27 gh	$0.12 \pm 0.02d$	2.31 ± 0.17 de	$0.053 \pm 0.002 \text{ h}$
$(ASE_{1.25} + Si_{60}) \times M_{50}$	6.5 ± 0.13 hi	$0.07 \pm 0.01 \text{ fg}$	$0.94 \pm 0.04 hi$	$0.086 \pm 0.002c$
$(ASE_{1.25} + Si_{60}) \times M_{75}$	9.0 ± 0.32 ef	$0.12 \pm 0.01d$	$1.83 \pm 0.18f$	0.074 ± 0.001 ef
$(ASE_{1.25} + Si_{60}) \times M_{100}$	10.7 ± 0.68 d	$0.15 \pm 0.01c$	2.50 ± 0.16 cd	0.065 ± 0.001 g
$(ASE_{2.5} + Si_{60}) \times M_{50}$	$7.9 \pm 0.16 \text{ fg}$	0.08 ± 0.02 ef	1.15 ± 0.03 gh	$0.098 \pm 0.003b$
$(ASE_{2.5} + Si_{60}) \times M_{75}$	10.1 ± 0.46 de	$0.15 \pm 0.01c$	$1.86 \pm 0.04 f$	0.074 ± 0.001 ef
$(ASE_{2.5} + Si_{60}) \times M_{100}$	$12.2 \pm 0.72c$	$0.16 \pm 0.01c$	2.36 ± 0.19 cde	0.070 ± 0.001 f
$(ASE_{3.75} + Si_{60}) \times M_{50}$	$9.2 \pm 0.24e$	$0.09 \pm 0.02e$	1.21 ± 0.02 gh	$0.104 \pm 0.003a$
$(ASE_{3.75} + Si_{60}) \times M_{75}$	12.7 ± 0.69 c	$0.16 \pm 0.01c$	2.01 ± 0.15 ef	$0.076 \pm 0.001e$
$(ASE_{3.75} + Si_{60}) \times M_{100}$	14.3 ± 0.41 b	$0.19 \pm 0.02b$	$2.69 \pm 0.20c$	0.070 ± 0.001 f
$(ASE_5 + Si_{60}) \times M_{50}$	$9.4 \pm 0.29e$	$0.11 \pm 0.01d$	1.34 ± 0.06 g	0.105 ± 0.003 a
$(ASE_5 + Si_{60}) \times M_{75}$	$16.4 \pm 0.40a$	0.20 ± 0.01 ab	$3.15 \pm 0.21b$	$0.081 \pm 0.002d$
$(ASE_5 + Si_{60}) \times M_{100}$	$17.6 \pm 1.08a$	$0.22 \pm 0.01a$	$3.66 \pm 0.20a$	0.071 ± 0.002 f

Within each parameter means within a column followed by the same letters are not significantly different by least significant difference test at $P \le 0.05$; data are means of four replications \pm standard errors



increase in free proline content compared with that of the control at 50%, 75%, and 100% FC, respectively.

4 Discussion

Different biotic and abiotic stress factors have a substantial influence on agricultural production and food security, and drought is a key abiotic stress that has a detrimental impact on crop growth and productivity at a global scale [9, 61]. It causes severe morphological, biochemical, and physiological damages to plants, resulting in significantly lower crop yields [8, 10, 39, 62]. Plant growth and fruit yield parameters of tomato were negatively impacted by soil moisture shortage in the present study. Cell division and cell elongation mechanisms are disrupted as a direct result of drought stress [8, 13, 63], which might be the reason for lower plant height and leaf area observed under water stress during the present study. Limiting soil moisture level from well-watered (100% FC) condition to severe water stress (50% FC) has been reported to reduce growth parameters, grain yield, and yield components of maize (Zea mays L.), which has been attributed to reduced cell division and cell elongation processes [10]. In the present study, growth and fruit yield characteristics were drastically reduced when tomato plants were grown under severe drought stress at 50% FC. These results are in close agreement with Kuscu et al. [64] who found that vegetables, especially tomatoes, are extremely susceptible to water stress. Nangare et al. [65] observed a substantial decrease in tomato growth parameters (plant height and leaf area index), root parameters (weight and depth), and chlorophyll content grown under controlled deficit irrigation level $(0.6 \times ET_c)$. Lower fruit yield at 50% FC might be attributed to a reduction in overall growth parameters, number of fruits per plant, fruit length, and fruit width caused by a poor physiological response since water stress decreases SPAD value, LRWC, and membrane stability index but increases electrolyte leakage [66]. A decrease in soil moisture regime improved tomato fruit quality parameters (TSS content, fruit firmness, and color index), which is consistent with Kuscu et al. [64], Nangare et al. [65], Zegbe-Dominguez et al. [67], and Helyes et al. [68]. Water stress inhibits the movement of water inside the plant but does not impair the dispersion of photoassimilates [69]. Water stress causes a reduction in fruit size and low dilution, which encourages the accumulation of assimilates inside fruits, resulting in an increase in fruit quality parameters [64, 67, 68]. Water stress also induces increased conversion of starch to sugars, resulting in an increase in TSS content [67]. Water productivity and LRWC declined with decreasing soil moisture regime, owing to poorer fruit production and vegetative development, respectively, whereas electrolyte leakage increased. Similarly, Hayat et al. [66] also observed an increase in electrolyte leakage and a decrease in LRWC in tomato when soil moisture regime was reduced.

