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# Effects of seaweed extract on the growth, yield and quality of cherry tomato under different growth conditions

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**ABSTRACT**

An experiment was carried out to determine the effect of foliar application of seaweed extract (0.2 %) on the growth, yield and quality of cherry tomato under stress and non-stress conditions. The greenhouse experiment was set up in a randomized block design with four treatments in three replications. Treatments were as follows: $V_1$ - seedlings treated by seaweed extract and subjected to drought; $V_2$ - seedlings treated by seaweed extract and regularly watered; $V_3$ - non-treated seedlings subjected to drought; $V_4$ - non-treated seedlings regularly watered. Cherry tomato seedlings treated by seaweed extract had a lower content of proline and higher leaf water potential compared to non-treated seedlings under stress conditions, indicating that application of this fertilizer contributes to better adaptation of cherry tomato seedlings to stress. Treatment with seaweed extract also positively influenced the yield and quality of cherry tomato (total soluble solids, vitamin C, lycopene) under both standard and drought stress conditions as compared to untreated plants in same conditions. Positive effects of seaweed extract on growth and quality of cherry tomato are result of its specific composition, as well as ability of cherry tomato plants to utilize bioactive substances in seaweed extracts for its growth and development.

**Key words:** cherry tomato; seaweed extract; osmotic adjustment; photosynthesis; antioxidants; growth conditions

**IZVLEČEK**

**UČINKI IZVLEČKOV MORSKIH ALG NA RAST, PRIDELEK IN KAKOVOST ČEŠNJEVEGA PARADIŽNIKA V RAZLIČNIH RASTNIH RAZMERAH**

Izveden je bil poskus za določanje učinkov foliarnega gnojenja z izvlečkom morskih alg (0, 2 %) na rast, pridelek in kakovost češnjevega paradižnika v stresnih in nestresnih razmerah. V rastlinjaku je bil postavljen naključni bločni poskus s štirimi obravnavanji in tremi ponovitvami. Obravnavanja so bila: $V_1$ – tretma sadik z izvlečkom morskih alg in izpostavitev suši; $V_2$ - tretma sadik z izvlečkom morskih alg in redno zalivanje; $V_3$ – netretirane sadike so bile izpostavljene suši; $V_4$ – netretirane sadike so bile redno zalivane. Sadike češnjevca, ki so bile tretirane z izvlečkom morskih alg, so imele manjšo vsebnost prolina in večji vodni potencial listov v primerjavi z netretiranimi v stresnih razmerah, kar kaže, da je uporaba tega gnojila prispevala k boljši prilagoditvi sadik na stres. Foliarno gnojenje z izvlečekom morskih alg je tudi pozitivno vplivalo na pridelek in kakovost češnjevca (celokupno vsebnost topnih snovi, vitamina C, likopena) v kontroli in stresnih razmerah v primerjavi z netretiranimi rastlinami v enakih razmerah. Pozitivni učinki izvlečka morskih alg na rast in kakovost češnjevca so posledica njegove specifične sestave kot tudi sposobnosti tega paradižnika, da bioaktivne snovi iz izvlečkov morskih alg uporabi za rast in razvoj.

**Ključne besede:** češnjev paradižnik; izvleček morskih alg**;** osmotsko uravnavanje; fotosinteza; antioksidanti; rastne razmere

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## 1 INTRODUCTION

Water stress caused by drought induces morphology, biochemistry and physiology changes in plant, leading to considerable reductions in plant growth and productivity (Atkinson and Urwin, 2012; Li and Mattson, 2015). Besides, drought stress is able to promote reactive oxygen species (ROS) production which in turn leads to damage of all cellular components primarily proteins, lipids and nucleic acids (Ali and Anjum, 2016; Ali et al., 2016).

Plants possess a number of defense mechanisms to cope with stress and some of the more important are osmotic adjustment and efficient antioxidant systems. The osmotic adjustment is indicated by the accumulation of proline, glycine betaine and other metabolites the structural capabilities to maintain homeostasis and improve plant functioning under drought stress (Hayat et al., 2012). The ability of plants to improve their defense mechanism against stress also depends on the possibility of plant to produce secondary metabolites with strong antioxidant activity, among them phenolic compounds (Sanchez-Rodriguez et al., 2011).

