Adv. Hort. Sci., 2021 35(2): 129-137 DOI: 10.36253/ahsc-8354

AHS
Advances in Horticultural Science

Effect of biostimulants and media compositions on growth and yield of *Capsicum annuum* L. under drought stress conditions

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Key words: chili pepper, drought stress, growth stimulant, organic farming, sustainable agriculture, *Trichoderma*.

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Citation:
ICHWAN B., ELIYANTI E., ZULKARNAIN Z., 2021 - *Effect of biostimulants and media compositions on growth and yield of Capsicum annuum L. under under drought stress conditions.* - Adv. Hort. Sci., 35(2): 129-137

Data Availability Statement:
All relevant data are within the paper and its Supporting Information files.

Competing Interests:
The authors declare no competing interests.

Received for publication 12 March 2020
Accepted for publication 19 March 2021

Abstract: The study was conducted at the Teaching and Research Farm of Agricultural Faculty, University of Jambi, Indonesia, from April through to September 2019. The aim of this study was to investigate the effect of biostimulants and media compositions on the growth and yield of chili pepper during restricted soil water content. The study was arranged in a split plot design with 3 replicates (groups). Different types of biostimulants (Citorin®, Hantu®, and a control) were designated as main plot, whereas media compositions (2:2:1, 2:1:1, 1:2:1 and 1:1:2) made of soil+trichocompost+rice husk charcoal were employed as sub plot. At the time of transplanting, soil water content was set to approximately 75% of field capacity to create stress conditions. The results showed that the proper choice of biostimulant and medium composition could increase nutrient status, total sugar and chlorophyll contents, and reduce proline level in plants grown under restricted water availability. Citorin® application on chili plants grown on organic media (soil+trichocompost+rice husk charcoal) with ratio of 2:1:1 could be recommended to support plant growth and production under drought stress conditions.

1. Introduction

Chili pepper (*Capsicum annuum* L.) is one of important vegetable crops in Indonesia which is cultivated almost throughout the country. The demand of this commodity has significantly increased, but the production is not yet able to meet the requirement. Current average yield of chili pepper is about 8.47 ton ha^{-1}, and this was far lower than its potential yield that may reach 15-20 ton ha^{-1} (Statistics Indonesia, 2019). Therefore, efforts should be done to promote chili pepper production through the improvement of cultivation techniques and expansion of planting area.

The improvement of chili production through expansion of planting area both in dry and rainy season is restricted by uncertain condition of growing environment, resulting in poor plant growth and development.

Chili pepper growing during dry season is constrained by limited soil water availability, causing environmental stress to the plants. Study by Ichwan *et al.* (2017) revealed that chili pepper grown under 50% field capacity showed slow growth, reduced yield, high proline content, and low sugar content.

Efforts to improve the ability of chili plants to survive drought stress and to be able to grow and result in maximum yield can be done by applying effective biostimulants to plants grown under a mixture of soil, trichocompost and rice husk charcoal. The effect of biostimulant in enhancing plant growth and production had been reported by many authors (López-Bucio *et al.*, 2015; Rady and Ur Rehman, 2016; Niyokuri *et al.*, 2018; Drobek *et al.*, 2019). Also, the use of trichocompost or *Trichoderma*-enriched compost in supporting plant growth and development under limited soil water content had been studied with positive results (Bae *et al.*, 2009; Mastouri *et al.*, 2010; Khoshmanzar *et al.*, 2019).

The use of organic materials may increase plant tolerance to drought stress due to their ability to improve soil structure, increase aeration in root zone, reduce mass density, increase cation exchange capacity, and maintain primary nutrients such as N and P, in addition to increasing soil water holding capacity. Trichocompost (*Trichoderma* in compost) is one of organic materials that can be used to improve plant resistance to drought stress. In addition, the application of husk charcoal in growing media is another way to enhance soil organic matter and improve chemical, physical and biological properties of soil. Husk charcoal contains 0.32% N, 15% PO, 31% KO, 0.95% Ca, 180 ppm Fe, 80 ppm Mn, 14.1 ppm Zn and pH 6.8. Another characteristic of husk charcoal is light (specific gravity of 0.2 kg L^{-1}). Rice husk charcoal also increased field capacity and water content by increasing the porosity of the amended soil, and reduced the acidity of soil (Mishra *et al.*, 2017).