There has been an increased focus on the use of various exogenous protectants in mitigating the negative effects of drought on various crops. An aqueous alkaline extract of A. nodosum applied to the soil has been reported to enhance chlorophyll contents in the leaves of the treated plants [tomato, dwarf French bean (*Phaseolus vulgaris* L.), wheat (Triticum aestivum L.), barley (Hordeum vulgare L.), and maize] compared with the untreated control plants [25, 30]. In the present study, exogenous combined application of ASE with Si enhanced growth and physiological traits of tomato. The increase in SPAD value and subsequently that of P_n and g_s might be attributed to the presence of betaines in the seaweed extract raising plant's chlorophyll levels [31]. Application of ASE can enhance endogenous phytohormones including cytokinin, auxin, indole acetic acid, and gibberellic acid, which can help plant growth and development as well as protect it from environmental challenges, such as drought, salt stress, and temperature extremes [25, 70–72]. Many studies show that the "stay-green" feature (increased chlorophyll content) relates to greater transpiration efficiency and productivity in sorghum [Sorghum bicolor (L.) Moench] and wheat under limited water availability [73, 74]. Increased plant's chlorophyll levels due to the application of ASE following water stress might be beneficial for reestablishing photosynthetic capacity of the leaves, leading to growth recovery. Kumari et al. [75] observed that seaweed extract significantly increased tomato growth regardless of application techniques. The application of Si, another exogenous protectant, has been found effective in alleviating the detrimental effects of drought stress in various agronomic and horticultural crops [8, 10, 11, 39, 40]. The beneficial effects of Si supplementation in enhancing plant tolerance against various biological and environmental challenges have been well established.

The synergetic effect between ASE and Si under different soil moisture regimes revealed an overall trend of growth and yield enhancement as indicated by an improvement in growth parameters, physio-biochemical traits, and fruit yield parameters, while only Si supplementation was also effective compared with the control plants and helped in growth and productivity improvement. Gowda et al. [76] applied a specific Si fertilizer (OSV-5 containing 8% Si) at 750 kg ha⁻¹ (60 kg ha⁻¹ soluble Si) in combination with the recommended NPK fertilizer dose and found longer tomato plants with more branches and fruit yield than plants grown without any exogenous Si fertilization. A Si-induced increase in root dry matter was observed at moderate water stress of 75% FC in comparison to the control plants, indicating that Si plays a positive role in enhancing plant development even without ASE supplementation. Improved root system development is critical under water stress, allowing plants



to maintain plant water relations. Ullah et al. [77] reported that the addition of 50 ppm Si to tomato plants increased shoot dry matter and root dry matter by about 36% and 45%, respectively. Most of the Si-mediated beneficial effects have been attributed to higher root hydraulic conductivity and cell wall integrity. Higher root dry matter and root hydraulic conductivity might confer drought tolerance to tomato plants because of Si supplementation. Other possible Si-induced stress-reduction processes include reduced transpirational water loss, increased root water uptake and metabolism, and enhanced antioxidant defense system [78]. A possible drought stress response has been proposed to include Si deposition in cell walls of the leaf epidermis, thereby decreasing E [79]. However, this is not always the case, as no change in E has been reported following Si application in sorghum [34] and wheat [33]. The mechanism of improved drought tolerance induced by Si supplementation might be more closely linked to an enhancement of water uptake ability. Silicon-mediated increase in yields under drought stress has been widely demonstrated in various crops, such as rice [8], maize [10], wheat [80], potato (Solanum tuberosum L.) [81], cantaloupe (Cucumis melo L.) [39], and grape tomato (Solanum lycopersicum L. var. cerasiforme) [40].

Increased leaf area and LRWC induced by the application of ASE under drought stress reveals that ASE might enhance tomato leaf water relations and support in the maintenance of cell turgor pressure and expansion, resulting in a large leaf area. Similarly, Spann and Little [28] reported the maintenance of orange plant growth and development with the application of ASE during drought stress, which was credited to enhanced plant water relations. Neily et al. [82] reported a significant improvement in leaf water content and improved recovery of wilted plants when a commercial extract from A. nodosum was applied along with fertilizer in different vegetables, including tomato. Increased leaf area has a direct positive impact on P_n, which in turn promotes growth and overall development of plants. Kumari et al. [75] reported that seaweed extract enhanced the content of photosynthetic pigments (chlorophyll and carotenoids) in tomato leaves regardless of application methods. Seaweed extract has been also reported to be beneficial in boosting chlorophyll biosynthesis in maize and black gram (Phaseolus mungo L.) [83] and common sage (Salvia officinalis L.) [84]. In the present study, ASE at 5 mL L⁻¹ with 60 kg ha⁻¹ Si improved leaf water relations and helped in maintaining cell turgor pressure by decreasing stomatal closure, resulting in a wide leaf area and high P_n, and, therefore, increased growth. Seaweed extract possesses trace elements that can be assimilated into plants. A. nodosum seaweed extract contains polyuronides (alginates and fucoidans), which improve soil water retention capacity, crumb structure, aeration, and capillary action, all of which can stimulate plant root system, increase soil microbial activity, and improve mineral availability and uptake [25].