Tomatoes are very sensitive to drought (Nuruddin et al., 2003). There are currently several approaches that potentially reduce the impact of drought stress on vegetable cultivation, such as development of drought stress tolerant cultivars, adopting agronomic practices, efficient irrigation systems and use organic fertilizers that can contribute to mitigate drought stress (Tilman et al., 2002; Mikiciuk and Dobromilska, 2014). Application of seaweed extract also might contribute to the strengthening of the plant defense system against stress since the seaweed extracts are very rich in bioactive compounds, including betaine, proline, and aromatic amino acids (Arioli et al., 2015). Currently, many types of seaweed extracts can be purchased for commercial agriculture, especially for vegetable cultivation (Craigie, 2011). Bio-algeen S-92 (Shulze & Hermsen GmbH, Germany) is an organic fertilizer for foliar supplemental feeding, derived from seaweed *Ascophyllum nodosum* (L.) Le Jol. According to the product specification Bio-algeen S-92 contains 96 % water, 0.02 % N, 0.006 % P, 0.096 % K, 0.31 % Ca, 6.3 mg $l^{-1}$ Fe, 1 mg $l^{-1}$ Zn, 0.6 mg $l^{-1}$ Mn, vitamins ($B_1$, $B_3$, $B_6$, $B_9$ and vitamin E), a certain amount of essential amino acids: alanine, glycine, tryptophan, histidine, proline, glutamine and other active natural substances such as organic acids and microelements that provide lots of benefits for plant growth and development (Dobromilska et al., 2008). As far as we know, the possible application of this preparation in greenhouse production of cherry tomato has not been tested so far, especially under drought stress conditions.

Cherry tomato was selected as the subject of this study, particularly because this species is commonly affected by a lack of moisture, and also because this vegetable an important part of a healthy diet.

## 2 MATERIALS AND METHODS

### 2.1 Field experiment

The study was carried out in 2015 under controlled conditions, in the greenhouse of public communal company 'Park' in Sarajevo. In the experiment air temperature in greenhouse was maintained at 23 to 25 °C during day and 20 to 22 °C at night. Relative humidity (RH) was maintained between 60 % and 70 %, with combined venting to reduce RH, and with high-pressure fogging to increase RH. During warm days shade cloth over the top of a greenhouse was used to reduce solar radiation entry.

The first part of the study involved transplanting of cherry tomato seedlings into individual pots (20 cm diameter × 13 cm height), containing substrate Florahum-SP (8 April 2015). Cherry tomato seedlings used in the experiment were produced at a certified nursery located near the greenhouse and showed no significant difference in terms of size and appearance. The substrate used in this study represented a mixture of white and black peat enriched with nutritional supplements. The main chemical characteristics of substrate were as follows: pH 5.5 - 6.5, EC 1.2 - 1.8 mS $cm^{-1}$, content of N 140 - 180 mg $l^{-1}$, content of $P_2O_5$ 160 - 300 mg $l^{-1}$, and content of $K_2O$ 180 - 400 mg $l^{-1}$.

The second part of the study related to setting up an experiment in which the cherry tomato seedlings were treated by 0.2 % solution of Bio-algeen S92, extract from *Ascophyllum nodosum* (ANE). The experimental trial was set up in a randomized block design with four treatments in three replications. Each of these treatments was present with sixty seedlings. The treatments were as follows:

$V_1$ - double foliar treatment with ANE (100 ml per plant) before exposure to drought stress,
$V_2$ - double foliar treatment with ANE (100 ml per plant) and regularly watered (non-stressed),

$V_3$ - without ANE treatment before exposure to drought stress,
$V_4$ - without ANE treatment and regularly watered (non-stressed).
ANE application was carried out manually, using small bottles with sprayers. The first ANE application was done immediately after the transplanting of seedlings (8 April 2015), and the second fifteen days later. Five days after the second treatment, the cherry tomato seedlings ($V_1$ and $V_3$) were exposed to drought stress conditions (non-watering), while the other cherry tomato seedlings ($V_2$ and $V_4$) were not exposed to drought stress, that is, they were regularly watered.

Exposure of cherry tomato seedlings to drought stress conditions lasted until the moment in which first visually observable effects of drought appeared on the seedlings as wilting leaves. This moment was also represented the beginning of the third part of study, which included measurement of selected physiological parameters for evaluating of drought tolerance in cherry tomato seedlings: leaf water potential, leaf area, photosynthetic pigments, content of proline, total phenolic and flavonoid content, and total antioxidant capacity. Leaf water potential was estimated by the dye method (Knipling, 1967), content of proline was measured by acid-ninhydrin method (Bates et al., 1973), photosynthetic pigments were extracted with 80 % acetone (Wettstein, 1957) and the total amount of pigments were determined with equations recommended by Lichtenthaler and Wellburn (1983), leaf area was measured by millimeter graph paper method (Pandey and Singh, 2011), total phenolic content was estimated using Folin Ciocalteu method (Ough and Amerine, 1988), total flavonoids according to Aluminium chloride colorimetric assay (Zhishen et al., 1999), and the ferric reducing/antioxidant power (FRAP) assay was used to determine total antioxidant capacity (Benzie and Strain, 1996).

