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ORIGINAL PAPER

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EFFECT OF BIOSTIMULANTS ON CHLOROPHYLL FLUORESCENCE PARAMETERS OF BROCCOLI (*Brassica oleracea* var. *Italica*) UNDER DROUGHT STRESS AND REWATERING

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ABSTRACT

The aim of the research was to determine the influence of biostimulants amino acids and amino acids + *Ascophyllum nodosum* filtrate on two broccoli cultivars 'Agassi' and 'Tiburon' and their response to soil drought. The plants were watered with *Ascophyllum nodosum* filtrate before planting and sprayed with amino acids after planting three times. Chlorophyll fluorescence measurements were performed before, during and after stress. They showed a considerable difference in cultivars' response to stress, with 'Agassi' being more sensitive. Application of biostimulants enhanced the tolerance to drought stress. Maximum photochemical efficiency of PSII was unchanged, whereas the quantum yield of electron transport and photochemical fluorescence quenching values increased and the non-photochemical fluorescence quenching decreased. Moreover, the apparent photosynthetic electron transport rate rose. Chlorophyll content index was affected by the cultivar and application of biostimulants.

Key words: amino acids, *Ascophyllum nodosum* filtrate, quantum yield of electron transport, fluorescence quenching, chlorophyll

Abbreviations: F_0 – initial fluorescence, F_v/F_m – maximum photochemical efficiency of PSII, F_m – maximal fluorescence in the dark-adapted state, Y – quantum yield of electron transport, qP – photochemical fluorescence quenching, qN – non-photochemical fluorescence quenching, ETR – apparent photosynthetic electron transport rate, CCI – chlorophyll content index

INTRODUCTION

Plant biostimulants include diverse substances and microorganisms that enhance plant growth [Calvo et al. 2014]. They are classified into the following major groups: humic and fulvic acids, protein hydrolysates, seaweed extracts, chitosan and other biopolymers, inorganic compounds, and beneficial fungi and bacteria [du Jardin 2015]. Some of them influence the growth of shoots and roots [Lisiecka et al. 2011], as well as yield quality and quantity [Mikiciuk and Dobromilska 2014]. Both protein hy-

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drolysates and seaweed extracts affect primary and secondary metabolism [Ertani et al 2013, Xu and Leskovar 2015]. They lead to an increase in antioxidant content, such as carotenoids, polyphenols and flavonoids [Colla et al. 2015].

Seaweed extracts contain cytokinins, auxins, abscisic acid, gibberellins, sterols and polyamines. They are also a source of micro and macronutrients [du Jardin 2015]. A typical response of plants to seaweed extracts is an increase in chlorophyll content. The effect is caused by the cytokinins found in seaweed extracts which impart protective effects on chloroplast [Zavaleta-Mancera et al. 2007] and, consequently, affect the chlorophyll content.

Protein hydrolysates have a considerable impact on the enzymes participating in carbon and nitrogen metabolism [Ertani et al. 2013]. An increased assimilation process after using protein hydrolysates may result from their beneficial effect on production of C skeletons and energy needed for amino acid biosynthesis [Colla et al. 2015].

Drought stress is one of the main factors limiting the yield quality and quantity of vegetables. It is especially important in plants with high water requirements, broccoli being one of them. The experiments conducted on various plant species have proved that application of biostimulants may lead to increased resistance to stress conditions [Ertani et al. 2013, Calvo et al. 2014]. However, to date the research on the effect of biostimulants on broccoli plants is limited. Hence, the aim of the experiment presented in this paper was to determine the response of two broccoli cultivars to drought stress after the application of two popular and easily accesssible biostimulants containing amino acids and *Ascophyllum nodosum* filtrate. The measurements were taken prior to and during the stress treatment, and also after rewatering to reveal the possibility to plants to regenerate, if possible.

To determine the changes occurring in plants under drought stress, chlorophyll fluorescence parameters were measured. The method is commonly used to record physiological changes in plants under different stress factors in many plant species [Björkman and Demmig 1987, Rohaček 2002, Dias and Brűggemann 2010, Bączek-Kwinta et al. 2011]. In our study the changes in the following parameters were discussed: initial fluorescence, maximum photochemical efficiency of PSII, quantum yield of electron transport, photochemical and non-photochemical fluorescence quenching, maximum primary yield of photochemistry of photosystem II and photosynthetic electron transport rate.