Rice husk charcoal treatments on pot grown lettuce (*Lactuca sativa*) and cabbage (*Brassica chinensis*) were found to increase the final biomass, root biomass, plant height and number of leaves in all the cropping cycles in comparison to charcoal free treatments (Carter *et al.*, 2013). Mannan and Shashi (2019) reported that the application of rice husk charcoal increased plant height, days to maturity, total dry weight, cob diameter, cob length, 100-grain weight, and yield of maize under drought conditions. Chlorophyll content was also found to increase, but proline content decreased due to rice husk charcoal application. Meanwhile, Imanda and Ketty (2018) claimed that rice husk charcoal along with cow manure was best composition for the growth of papaya plant. In chili pepper, however, study on the use of mixture of soil, trichocompost and rice husk charcoal, particularly under drought stress, is not yet well documented.

The application of biostimulant along with the improvement of growing media is expected to be synergistically improve the resistance of chili pepper against drought stress while sustaining best growth and yield. The purpose of this study was to obtain a proper combination of biostimulant and growing media compositions that enhance the growth and yield of chili pepper during drought stress.

2. Materials and Methods

Experimental design and plant handling

The study was conducted at the Teaching and Research Farm, Faculty of Agriculture, University of Jambi from April 2019 through to September 2019. A split plot design with three replications was employed in this study. The main plot consisted of different types of commercial mixed biostimulants (Citorin® and Hantu®) and a control, while the subplot consisted of different ratio of soil+trichocompost+rice husk charcoal (2:2:1, 2:1:1, 1:2:1, and 1:1:2). The biostimulant Citorin® contained gibberellic acid, P_2O_5, K_2O, MgO, Mn, antioxidant and vitamins (Amanah and Putra, 2018), and Hantu® contained gibberellic acid, indoleacetic acid, kinetin, zeatin, N, P, Na, Mg, Cu, Fe, Mn, Zn, Co, Cd, and Pb (Lidar and Mutryarny, 2017).

Seeds of chili pepper cv. Lado were sown on media consisted of soil+trichocompost+rice husk charcoal (2:1:1) on a seedbed. Seven days later seedlings were transferred to nursery, and left for 21 days before transplanting on individual pots with different media compositions according to the treatment. Soil water content in the media was set to 75% of field capacity to create drought condition, except those controls. This is in accordance with our previous investigation (Ichwan *et al.*, 2017).

Biostimulants were applied to the plants on weekly basis from week 2nd to week 12th after transplanting by foliar spraying. Fertilization and maintenance of plants were carried out in accordance with the standard of chili pepper cultivation (Zulkarnain, 2013).

Variables observed

Data on plant growth and yield were collected 10 weeks after transplanting (the time of fruit formation) and 14 weeks after transplanting (the time of first harvest). Variables observed were plant height, number of productive branches, total leaf area, dry weight (total and above-ground parts), fruit number, and total weight of fruits per plant.

Chemical analysis

Data on N, P, K^+, Ca^{2+} and Mg^{2+} content in plant tissues were also recorded as well as total sugar, chlorophyll content, and proline content within leaf tissues. Composite leaf samples were made by physically mixing individual leaves taken from 3 sample plants of 3 replicates into one homogenous sample. Compositing reduced the number of analyses to be performed and was designed to provide a representative sample of the treatment. Ten youngest mature leaves on main stem were collected at 10 weeks after transplanting (WAT). Dry and clean leaf samples were placed in a sample bag prior to laboratory analysis.

Total nitrogen content was determined by Kjeldahl method (Labconco Corporation, 1998), phosphorus concentration was determined by vanadium molybdate yellow colorimetric method (Karlberg and Pacey, 1989), whereas K^+, Ca^{2+} and Mg^{2+} were analysed using Atomic Absorption Spectrometry (AAS) method (The Perkin-Elmer Corporation, 1996). Total sugar was determined according to Irigoyen *et al.* (1992), chlorophyll content was determined according to Hall and Rao (1986), and proline content was analysed according to Bates *et al.* (1973).

Statistical analysis

Data were analysed statistically using Analysis of Variance (ANOVA) module (Petersen, 1985) to judge the significance of the effect of biostimulants and growing media compositions. If the result from ANOVA is significant ($p<0.05$), then the Fisher's Least Significant Different (FLSD) is applied to see the difference between two treatment means. Any difference larger than FLSD is considered a significant result.