The next part of study involved the cultivation of cherry tomato under standard growth conditions in all variants until the time of technological maturity of fruits, in order to test how the exposure of seedlings to drought stress and application of ANE affect the yield and quality of fruit. Fruit nutritional quality was analyzed by detecting the following parameters: total soluble solids, titratable acidity, lycopene, vitamin C, total phenolic and flavonoid content, content of rutin and naringenin, and total antioxidant capacity of cherry tomato fruits. Estimation of total soluble solids was performed by digital refractometer according to the International standard method (ISO 2173, 2003), titratable acidity was estimated by titration with NaOH according to AOAC Official method No. 942.15 (AOAC, 2000) and vitamin C by titration with 2,6-dichlorophenolindophenol according to AOAC Official method No. 967.21 (AOAC, 2006).

### 2.2 Estimation of proline

Estimation of proline was carried out as follows: 1 g of fresh leaf samples was homogenized in 3 % (w/v) aqueous 5-sulfosalicylic acid and the homogenate was filtered through a glass-fiber filter to a plastic test tube. 2 ml of filtrate was mixed with 2 ml of ninhydrin reagent and 2 ml of glacial acetic acid in a test tube and boiled for 1 hour at 100 $^0$C (ninhydrin reagent was prepared as follows: 2.5 g of ninhydrin was dissolved in a mixture of 60 ml glacial acetic acid and 40 ml 6 mol $l^{-1}$ phosphoric acid). After termination of reaction in ice bath, the reaction mixture was extracted with 4 ml of toluene, and mixed vigorously with a vortex mixer for 15 - 20 sec. The reddish layer of mixture was transferred to cuvette and absorbance read at 520 nm with a UV/Vis spectrophotometer (Thermo Scientific, Madison, USA) using toluene as blank. The proline concentration was determined from a standard curve (0 - 5 μg $ml^{-1}$) and then the values were recalculated on fresh mass (μg $g^{-1}$ FM).

### 2.3 Estimation of photosynthetic pigments

Extraction of pigments was made from 200 mg of fresh leaves in acetone (80 %) and absorbance of extract was read spectrophotometrically at 662 nm, 645 nm, and 470 nm. The total amounts of pigments were determined with equations recommended by Lichtenthaler and Wellburn (1983) as follows:
Chlorophyll *a* = 11.75 $A_{662}$ - 2.350 $A_{645}$
Chlorophyll *b* = 18.61 $A_{645}$ - 3.960 $A_{662}$
Carotenoids = 1000 $A_{470}$ - 2.270 Chl *a* - 81.4 Chl *b*/227
The results were expressed as mg of pigment per g of fresh mass (mg $g^{-1}$ FM).

### 2.4 Extraction of the plant material

Extraction of phenolic compounds from dry leaves and fruits of cherry tomato was performed in reaction flasks using a 30 % aqueous solution of ethanol. The flasks were boiled at 60 $^0$C for 1 hour using a reflux condenser. Extracts thus obtained were used for the estimation of the total content of phenolic and flavonoids, and total antioxidant capacity.

### 2.5 Estimation of total phenolic content

The total phenolic content of the extract was determined as follows: 0.25 ml of extract, 15 ml of distilled water, and 1.25 ml of Folin-Ciocalteu's reagent (diluted by distilled water in the ratio 1:2) was mixed into 25 ml flask. The mixture was incubated at room temperature for 15 min and then 3.75 ml saturated sodium carbonate solution was added. Flask was filled to the mark with 30 % ethanol and heated in water bath at 50 ºC, for 30

min. After cooling to room temperature absorbance was measured at 765 nm. The total phenolic content was calculated using a standard curve with gallic acid (0 - 500 mg $l^{-1}$), and results were expressed as mg of gallic acid equivalent per g dry mass (mg eq. GA $g^{-1}$ DM).