MATERIALS AND METHODS

Plant materials and treatments

The experiment was conducted on plants of two broccoli cultivars, 'Agassi' and 'Tiburon' in a growing chamber. The temperature was 18/16°C (day/night), photoperiod 16 hours, relative humidity 90%, photosynthetic photon flux density 150 $\mu mol \cdot m^{-2}\ s^{-1}$. Broccoli seedlings were produced in 0.09 dm^3 pots filled with peat substrate for growing cruciferous vegetables (Kronen-Klasmann, Klasmann–Deilman Polska Sp. z o.o., Poland). When the seedlings had three-four leaves, they were transplanted to bigger containers (5 dm^3). Before planting, the nutrients were supplemented to the maximum optimum level ($mg \cdot dm^{-3}$ of substrate): $N–NO_3$ – 250; P – 200; K – 600; Ca – 1600; Mg – 160 + microelements (Fe – 10; Mn – 3; Cu – 12; B – 3; Zn – 1; Mo – 1). Additionally, during the growing period the plants were fed with 0.5% Kristalon^{TM} blue (Yara Poland Sp. z o.o.).

The plants were watered with *Ascophyllum nodosum* filtrate (Arysta LifeScience Polska Sp z o.o., Poland) three days before planting and sprayed with amino acids (Arysta LifeScience Polska Sp z o.o., Poland) three times – two, four and six weeks after planting. Control plants were not treated with biostimulants. Drought stress was achieved by discontinuation of watering for two days. Physiological measurements were taken before stress, during stress and two days after rewatering. The water content in the substrate was determined with moisture sensors 5TE and Em50 data logger (Decagon Devices, Inc., USA). The water capacity of the soil was a 40% (v/v) and during the drought stress water content decreased to 15% (v/v). After rewatering water content increased to 40% (v/v).

Measurements

Plant stress level caused by the studied factors was determined by measuring chlorophyll fluorescence (OS1–FL Fluorometer, OptiSciences Inc., USA). Fluorescence parameters: F_0, F_m, F_v were determined after eight hours of darkness using photosynthetic photon flux density (PPFD) <0.15 μmol·m^{-2}s^{-1}. F_m was measured after 0.8 s saturating white light pulse (>15 000 μmol·m^{-2}s^{-1} PPFD). F_v/F_m was calculated. Y was calculated according to Genty et al. [1989], while qP and qN parameters were calculated according to Schreiber et al. [1986].

Chlorophyll content index (CCI) was measured with OSI CCM–200 Plus leaf chlorophyll meter (ADC BioScientific Ltd., UK) on the same leaves that were used for chlorophyll fluorescence measurements.

Statistical analysis

The experiment was established as a three-factor design, in four replicates (one plant in each). The significance of the biostimulants and the time of measurement (before drought stress, during stress and after rewatering) to the physiological parameters was determined with the ANOVA. Differences between the means were estimated with the Newman-Keuls test at the significance level of P = 0.05.

RESULTS

Analysis of ANOVA showed that the values of F_0, F_m, Y, qP, qN, ETR and CCI parameters depended on the cultivar, while there was no such correlation for F_v/F_m. Application of biostimulants affected F_v/F_m values significantly and had a considerable influence on Y, qP, qN, ETR, and CCI (tabs 1 and 2).