3. Results

There was no significant effect of biostimulants on plant height at 10 weeks after transplanting (WAT). However, significant response was shown by plants grown on different media compositions. In the absence of biostimulant, the composition of 2:1:1, 1:2:1 and 1:1:2 were found to enhance plant height. When Citorin® was used as plant biostimulant, the medium composition of 1:1:2 was the best. However, with the use of biostimulant Hantu® none of medium composition showed significant effect on plant height (Table 1).

Table 1 - The effect of biostimulants on the height of chili pepper (cm) grown on different media compositions with limited soil water content (10 WAT)

Biostimulants	Plant height			
	Media compositions [z]			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	88.00 b	105.33 a	106.67 a	102.33 a
Citorin®	98.00 b	114.33 ab	99.00 b	118.00 a
Hantu®	102.00 a	108.00 a	95.33 a	104.67 a

[z] Media compositions= soil : trichocompost : rice husk charcoal. Numbers followed by the same lowercase in the rows are not significantly different according to Fisher's Least Significant Different test (α = 0.05).

The interaction between biostimulants and media compositions showed significant effect on the number of productive branches at 10 WAT (Table 2). The application of Citorin® on plant grown on medium composition of 1:1:2 resulted in the highest number of productive branches, followed by those grown on 2:2:1 medium composition, and they differed significantly from those grown on 1:2:1 medium composition. While in the use of Hantu®, medium composition of 2:2:1 was the best and the effect was significantly different from 1:2:1 and 1:1:2 compositions, but not 2:1:1 composition. In the absence of biostimulant, however, there was no significant difference in the number of productive branches on plants grown on different media compositions (Table 2).

Table 2 - The effect of biostimulants on the number of productive branches of chili pepper grown on different media compositions with limited soil water content (10 WAT)

Biostimulants	Number of productive branches			
	Media compositions [z]			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	88.00 aA	94.00 aA	90.66 aA	71.33 aB
Citorin®	106.66 aA	96.00 abA	78.66 bA	121.33
Hantu®	112.66 aA	100.00 abA	83.00 bA	79.00 bB

[z] Media compositions= soil : trichocompost : rice husk charcoal. Numbers followed by the same lowercase in the rows are not significantly different according to Fisher's Least Significant Different test (α = 0.05).

At 14 WAT, the number of productive branches was not affected by biostimulant application, but the composition of the media was found to affect this parameter significantly (Table 3). Hantu® was effective in promoting the number of productive branches on plants grown on medium composition of 2:1:1, 1:1:2 and 2:2:1, which were significantly different from those grown on 1:2:1 medium composition. With Citorin® application as well as in the absence of biostimulant, the effect of media compositions did not show any difference on the number of productive branches (Table 3).

Table 3 - The effect of biostimulants on the number of productive branches of chili pepper grown on different media compositions with limited soil water content (14 WAT)

Biostimulants	Number of productive branches			
	Media compositions (z)			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	161.00 a	176.67 a	129.00 a	170.67 a
Citorin ®	195.67 a	211.00 a	135.33 a	186.67 a

(z) Media compositions= soil : trichocompost : rice husk charcoal. Numbers followed by the same lowercase in the rows are not significantly different according to Fisher's Least Significant Different test (α = 0.05).

The interaction between biostimulants and media compositions did not significantly affect total leaf area and total dry weight (Tables 4 and 5). The largest absolute total leaf area was found in the combination of Hantu® and 2:1:1 medium composition (2,644.66 cm^2) (Table 4). In addition, the greatest total dry weight was obtained in the combination of Citorin® and 1:1:2 medium composition (Table 5).

Significant effect of the interaction between biostimulants and media compositions was noted on dry weight of above-ground parts (Table 6), fruit number (Table 7) and fruit weight (Table 8). Data presented in Table 6 show that the dry weight of above-ground parts of plants treated with Citorin® on medium composition of 1:1:2 was significantly higher than those without biostimulant. In the absence of biostimulant, media composition of 2:1:1 and 1:2:1 were better than 2:2:1 or 1:1:2, while in the application of Citorin®, the composition of 1:1:2 and 2:1:1 were better than 2:2:1 or 1:2:1. However, when Hantu® was applied there was significant difference in the dry weight of above-ground parts of chili peppers grown on all media compositions (Table 6).

Table 4 - The effect of biostimulants on total leaf area of chili pepper (cm^2) grown on different media compositions with limited soil water content

Biostimulants	Total leaf area (cm^2)			
	Media compositions			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	1497.46	1930.62	1827.07	1784.26
Citorin®	2115.81	2285.40	1531.72	2361.31
Hantu®	1997.24	2644.66	1556.42	2413.54

(z) Media compositions= soil : trichocompost : rice husk charcoal.