### 2.6 Estimation of total flavonoids content

The total flavonoid content of the extract was determined by the aluminium chloride colorimetric assay as follows: 1 ml of extract was added to 10 ml volumetric flask containing 4 ml of distilled water and 0.3 ml 5 % $NaNO_2$. After 5 min. 0.3 ml 10 % $AlCl_3$ was added, and the mixture was incubated at room temperature for 6 min. Then 2 ml of 1 mol $l^{-1}$ NaOH was added and the flask was made up to 10 ml with distilled water. The flask was incubated at room temperature for 15 min, and absorbance was read at 510 nm. The total flavonoid content was calculated using a standard curve with catechin (0 - 100 mg $l^{-1}$) end results were expressed as mg of catechin equivalent per g of dry mass (mg eq. C $g^{-1}$ DM).

### 2.7 Estimation of total antioxidant capacity

The total antioxidant capacity of the extract was determined by ferric reducing antioxidant power (FRAP) assay as follows: 240 µl of distilled water, 80 µl of extract, and 2080 µl of FRAP reagent (reagent was obtained by mixing 0.3 mol $l^{-1}$ acetate buffer (pH = 3.6), 10 mmol $l^{-1}$ TPTZ (2,4,6-tripyridyl-s-triazine) and 20 mmol $l^{-1}$ $FeCl_3$ x 6 $H_2O$ in ratio 10 : 1 : 1) were added into a 10 ml Erlenmeyer flask and heated in water bath at 37 ºC, for 5 min and the absorbance was measured at 595 nm. The values of total antioxidant capacity were calculated using a standard curve with $FeSO_4$ x 7 $H_2O$ (0 - 2000 µmol $l^{-1}$) and results were expressed as µmol $Fe^{2+}$ per g of dry mass of extract (µmol $Fe^{2+}$ $g^{-1}$ DM).

### 2.8 Estimation of lycopene

Lycopene content was determined according to method of Davis et al. (2003) as follows: Approximately 0.3 to 0.6 g of the homogenized samples of cherry tomato fruits were weighed in Erlenmeyer flasks and 5 ml of 0.05 % (w/v) butylated hydroxytoluene (BHT) in acetone, 5 ml of ethanol and 10 ml of hexane were added. Samples were extracted on an orbital shaker for 15 min on ice. After shaking, 3 ml of deionized water were added to each flask and the samples were shaken for an additional 5 min on ice. Samples were then left at room temperature for 5 min to allow the separation of both phases. The absorbance of the upper layer was measured in a 1-cm-path-length quartz cuvette at 503 nm blanked with hexane, and results were expressed as mg lycopene per g of fresh mass (µg $g^{-1}$ FM).

### 2.9 Individual flavonoid compounds extraction and analysis

The extraction of samples (5 g) was made in 10 ml of extracted solution (methanol + 3 % formic acid + 1 % m/v 2,6-di-tert-butyl-4-methyilphenol/BHT) according to Escarpa and Gonzales (2000).

Individual flavonoid compounds (naringenin and rutin) were analyzed by using Thermo Scientific Finnigan Surveyor HPLC-DAD system, controlled by a ChromQuest 4.0 chromatography workstation software system (Thermo Scientific, San Jose, CA, USA). Separation of flavonoid compounds was achieved by using Pursuit XRs 3 C-18 column (4.6 × 150 mm, 5 µm; Agilent Technologies, Santa Clara, CA, USA) operated at 25 ºC. The mobile phase consisted of the following linear gradient: 97 % acetonitrile + 3 % redistilled water + 0.1 % formic acid (A) and 97 % redistilled water + 3 % acetonitrile + 0.1 % formic acid (B). Sample injection volume was 20 µL and the flow rate was 0.6 ml $min^{-1}$. The sample was eluted in accordance with method described by Marks et al. (2007). Detection of flavonoid compounds was performed with a diode array detector (DAD) at 280 and 350 nm. Naringenin and rutin were identified on the basis of their retention times and addition of external standards and quantification was made according to concentrations of corresponding external standard and expressed as mg per 100 g of fresh mass (mg 100 $g^{-1}$ FM).

### 2.10 Statistic data processing

All experimental measurements were carried out in triplicate and the results were expressed as mean ± standard deviation. The data obtained were processed by application of standard statistic methods of variance analysis (ANOVA) using Microsoft Excel 2013 software program, and the significant differences between the variants were determined using Least Significant Differences at 0.05 level of probability ($LSD_{0.05}$).

## 3 RESULTS AND DISCUSSION

### 3.1 Leaf water potential

Exposure of cherry tomato seedlings to drought stress caused a decrease in leaf water potential (Ψ) as compared with control (non-stressed seedlings), regardless of ANE treatment, and as expected, with the progression of the stress, plant water potential

decreased. However, the reduction of Ψ being less pronounced for stressed plant treated by ANE (Figure 1), suggesting that this treatment helps the maintain homeostasis in plant cells under stress conditions.