Table 1. ANOVA F-values for F_0, F_m, F_v/F_m, and Y parameters

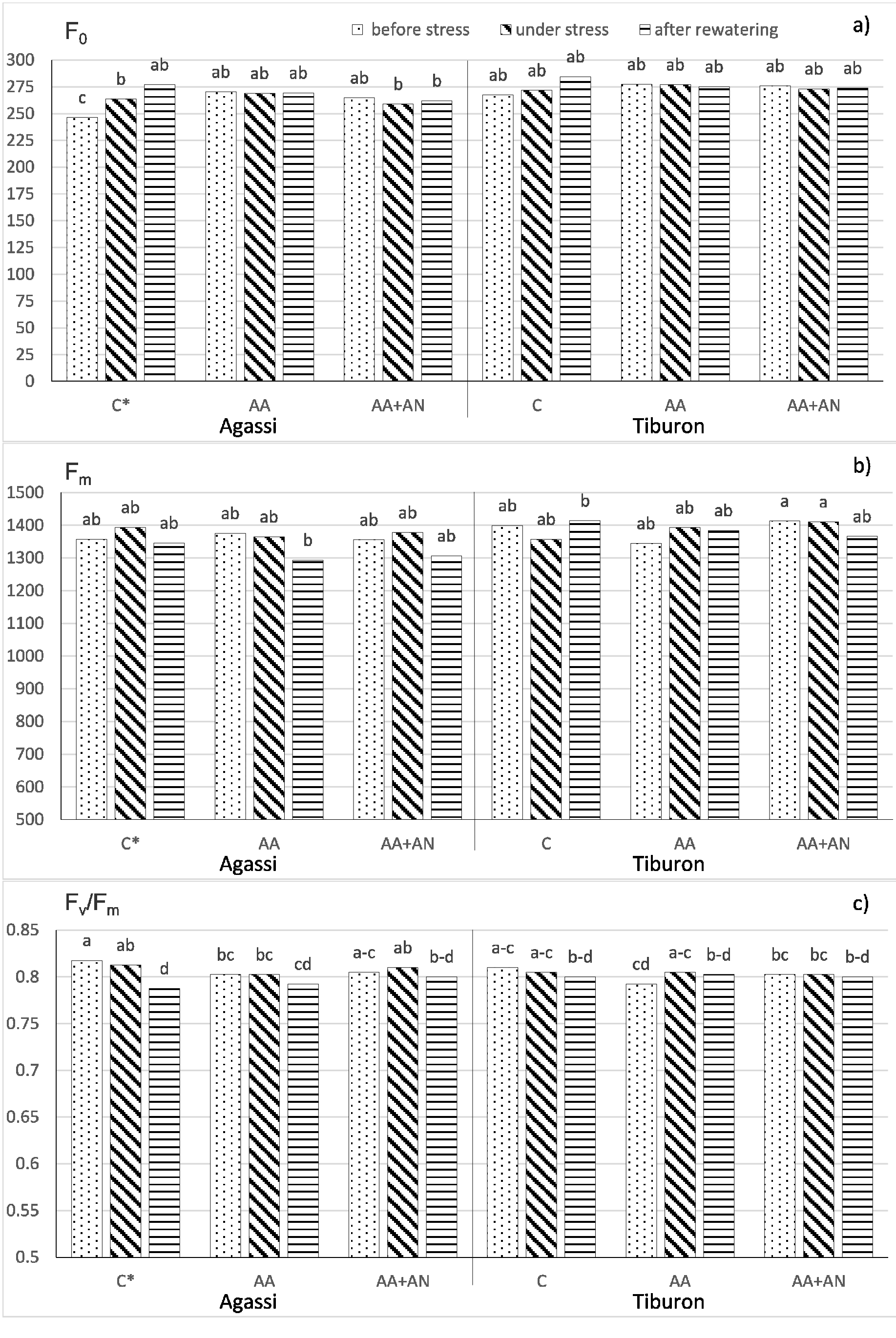
Object	F_0	F_m	F_v/F_m	Y
A	30.01**	11.95**	0.47 ^{ns}	38.44**
B	2.75 ^{ns}	1.20 ^{ns}	4.45*	33.66**
A × B	0.83 ^{ns}	0.61 ^{ns}	0.56 ^{ns}	57.16**
C	4.17*	3.40*	12.58**	7.46**
A × C	0.51 ^{ns}	3.87*	7.28**	16.70**
B × C	6.77**	1.33 ^{ns}	4.85**	29.78**
A × B × C	0.67 ^{ns}	1.91 ^{ns}	1.66 ^{ns}	12.78**

A – cultivar, B – treatment (control, AA, AA + AN), C – term of measurementbefore stress, under stress, after rewatering)
* significance level P = 0.90
** significance level P = 0.95, ^{ns} not significant at P = 0.95