Application of ASE improves plant tolerance against abiotic stress by providing (i) faster reestablishment of osmotic adjustment, (ii) increased photosynthetic activity, (iii) and enhanced g_s [25, 85]. Karabudak et al. [86] and Murtic et al. [87] mentioned that chemicals present in ASE, such as glycine betaine and sterols, operate as a buffer against significant osmotic changes in plant cells, thereby mitigating the harmful effects of stress on plants. Additionally, it also contains a variety of osmolytes, such as proline, valine, isoleucine, and aspartic acid, vitamins, and microelements, and various other active natural chemicals that enhance crops stress tolerance [28].

Proline accumulation has been linked to stress tolerance in a variety of plant species, and its content has been found to be usually higher in stress-tolerant plants than in stresssensitive ones. However, proline accumulation cannot be solely used as a specific marker for drought tolerance since it is a generic response of plants to diverse abiotic stresses [16]. A gradual increase in free proline content was observed in plants treated with ASE, which was maximized at 5 mL L⁻¹ compared with the untreated control plants regardless of soil moisture regimes. High proline accumulation facilitates plants to sustain under stresses that cause dehydration of the plant tissue, such as drought (low water potential), allowing them for extra water uptake from the environment and, therefore, buffering the immediate effect of water scarcity [88]. It has been reported that tomato genotypes that are more susceptible to soil water deficit respond to drought stress by accumulating less proline in the leaves [89]. Therefore, a significant proline buildup cannot be regarded as a typical reaction to seaweed extract in drought-stressed crops, and the degree of its accumulation may vary according to the type of seaweed used, application method, and crop class.

The application of ASE and Si influenced fruit quality parameters. Fruit quality (length, width, firmness, and color index) exhibited a gradual increase with increasing ASE dose in combination with Si. In addition, plants supplemented only with Si also had better fruit quality in comparison to the control plants, indicating that both ASE and Si are effective in improving tomato fruit quality. This improvement in fruit quality is advantageous since the optimum market price for tomato is mostly determined by the size of the fruit. Sugar level strongly correlates with TSS content in fruit crops, and sugar level markedly controls fruit quality [90]. In this study, there was a strong tendency toward increased TSS content with decreasing soil moisture level, and the application of ASE and Si substantially increased fruit TSS content. The overall performance of the plant was improved when 5 mL L⁻¹ ASE was applied in combination with 60 kg ha⁻¹ Si.



5 Conclusion

Abiotic stress, such as drought, is a severe environmental constraint that has a negative impact on growth and development of almost all crops including tomato. The results indicate that tomato is susceptible to water stress, with the overall performance being negatively impacted at a severe soil moisture level of 50% FC. However, at moderate soil moisture level of 75% FC and well-watered conditions of 100% FC, an exogenous application of ASE at 5 mL L⁻¹ in combination with 60 kg ha⁻¹ Si was beneficial. The advantage of ASE and Si application on fruit yield was demonstrated by similar yields between plants grown under 100% FC with ASE and Si doses of 3.75 mL L⁻¹ and 60 kg ha⁻¹, respectively, and plants grown under 75% FC with ASE and Si doses of 5 mL L⁻¹ and 60 kg ha⁻¹, respectively, resulting in increased water productivity by deficit irrigation. In addition, fruit quality was significantly improved (increased fruit length, fruit width, TSS content, fruit firmness, and color index) in response to ASE application and the best quality fruits were obtained at 5 mL L⁻¹ ASE dose. Plants grown with only Si without any ASE supplementation also had an overall better performance compared with the control plants; however, the effects were lower compared with the integrated application of ASE and Si. Better recovery of tomato plants after water deficit was conferred by the integrated application of ASE and Si by providing increased leaf greenness, plant water relations, and membrane stability as well as enhanced stomatal conductance and photosynthetic activity. Exogenous application of ASE at 5 mL L⁻¹ in combination with Si at 60 kg ha⁻¹ is recommended when tomato is cultivated on soils with moderate to adequate moisture availability. However, the same combination is not effective at extreme soil moisture deficit condition of 50% FC. Future field experiments using various doses of ASE with Si and soil moisture regimes are needed to confirm the present findings.

Authors Contribution All authors contributed to the study's conception and design. MA acquired the data and performed the statistical analysis with guidance from HU and AD. MA drafted the manuscript, and HU, AA, RT, SC, and AD critically reviewed it for important intellectual content. All authors read and approved the final manuscript.

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Data Availability The datasets used and/or analyzed during the current study are available from the corresponding author on request.

Declarations

Competing interests The authors declare no competing interests.

Ethics Approval Not applicable.

Consent to Participate Not applicable.

Consent for Publication Not applicable.

Conflict of Interest The authors declare no competing interest.

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