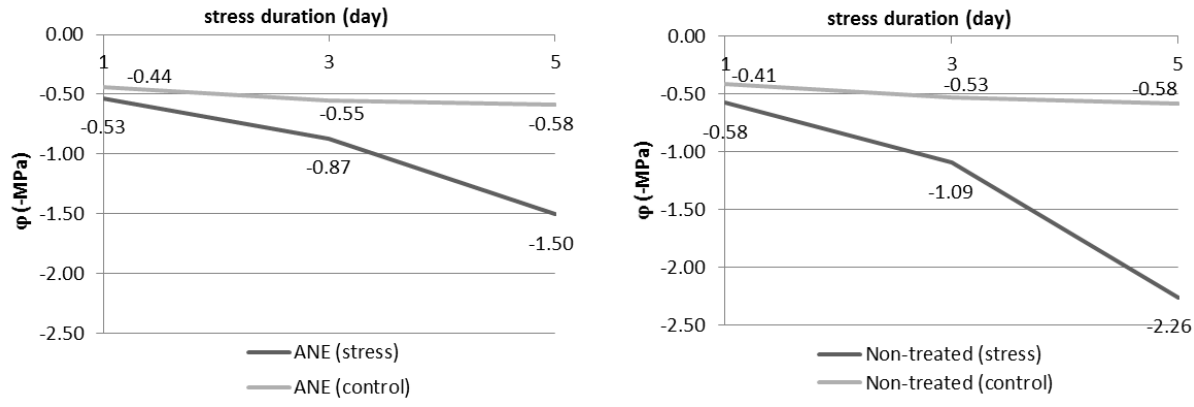


**Figure 1:** Leaf water potential (-MPa) depending on treatment with ANE and exposure to water stress

Numerous studies have also found that ANE application contributes to better osmotic adjustment of plants to stress (Khan et al., 2009; Ha et al., 2014). Karabudak et al. (2014) reported that substances such as glycine betaine and sterols present in ANE act as a buffer against major osmotic changes in plant cells and thus reduces negative effects of stress on plants. In addition, ANE contain many other osmolytes including amino acids: proline, valine, isoleucine and aspartic acid, vitamins and microelements and numerous other active natural substances that improve the stress tolerance of agricultural crops (Spann and Little, 2011).

### 3.2 Proline content

The results of the proline estimation indicated an increase of the proline level in leaves of all cherry tomato seedlings exposed to drought stress conditions. Furthermore, in experiment variant where cherry tomato seedlings were treated by ANE, the increase of proline in leaves during drought stress was much lower compared with non-treated seedlings grown under same conditions (Figure 2).

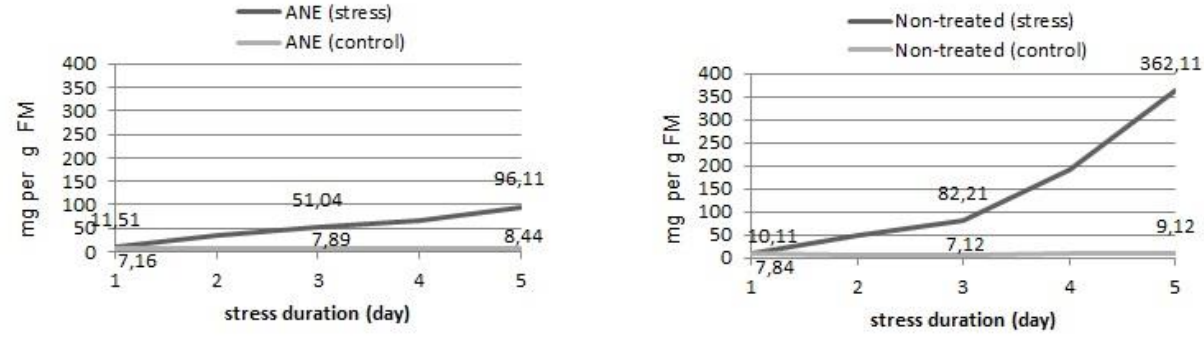


**Figure 3**: Proline content (μg $g^{-1}$ FM) depending on the ANE treatment and exposure to water stress

Since the faster increase of proline content in leaves indicates plant stress, the results of the present study suggest that application of ANE contributes to a better osmotic adjustment of cherry tomato seedlings to stress conditions. Extracts from seaweed have also been reported to reduce drought stress in cultivation of vegetable, ornamental crops and grasses (Zhang and Ervin, 2004; Neily et al., 2010).