Table 2. ANOVA F-values for qP, qN, ETR and CCI parameters

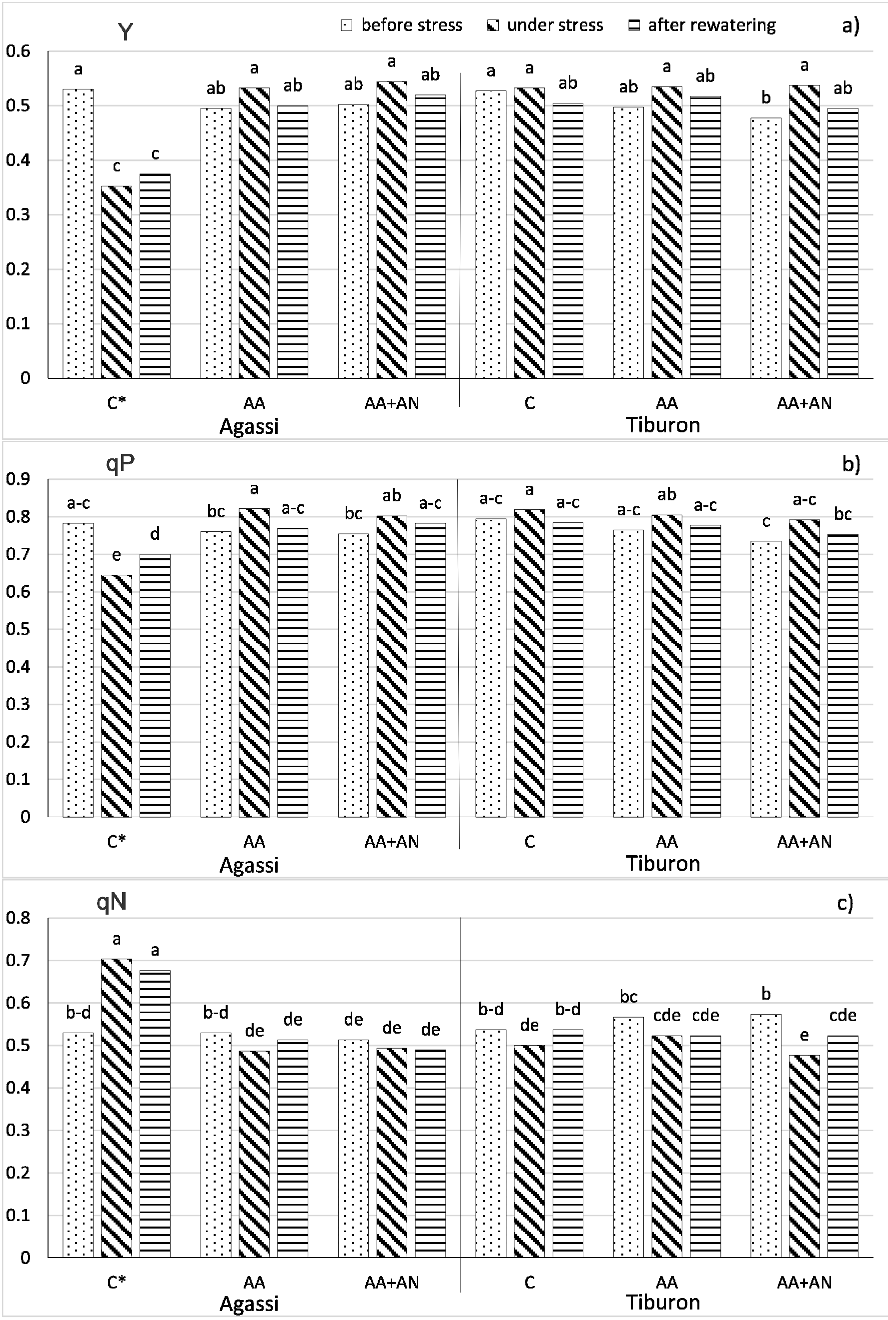
Object	qP	qN	ETR	CCI
A	15.64**	45.07**	71.01**	23.17**
B	8.12**	48.38**	64.77**	6.62**
A × B	34.62**	49.67**	86.94**	1.01 ^{ns}
C	4.37*	97.88**	7.85**	20.99**
A × C	6.17**	8.74**	31.14**	7.13**
B × C	12.98**	81.06**	33.05**	3.28*
A × B × C	8.22**	69.32**	24.46**	1.28 ^{ns}

Explanations, see Table 1



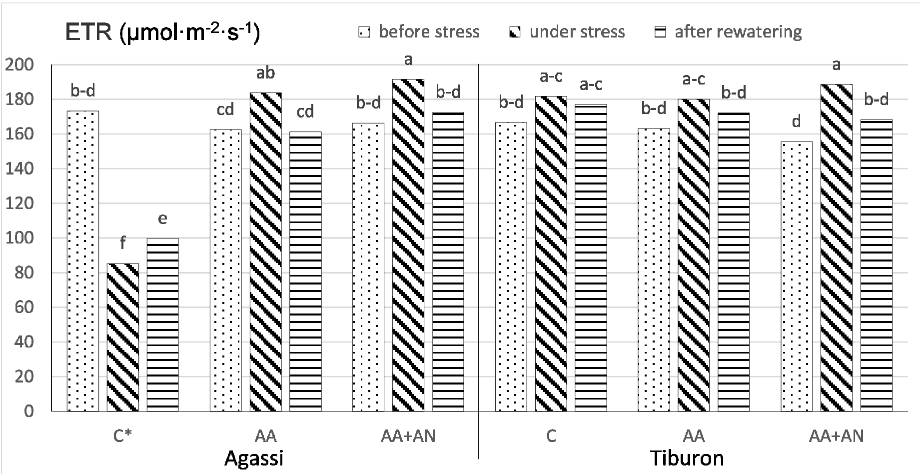
* C = Control; AA = amino acids; AA + AN = amino acids + *Ascophyllum nodosum* filtrate

Fig. 1. The influence of AA and AA + AN on F_0, F_m and F_v/F_m parameters of two broccoli cultivars before drought stress, under stress and after rewatering. The values are means of four determinations SD. Means sharing the same letter are not significantly different from each other



* C = Control; AA = amino acids; AA + AN = amino acids + *Ascophyllum nodosum* filtrate

Fig. 2. The influence of AA and AA + AN on Y, qP and qN parameters of two broccoli cultivars before drought stress, under stress and after rewatering. The values are means of four determinations SD. Means sharing the same letter are not significantly different from each other



* C = Control; AA = amino acids; AA + AN = amino acids + *Ascophyllum nodosum* filtrate

Fig. 3. The influence of AA and AA + AN on ETR parameter of two broccoli cultivars before drought stress, under stress and after rewatering. The values are means of four determinations SD. Means sharing the same letter are not significantly different from each other

Under drought stress the changes in chlorophyll fluorescence parameters depended on the biostimulants and the cultivar. In 'Agassi' control plants, both under drought stress and after rewatering, the value of F_0 increased in comparison with the value before stress (fig. 1 a). No significant changes were observed in the plants treated with biostimulants, both amino acids (AA) and amino acids + *Ascophyllum nodosum* filtrate (AA+AN) in 'Agassi' and in all objects in 'Tiburon' cultivar (fig. 1 b). F_m value decreased only in 'Agassi' plants after rewatering when AA were applied (fig. 1 b). A different tendency was observed for F_v/F_m values which did not differ significantly in 'Agassi' objects treated with biostimulants, while dropped in control plants to the lowest level. F_v/F_m values slight increase in 'Tiburon' plants during stress when AA were used. The difference still could have been noticed after rewatering (fig. 1 c).