Table 5 - The effect of biostimulants on total dry weight of chili pepper (g) grown under drought stress condition on different media compositions with limited soil water content

Biostimulants	Total dry weight			
	Media compositions (z)			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	10.11	13.68	13.34	10.41
Citorin®	11.38	14.31	11.51	16.44
Hantu®	13.58	15.38	12.48	13.04

(z) Media compositions= soil : trichocompost : rice husk charcoal.

Table 6 - The effect of biostimulants on dry weight of above-ground parts of chili pepper (g) grown on different media compositions with limited soil water content

Biostimulants	Dry weight			
	Media compositions (z)			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	4.73 bA	7.13 aA	6.93 aA	4.83 bB
Citorin®	5.03 bA	7.86 aA	5.93 bA	8.23 aA
Hantu®	6.60 aA	7.86 aA	6.46 aA	6.50 aAB

(z) Media compositions= soil : trichocompost : rice husk charcoal. Numbers followed by the same lowercase in the rows and the same uppercase in the columns are not significantly different according to Fisher's Least Significant Different test (α = 0.05).

Data on fruit number presented in Table 7 show that plants grown on media with composition of 2:2:1 and treated with biostimulants produce more fruits than those without biostimulants. On 2:1:1 media composition, the application of Hantu® resulted in the greatest number of fruit, which was significantly different from either the application of Citorin® or no biostimulant treatment. On 1:1:2 media composition, however, the effect of Hantu® was significantly different to those plants grown in the absence of biostimulant only. On media composition of 1:2:1, the control treatment produced more fruits than those plants treated with Citorin® (Table 7).

Table 7 - The effect of biostimulants on the number of fruit of chili pepper grown on different media compositions with limited soil water content

Biostimulants	Fruit number Media compositions (z)			
	2:02:01	2:01:01	1:02:01	1:01:02
No Biostimulant	25.00 bB	38.66 aB	41.66 aA	33.00 abB
Citorin®	42.00 aA	45.33 aB	24.66 bB	40.33 aAB
Hantu®	54.00 bA	73.66 aA	29.66 cAB	61.33 abA

(z) Media compositions= soil : trichocompost : rice husk charcoal.
Numbers followed by the same lowercase in the rows and the same uppercase in the columns are not significantly different according to Fisher's Least Significant Different test (α = 0.05).

Table 8 - The effect of biostimulants on the weight of fruit of chili pepper (g) grown on different media compositions with limited soil water content

Biostimulants	Fruit weight Media compositions (z)			
	2:02:01	2:01:01	1:02:01	1:01:02
No	54.89 cB	96.89 aA	81.08 abA	67.61 bcA
Citorin®	99.03 abA	107.75 aA	31.37 cB	76.77 bA
Hantu®	67.58 bB	106.95 aA	28.70 cB	71.47 bA

(z) Media compositions= soil : trichocompost : rice husk charcoal.
Numbers followed by the same lowercase in the rows and the same uppercase in the columns are not significantly different according to Fisher's Least Significant Different test (α = 0.05).

Data presented in Table 8 show that biostimulant Citorin® significantly increased fruit weight when applied on plants grown on medium with 2:2:1 composition. On medium with 1:2:1 composition, the application of either Citorin® or Hantu® was found to result in less fruit weight significantly. In the absence of biostimulant, the composition of 2:1:1 significantly increased fruit weight than 2:2:1 and 1:1:2 media compositions. Also, in the application of Citorin®, the fruit weight of plants grown on medium with the composition of 2:1:1 was significantly heavier than of those grown on medium composition of 1:2:1. Further, in the application of Hantu® the fruit weight of plants grown on the media composition of 2:1:1 was significantly heavier than the weight fruits produced by plants grown on any other media compositions (Table 8).

The use of biostimulants and growing media containing soil+trichocompost+rice husk charcoal was able to maintain nutrient status within plant tissues to remain at optimal level during limited soil water availability, except for potassium which was at luxury consumption level and calcium which was below critical limit. Nutrient content of chili leaf tissues was measured when the plant was 14 weeks old after transplanting. The results of leaf tissue nutrient measurement is presented in Table 9.