### 3.3 Photosynthetic pigments and leaf area

The results of the analysis of photosynthetic pigments showed that the content of chlorophyll *a* (Chl *a*), chlorophyll *b* (Chl *b*) and carotenoids in leaves of

cherry tomato seedlings were decreased under drought stress conditions, regardless of ANE treatment (Table 1). This decrease was statistically significant for pigment Chl *a*, while for pigment Chl *b* and carotenoids were not. Numerous studies have also found that plant reduces the content of pigments in leaves under stress conditions (Ghorbanli et al., 2013; Yuan et al., 2016). Jaleel et al. (2009) reported that reducing the photosynthetic pigments content in leaves may be the result of the impairment in pigment biosynthesis or destruction of pigments due to disturbing in uptake of nutrients under drought stress conditions. Anjum et al. (2011) found that drought causes not only the reduction of the photosynthetic pigments content but it also leads to the destructive changes in the chloroplast, resulting in decreased photosynthetic capacity of plant.

The research results also indicate that ANE application contributes to higher synthesis of photosynthetic pigments in cherry tomato leaves and thus improving plant survival under subsequent stress. González et al. (2013) reported that the efficiency of seaweed extracts application to increase the content of pigments has been mainly attributed to large number of natural nitrogenous compounds present in seaweed which are important for the synthesis of chlorophyll pigments.

**Table 1**: Photosynthetic pigments content and leaf area of cherry tomato seedlings

| Treatment | Chl *a* (mg $g^{-1}$ FM) | Chl *b* (mg $g^{-1}$ FM) | Carotenoids (mg $g^{-1}$ FM) | Leaf area ($cm^2$) |
|---|---|---|---|---|
| $V_1$ ANE (stress) | $1.36 \pm 0.12^{bc}$ | $0.46 \pm 0.03$ | $0.45 \pm 0.01$ | $16.06 \pm 3.01^{bc}$ |
| $V_2$ ANE (non-stress) | $1.51 \pm 0.07^{a}$ | $0.51 \pm 0.10$ | $0.53 \pm 0.11$ | $19.86 \pm 5.88^{a}$ |
| $V_3$ Non-treated (stress) | $1.13 \pm 0.11^{d}$ | $0.45 \pm 0.05$ | $0.46 \pm 0.05$ | $13.21 \pm 2.95^{d}$ |
| $V_4$ Non-treated (non-stress) | $1.38 \pm 0.04^{b}$ | $0.47 \pm 0,05$ | $0.47 \pm 0.05$ | $17.84 \pm 3.54^{b}$ |
| $LSD_{0.05}$ | 0.095 | - | - | 2.186 |

Values expressed as main ± standard deviation.
Different letters in each column represent significant difference among variants at 0.05 level of probability

As shown in Table 1, leaf area in stressed cherry tomato seedlings were lower than in treatments where the seedlings were regularly watered. Many studies have shown the similar results about the effect of drought stress on leaf area (Jureková et al., 2011; Aldana et al., 2014). Galmés et al. (2013) reported that reducing the leaf area of plant under stress conditions is primarily result of reduction of cell enlargement and limited cell division due to lack of water.

The present data also showed that in stressful conditions, cherry tomato seedlings treated by ANE had a higher leaf area compared to untreated plants. This data indicates that the treated plants were less stressed, supporting the hypothesis that the application of ANE postpones and thus reduces negative effects of drought on cherry tomato seedlings.

### 3.4 Total phenolic, total flavonoid content, and antioxidant capacity of leaf extracts

A secondary effect of drought stress on plants is the increased of reactive oxigen species (ROS) such as superoxide radicals ($O^{2-}$), hydroxyl radicals ($OH^{\cdot}$), hydrogen peroxide ($H_2O_2$), and other oxidant substances (Appel and Hirt, 2004). In order to achieve balance between the production and scavenging of ROS, plants activate their defense systems, which include enzymatic and non-enzymatic systems. Plants have ability to synthesize a wide range of antioxidants that can contribute the strengthening of the defense system of the plant, and some of these substances are phenolic compounds. The protection activity of phenolic compounds is achieved mainly due to their redox potential, which allowed them to act as scavenger of free radicals (Atmani et al., 2009).

The results of this study showed that the total antioxidant capacity (FRAP), total phenolic (TPC) and total flavonoid content (TFC) were significantly higher in leaves of cherry tomato seedlings exposed to drought stress than in non-stressed seedlings (Table 2), suggesting that plant initiates the intensive synthesis of phenolic compounds as a response to drought stress, and this hypothesis has been confirmed by many scientists (Basu et al., 2010; Cramer et al., 2011).