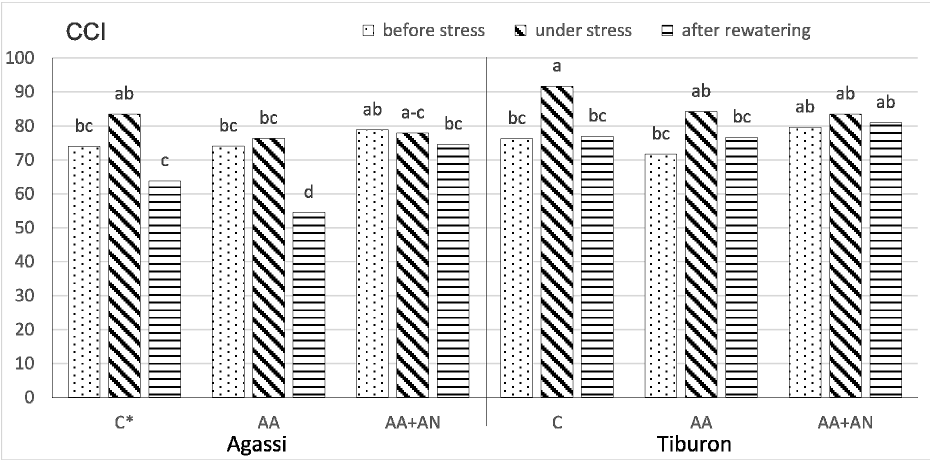
Plants treated with biostimulants showed an increased Y value in 'Agassi' cultivar (fig. 2 a). An increase in qP and a decrease in qN only in 'Agasssi' cultivar in comparison with the control object was observed (fig. 2 b, c).

When biostimulants were used, the ETR value in 'Agassi' cultivar increased during drought stress (fig. 3 a). It should be emphasized that during stress a considerable decline in ETR value in 'Agassi' control plants was observed, while the value increased in the 'Tiburon' cultivar. There were no statistically significant differences in ETR value between the control objects and the plants treated with biostimulants during drought stress in the 'Tiburon' plants.

The highest value of CCI in control object in 'Tiburon' cultivar was stated, while the lowest after AA treatment in 'Agassi' cultivar.

DISCUSSION

Measurement of chlorophyll fluorescence is a non--invasive method of determining the physiological state of plants [Roháček and Barták 1999, Zlatev and Yordanov 2004, Efeoglu et al. 2009]. Our study aimed to determine the influence of the applied biostimulants on the following parameters of chlorophyll fluorescence: F_0, F_m, F_v/F_m, Y, qP, qN and ETR.



* C = Control; AA = amino acids; AA + AN = amino acids + *Ascophyllum nodosum* filtrate

Fig. 4. The influence of AA and AA + AN on CCI of two broccoli cultivars before drought stress, under stress and after rewatering. The values are means of four determinations SD. Means sharing the same letter are not significantly different from each other

In plants subjected to drought stress F_0 increases and F_m decreases [Zlatev and Yordanov 2004, Bączek-Kwinta et al. 2011, Caulet et al. 2014]. F_0 increase may result from inhibition of the acceptor side of PSII or indicate the occurence of photoinhibitory damage in response to water stress [Bertamini and Nedunchezhian 2003]. In our study, there was an increase in F_0 only in the control object indicating the protective impact of biostimulants on photosynthetic apparatus in broccoli.

In our experiment, F_m decrease in comparison with the value before the stress was found only after rewatering following application of AA in 'Agassi'. In 'Tiburon' plants treated biostimulants the changes in F_m values were not significant (fig. 1 b). A decrease in the values of F_m under drought stress was also observed in the study by Efeoglu et al. [2009]. According to Borek et al. [2016] F_0 and F_m values are connected with the content of photosynthetic pigments, and low content of photosynthetic pigments results in low F_0 and F_m values. However, our study does not confirm this correlation. A significantly lower value of F_0 was found only in the control object in 'Agassi' cultivar before drought stress, whereas CCI was considerably lower after rewatering (fig. 4).