**Table 2:** Total phenolic (TPC), total flavonoid content (TFC) and antioxidant capacity (FRAP) in leaves of cherry tomato seedlings

| Treatment | FRAP ($\mu$mol $Fe^{2+}$ $g^{-1}$ DM) | TPC (mg $g^{-1}$ DM) | TFC (mg $g^{-1}$ DM) |
|---|---|---|---|
| $V_1$ ANE (stress) | $140.03 \pm 5.78^{a}$ | $8.06 \pm 0{,}44^{a}$ | $4.08 \pm 0.23^{a}$ |
| $V_2$ ANE (non-stress) | $123.68 \pm 3.29^{c}$ | $7.13 \pm 0{,}22^{b}$ | $3.74 \pm 0.19^{bc}$ |
| $V_3$ Non-treated (stress) | $133.93 \pm 5.63^{b}$ | $7.07 \pm 0{,}06^{bc}$ | $3.86 \pm 0.09^{b}$ |
| $V_4$ Non-treated (non-stress) | $102.87 \pm 1.68^{d}$ | $5.99 \pm 0{,}22^{d}$ | $2.88 \pm 0.09^{d}$ |
| $LSD_{0.05}$ | 5.70 | 0.427 | 0.127 |

Values expressed as main ± standard deviation.
Different letters in each column represent significant difference among variants at 0.05 level of probability

As shown in Table 2 cherry tomato seedlings treated by ANE have a higher content of phenolic and among them flavonoids in leaves of cherry tomato seedlings compared to non-treated seedlings, both in standard as well as stressful growth conditions. These effects can also be attributed to the specific chemical composition of ANE. It's well known that seaweed extracts contains amino acid phenylalanine, tyrosine and tryptophan which serve as precursor for the synthesis of a wide range of phenolic compounds, so it can be assumed that application of ANE promote the synthesis of phenolic compounds in cherry tomato plants.

### 3.5 Yield and quality parameters of cherry tomato fruits

In order to test how the exposure of cherry tomato seedlings to drought and application of ANE impact on the yield and quality parameters of cherry tomato fruits, the analysis of yield and quality parameters of fruit were carried out, and the results are shown in Table 3 and 4.

**Table 3:** Yield, total soluble solids (TSS), titratable acidity (TA) and vitamin C content of cherry tomato fruits

| Treatment | Yield (kg per plant) | TSS (Brix) | TA (%) | Vitamin C (mg 100 $g^{-1}$ FM) |
|---|---|---|---|---|
| $V_1$ ANE (stress) | $1.77 \pm 0.2^{bc}$ | $6.59 \pm 0.13^{ab}$ | $0.64 \pm 0.01^{b}$ | $13.77 \pm 1.34$ |
| $V_2$ ANE (non-stress) | $2.41 \pm 0.25^{a}$ | $6.54 \pm 0.09^{abc}$ | $0.61 \pm 0.02^{c}$ | $13.33 \pm 0.67$ |
| $V_3$ Non-treated (stress) | $1.07 \pm 0.8^{d}$ | $6.66 \pm 0.13^{a}$ | $0.66 \pm 0.02^{a}$ | $13.66 \pm 0.66$ |
| $V_4$ Non-treated (non-stress) | $2.07 \pm 0.41^{ab}$ | $6.34 \pm 0.12^{d}$ | $0.62 \pm 0.01^{c}$ | $13.22 \pm 1$ |
| $LSD_{0.05}$ | 0.395 | 0.126 | 0.019 | - |

Values expressed as main ± standard deviation.
Different letters in each column represent significant difference among variants at 0.05 level of probability

**Table 4:** Lycopene, total phenolic content (TPC), total flavonoid content (TFC) and total antioxidant capacity (FRAP) of cherry tomato fruits

| Variant | Lycopene ($\mu$g $g^{-1}$ FM) | TPC (mg $g^{-1}$ DM) | TFC (mg $g^{-1}$ DM) | FRAP ($\mu$mol $Fe^{2+}$ $g^{-1}$ DM) |
|---|---|---|---|---|
| $V_1$ ANE (stress) | $90.66 \pm 2.69^{a}$ | $10.56 \pm 0.81^{a}$ | $5.39 \pm 0.49^{a}$ | $188.35 \pm 8.37^{ab}$ |
| $V_2$ ANE (non-stress) | $88.25 \pm 2.97^{ab}$ | $9.48 \pm 0.56^{c}$ | $4.71 \pm 0.44^{c}$ | $155.9 \pm 13.41^{c}$ |
| $V_3$ Non-treated (stress) | $88.25 \pm 2.12^{ab}$ | $10.44 \pm 1.06^{ab}$ | $5.31 \pm 0.34^{ab}$ | $194.26 \pm 10.82^{a}$ |
| $V_4$ Non-treated (non-stress) | $86.17 \pm 3.11^{b}$ | $8.55 \pm 0.37^{d}$ | $4.26 \pm 0.12^{d}$ | $141.43 \pm 4.90^{d}$ |
| $LSD_{0.05}$ | 2.47 | 0.617 | 0.346 | 12.91 |

Values expressed as main ± standard deviation.
Different letters in each column represent significant difference among variants at 0.05 level of probability

As shown in Table 3, total soluble solids, titratable acidity and vitamin C of fruits were higher in plants exposed to stress as compared to non-stressed plants, regardless of ANE treatment, but for vitamin C, that increase was not statistically justified. The total antioxidant capacity and the content of secondary metabolites such as lycopene, phenolic and flavonoids were also significantly higher in fruits of cherry tomato exposed to drought stress (Table 4). Besides, analyses of individual flavonoid compound indicate that the

dominant flavonoid in fruits of cherry tomato naringenin and rutin, also accumulates more in cherry tomato fruits as response to drought stress (Table 5).

**Table 5:** Naringenin and rutin content of cherry tomato fruits

| Treatment | Naringenin (mg 100 $g^{-1}$ FM) | Rutin (mg 100 $g^{-1}$ FM) |
|---|---|---|
| $V_1$ ANE (stress) | $3.87 \pm 0.36^{a}$ | $6.62 \pm 0.19^{b}$ |
| $V_2$ ANE (non-stress) | $3.01 \pm 0.21^{bc}$ | $5.64 \pm 0.08^{c}$ |
| $V_3$ Non-treated (stress) | $3.29 \pm 1.03^{b}$ | $7.55 \pm 0.07^{a}$ |
| $V_4$ Non-treated (non-stress) | $2.89 \pm 0.14^{c}$ | $5.71 \pm 0.63^{c}$ |
| $LSD_{0.05}$ | 0.306 | 0.217 |

Values expressed as main ± standard deviation.
Different letters in each column represent significant difference among variants at 0.05 level of probability

These results indicate that the contents of antioxidants in plant are closely related to the growth conditions and that their content in plant increases if the plant is exposed to controlled drought stress conditions. Many studies have shown the similar effect of drought stress on the content of antioxidant substances and generally secondary metabolites in tomato fruits (Atkinson et al., 2011; Giannakoula and Ilias, 2013). There is therefore no doubt that higher production of secondary metabolites is one of basic response of plant to controlled drought stress and conclusions of many studies support this hypothesis (Murshed et al., 2013; Okunlola et al., 2015). These observations are very interesting in terms of improving cherry tomato quality since that vegetable containing phytochemicals with high antioxidant power are drawing increased interest from consumers (Kubota et al., 2006).

The negative impact of drought stress on cherry tomato fruits in the present study was related to the yield, that was significantly lower in stressed plants, what was expected since the lack of water causes losses in tissue water content which reduce turgor pressure in cell, thereby inhibiting enlargement and division of cell, causing of reduce of yield. However, in experiment where cherry tomato seedlings were treated by ANE before exposure to stress, the yield was significantly higher compared to non-treated plants exposed to stress, indicating that this fertilizer reduces negative effects of drought on yield.

Furthermore, treatment of cherry tomato seedlings with ANE, has significantly contributed to increase the content of phenolic and flavonoids in fruits of cherry tomato under standard (non-stress) growth conditions, confirming that this fertilizer stimulate the synthesis of phenolic compounds, and thereby strengthen antioxidant defense mechanism of the plant. This fertilizer was also positively influenced by some quality parameters of cherry tomato (total soluble solids, content of ascorbic acid and lycopene) under standard growth conditions (non-stress) as compared to untreated plants.

## 4 CONCLUSIONS

ANE application in cherry tomato cultivation contributes to better adaptation of seedlings to drought conditions. The application of ANE also positively influenced by the yield and quality of cherry tomato under both standard and drought stress conditions as compared to untreated plants in same conditions. Positive effects of application of ANE are result of its specific composition, as well as ability of cherry tomato plants to utilize bioactive substances in seaweed extracts for its growth and development. The results of this study also indicate that the controlled exposure of cherry tomato plants to drought stress improves fruit quality, increasing nutritional components but decreasing yield.

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