F_v/F_m reflects the potential quantum efficiency of PSII and is used as a sensitive indicator of plant photosynthesis performance [Maxwell and Johnson 2000]. F_v/F_m decrease in stress conditions reveals that the photochemical capacity of PSII is diminished [Demmig and Björkman 1987], which is connected with a decrease in the transport of electrons from PSII to PSI [Sikder et al. 2015]. According to Brestic and Zivcak [2013], the values of F_v/F_m start to decline below 70% of relative water content in leaf. A significant decrease in the value of F_v/F_m under drought stress was also confirmed by Rahbarian et al. [2011] and Hu et al. [2010], while a slight decline in the value of F_v/F_m was found by Zlatev and Yordanov [2004] or Bączek-Kwinta el al. [2011]. In our study, the value of F_v/F_m in the 'Agassi' cultivar significantly decreased only after rewatering in the control object (fig. 1 c). In plants in which AA and AA+AN were used there was no difference in the value of F_v/F_m before stress, during stress, and after rewatering. In 'Tiburon' the F_v/F_m value in the control object slight decreased after rewatering, compared with the value prior to stress. It slight increased, however, during stress, and after rewatering in plants treated with AA (fig. 1c). A decrease in F_v/F_m after rewatering in the control object of 'Agassi' plants was accompanied by a decline in CCI value (fig. 4), which is consistent with the study by Borek et al. [2016].

In our study, the values of Y and qP in the control of 'Agassi' cultivar fell during drought stress and after rewatering, while qN value increased (fig. 2a, b and c, respectively). In 'Tiburon' qP value not changed. The decline in qP in the 'Agassi' plants in the control object suggests that the degree of openness of the PSII reaction center was lower than that of the 'Tiburon' ones [Li et al. 2006]. A decrease in the value of qP and an increase in qN under drought stress were also found by Subrahmanyam et al. [2006] and Przybysz et al. [2010] after applying the biostimulant Asahi, in drought stress conditions, too. The reduction of Y and photochemical quenching (qP) values suggests that Y is dependent mainly on the proportion of the reaction centers, which are photochemically "open", rather than on the efficiency of the absorbed photons in reaching a reaction center. The increase in qN accompanied by a decrease in F_v/F_m is the result of the large proportion of absorbed light energy not being used by the plants in the photosynthesis process [Zlatev and Yordanov 2004]. Hence, no change in the value of F_v/F_m, an increase in Y and qP values, a decrease in qN during drought stress in plants treated with biostimulants in our study, indicate their higher stress resistance as compared to the control.

Changes in chlorophyll fluorescence parameters are closely related to changes in basic physiological features such as photosynthetic rate (A), stomatal conductance (g_s) and internal CO_2 concentration (C_i). In the first part of our research [Kałużewicz et al. 2017] conducted on 'Agassi' and 'Tiburon' cultivars in the same conditions as presented in this paper, drought stress resulted in a more than fourfold decrease in A value, double fold decrease in g_s and a significant increase in C_i but only in control plants of 'Agassi' cultivar. This correlation was not found in 'Tiburon' cultivar. In both cultivars in plants treated with biostimulants, drought stress did not change A and C_i values, which indicates their increased tolerance to stress.

ETR is a measure of the electron transfer rate in leaves [Li et al. 2006]. According to Hura et al. [2007] the value of ETR under drought stress in wheat plants decreased from several to several tens of percent, depending on the cultivar. In our experiment, the value of ETR in 'Agassi' significantly decreased during drought stress and after rewatering only in the control plants, while it significantly increased in the plants treated with biostimulants during stress (fig. 3a). In the 'Tiburon' cultivar under drought stress the ETR value increased after AA+AN treatment. A similar tendency was also noted by Caulet et al. [2014] in two strawberry cultivars treated with furostanol glycosides. The increase in the value of Y and ETR meant that the PSII reaction centers were open, captured the light energy for photochemical reactions and photosynthesis proceeded normally.

According to many authors drought stress resulted in lower chlorophyll content [Arzani and Yazdani 2008, Farooq et al. 2009, Guerfel et al. 2009]. The correlation was not observed in our own study either in the control object or after application of biostimulants (fig. 4). The lack of correlation between *Ascophyllum nodosum* treatment and chlorophyll content was also confirmed by Xu and Leskovar [2015].

In our research CCI value depended on both the cultivar and the applied biostimulants (tab. 1). However, no significant interaction between these two factors was found. A considerable differences in chlorophyll content in different wheat cultivars was observed by Chandrasekar et al. [2000], and the cultivars that were more resistant to drought stress were characterised by higher chlorophyll content. Our study confirmed a significant effect of interaction between the cultivar and time of measurement (before, during or after drought stress) on the CCI value. However, an increase level of chlorophyll content was found only in the control object in 'Tiburon' cultivar.

CONCLUSIONS

The study shows that the response of broccoli cultivars to drought stress varied, with 'Agassi' being characterized by greater sensitivity. Plants of 'Agassi' cultivar treated with biostimulants had greater tolerance to drought stress. This was confirmed by the lack of change in F_v/F_m, accompanied by an increase in the value of Y and qP and a decrease in qN. In those plants, an increase in the value of ETR were observed. The CCI value depended on both the cultivars and the biostimulant treatment.

REFERENCES

Arzani, K., Yazdani, N. (2008). The influence of drought stress and paclobutrazol on quantitative changes of proteins in olive (*Olea europaea* L.) cultivars Bladi and Mission. Acta Hortic., 791, 527–530. DOI: 10.17660/Acta Hortic.2008.791.81.

Bączek-Kwinta, R., Kozieł, A., Seidler-Łożykowska, K. (2011). Are the fluorescence parameters of German chamomile leaves the first indicators of the anthodia yield in drought conditions? Photosynthetica, 49, 87–97. DOI: 10.1007/s11099-011-0013-3.

Bertamini, M., Nedunchezhian, N. (2003). Photoinhibition of photosynthesis in mature and young leaves of grapevine (*Vitis vinifera* L.). Plant Sci., 164, 635–644. DOI: https://doi.org/10.1016/S0168-9452(03)00018-9.

Björkman, O., Demmig, B. (1987). Photon yield of O_2 evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. Planta, 170, 489–504. DOI: 10.1007/BF00402983.

Borek, M., Bączek-Kwinta, R., Rapacz, M. (2016). Photosynthetic activity of variegated leaves of *Coleus* × *hybridus* hort. Cultivars characterized by chlorophyll fluorescence techniques. Photosynthetica, 54(3), 331–339. DOI: 10.1007/s11099-016-0225-7.

Brestic, M., Zivcak, M. (2013). PSII fluorescence techniques for measurement of drought and high temperature stress signal in crop plants: protocols and applications. In: Molecular stress physiology of plants. Rout, G.R., Das, A.B. (eds). Springer–Verlag, Berlin–Heidelberg, pp., 87–131. DOI: 10.1111/j.1469-8137.2004.01056.x.

Calvo, P., Nelson, L., Kloepper, J.W. (2014). Agricultural uses of plant biostimulants. Plant Soil, 383, 3–41. DOI: 10.1007/s11104-014-2131-8.

Caulet, R.P., Gradinariu, G., Lurea, D., Morariu, A. (2014). Influence of furostanol glycosides treatments on strawberry (*Fragaria*×*ananassa* Duch.) growth and photosynthetic characteristics under drought condition. Sci. Hort., 169, 179–188. DOI: https://doi.org/10.1016/j.scienta.2014.02.031.

Chandrasekar, V., Sairam, R.K, Srivastava, G.C. (2000). Physiological and biochemical responses of hexaploid and tetraploid wheat to drought stress. J. Agron. Crop Sci., 185, 219–227.

Colla, G., Nardi, S., Cardarelli, M., Ertani, A., Lucini, L., Canaguier, R., Rouphael, Y. (2015). Protein hydrolysates as biostimulants in horticulture. Sci. Hort., 196, 28–38.

Demmig, B., Björkman, O. (1987). Comparison of the effect of excessive light on chlorophyll fluorescence (77K) and proton yield of O_2 evolution in leaves of higher plants. Planta, 171, 171–184. DOI: 10.1007/BF00391092.

Dias, M.C., Brűggemann, W. (2010). Limitations of photosynthesis in *Phaseolus vulgaris* under drought stress: gas exchange, chlorophyll fluorescence and Calvin cycle enzymes. Photosynthetica, 48, 96–102. DOI: 10.1007/s11099-010-0013-8.

du Jardin, P. (2015). Plant biostimulants: Definition, concept, main categories and regulation. Sci. Hort., 196, 3–14. DOI: https://doi.org/10.1016/j.scienta.2015.09.021.

Efeoglu, B., Ekmekçi. Y., Çiçek, N. (2009). Physiological responses of three maize cultivars to drought stress and recovery. S. Afr. J. Bot., 75, 34–42. DOI: https://doi.org/10.1016/j.sajb.2008.06.005.

Ertani, A., Schiavon, M., Muscolo, A., Nardi, S. (2013). Alfalfa plant-derived biostimulant stimulate short–term growth of salt stressed *Zea mays* L. plants. Plant Soil, 364, 145–158. DOI: 10.1007/s11104-012-1335-z.

Farooq, M., Wahid, A., Kobayashi, N., Fujita, D., Basra S.M.A. (2009). Plant drought stress: effects, mechanisms and management. Agron. Sustain. Dev., 29, 185–212. DOI: 10.1051/agro:2008021.

Genty, B., Briantais, J.M., Baker, N.R. (1989). The relationship between the quantum yield of photosynthetic electron transport and quenching of chlorophyll fluorescence. Biochim. Biophys. Acta., 990, 87–92. DOI: https://doi.org/10.1016/S0304-4165(89)80016-9.

Guerfel, M., Baccouri, O., Boujnah, D., Chaïbi, W., Zarrouk, M. (2009). Impacts of water stress on gas exchange, water relations, chlorophyll content and leaf structure in the two main Tunisian olive (*Olea europaea* L.) cultivars. Sci. Hort., 119, 257–263. DOI: https://doi.org/10.1016/j.scienta.2008.08.006.

Hu, W.H., Xiao, Y.A., Zeng, J.J., Hu, X.H. (2010). Photosynthesis, respiration and antioxidant enzymes in pepper leaves under drought and heat stresses. Biol. Plantarum, 54, 761–765. DOI: 10.1007/s10535-010-0137-5.

Hura, T., Grzesiak, S., Hura, K., Thiemt, E., Tokarz, K., Wędzony, M. (2007). Physiological and biochemical tools useful in drought-tolerance detection in genotypes of winter triticale: accumulation of ferulic acid correlates with drought tolerance. Ann. Bot., 100, 767–775. DOI: org/10.1093/aob/mcm162.

Kałużewicz, A., Krzesiński W., Spiżewski T., Zaworska A. (2017). Effect of biostimulants on several physiological characteristics and chlorophyll content under drought stress and re-watering. Not. Bot. Hortic. Agrobot., 45, 197–202. DOI: 10.15835/nbha45110529.

Li, R., Guo, P., Baum, M., Grando, S., Ceccarelli, S. (2006). Evaluation of chlorophyll content and fluorescence parameters as indicators of drought tolerance in barley. Agri. Sci. China, 5, 751–757.

Lisiecka, J., Knaflewski, M., Spiżewski, T., Frąszczak, B., Kałużewicz, A., Krzesiński, W. (2011). The effect of animal protein hydrolysate on quantity and quality of strawberry daughter plants cv. 'Elsanta'. Acta Sci. Pol. Hortorum Cultus, 10, 31–40.

Maxwell, K., Johnson, G.N. (2000). Chlorophyll fluorescence – a practical guide. J. Exp. Bot., 51, 659–668. DOI: https://doi.org/10.1093/jexbot/51.345.659.

Mikiciuk, M., Dobrolmilska, R. (2014). Assessment of yield and physiological indices of small-sized tomato cv. 'Bianka F1' under the influence of biostimulators of marine algae origin. Acta Sci. Pol. Hortorum Cultus, 13, 31–41.

Przybysz, A., Wrochna, M., Słowiński, A., Gawrońska, H. (2010). Stimulatory effect of Asahi SL on selected plant species. Acta Sci. Pol. Hortorum Cultus, 9, 53–64.

Rahbarian, R., Khavari-Nejad, R., Ganjeali, A., Bagheri, A., Najafi, F. (2011). Drought stress effects on photosynthesis chlorophyll fluorescence and water relations in tolerant and susceptible chickpea (*Cicer arietinum* L.). Acta Biol. Cracov. Bot., 53, 47–56. DOI: 10.2478/v10182-011-0007-2.

Roháček, K. (2002). Chlorophyll fluorescence parameters: the definitions, photosynthetic meaning, and mutual relationships. Photosynthetica, 40, 13–29.

Roháček, K., Barták, M. (1999). Technique of the modulated chlorophyll fluorescence: basic concepts, useful parameters, and some applications. Photosynthetica, 37, 339–363.

Schreiber, U., Schliwa, U., Bilger, W. (1986). Continuous recording of photochemical and non-photochemical chlorophyll fluorescence quenching with a new type of modulation fluorometer. Photosynth. Res., 10, 51–56.

Sikder, S., Foulkes, J., West, H., De Silva, J., Gaju, O., Greenland, A., Howell, P. (2015). Evaluation of photosynthetic potential of wheat genotypes under drought condition. Photosynthetica, 53, 47–54. DOI: 10.1007/s11099-015-0082-9.

Subrahmanyam, D., Subash, N., Haris, A., Sikka, A.K. (2006). Influence of water stress on leaf photosynthetic characteristics in wheat cultivars differing in their susceptibility to drought. Photosynthetica, 44, 125–129.

Xu, C., Leskovar, D.I. (2015). Effect of *A. nodosum* extracts on spinach growth, physiology and nutrition value under drought stress. Sci. Hort., 183, 39–47. DOI: https://doi.org/10.1016/j.scienta.2014.12.004.

Zavaleta-Mancera, H.A., Lopez-Delgado, H., Loza-Tavera, H., Mora-Herrera, M., Trevilla-Garcia, C., Vargas-Suarez, M., Ougham, H. (2007). Cytokinin promotes catalase and ascorbate peroxidase activities and preserves the chloroplast integrity during dark-senescence. J. Plant Physiol., 164:1572–1582. DOI: 10.1016/j.jplph.2007.02.003

Zlatev, Z., Yordanov, I.T. (2004). Effects of soil drought on photosynthesis and chlorophyll fluorescence in bean plants. Bulg. J. Plant Physiol., 30, 3–18.