Plants grown on different media compositions and treated with both Citorin® and Hantu® showed a higher total sugar and chlorophyll contents and lower proline level compared to those without biostimulant application (control). In spite of media compositions, plants treated with Citorin® show a higher total sugar and lower proline content in comparison to those

Table 9 - Leaf nutrient contents of chili pepper as affected by biostimulants and different media compositions during limited soil water availability

Biostimulants	Media (z) compositions	Leaf tissue nutrient content (%) (y)				
		N	P	K^+	Ca^{2+}	Mg^{2+}
No biostimulant	2:02:01	2.46	0.31	7.22	0.88	0.56
	2:01:01	1.94	0.37	6.22	1.18	0.73
	1:02:01	2.55	0.32	6.53	1.02	1.05
	1:01:02	3.01	0.26	6.13	1.03	1.02
Citorin®	2:02:01	3.21	0.33	4.37	1.01	0.86
	2:01:01	2.83	0.31	5.99	1.12	0.89
	1:02:01	3.12	1.34	8.24	0.85	0.44
	1:01:02	3.11	0.30	7.03	0.85	0.99
Hantu®	2:02:01	3.44	0.29	7.78	1.05	0.72
	2:01:01	3.65	0.31	5.83	1.08	0.61
	1:02:01	3.14	0.34	7.47	0.65	0.48
	1:01:02	3.02	0.30	7.67	0.86	0.93

(z) Media compositions= soil : trichocompost : rice husk charcoal.
(y) Leaf nutrient content was determined compositely by physically mixing individual leaves taken from each 3 sample plants of 3 repli-

treated with Hantu®. However, plants grown on medium composition of 1:1:2 and treated with Hantu® produced the highest chlorophyll content.

Total sugar, proline and chlorophyll content of leaves were measured when the plants were 14 weeks after planting, and the results are presented in Table 10.

4. Discussion and Conclusions

The application of biostimulants on chili pepper grown on different growing media compositions with limited soil water content was found to increased plant growth and yield. Our results proved that biostimulant application was important for chili pepper grown on media consisting of soil+trichocompost+rice husk charcoal but with limited water supply. Plants treated with biostimulants grew better than those without biostimulant on all growing media. Plants treated with either Citorin® or Hantu® and grown on soil+trichocompost+rice husk charcoal with ratio of 2:1:1 were taller and bigger than those grown on other media compositions without biostimulant application (Fig. 1 and Fig. 2). This increased in pepper growth and yield is presumably due to hormones, organic acids, and macro and micro nutrients contained within the biostimulants. Citorin® contains gibberellic acid (GA_3) along with nutrients such as P, K, Mg, Mn, antioxidants and vitamins (Amanah and Putra, 2018). Meanwhile, Hantu® contains the gibberellic acids (GA_3, GA_5, GA_7), IAA, kinetin and zeatin along with nutrients such as N, P, Na, Mg, Cu, Fe, Mn, Zn, Co, Cd, and Pb (Lidar and Mutryarny, 2017).

Hedden and Thomas (2012) claimed that GA physiologically acted as growth stimulant of plant organs through cell division and elongation. Further, Gupta and Chakrabarty (2013) suggested that gibberellic

Fig. 1 - The effect of biostimulants on the growth of chili pepper during limited water supply (A = no biostimulant; B = Citorin®; C = Hantu®).

Table 10 - Total sugar, proline and chlorophyll contents as affected by biostimulants and different media compositions during limited soil water availability

Biostimulants	Media (z) compositions	Total sugar (y) (mg g^{-1})	Proline (y) (mM g^{-1})	Chlorophyll (y) (cm^2 mL^{-1})		
				a	b	Total
No biostimulant	2:02:01	3.045	0.925	6.096	9.523	15.619
	2:01:01	3.136	0.592	6.688	10.175	16.863
	1:02:01	2.091	1.048	7.386	11.380	18.766
	1:01:02	3.091	0.304	7.527	10.695	18.222
Citorin®	2:02:01	3.136	0.439	7.731	11.723	19.455
	2:01:01	4.841	0.254	8.366	11.325	19.691
	1:02:01	2.614	0.921	5.252	7.109	12.361
	1:01:02	3.909	0.347	7.585	10.568	18.154
Hantu®	2:02:01	4.091	1.209	7.430	9.888	17.318
	2:01:01	3.727	0.803	6.681	9.217	15.899
	1:02:01	3.864	1.141	3.723	5.076	8.799
	1:01:02	2.455	0.566	8.695	12.011	20.706

(z) Media compositions= soil : trichocompost : rice husk charcoal.

(y) Total sugar, proline and chlorophyll contents was determined compositely by physically mixing individual leaves taken from each 3 sample plants of 3 replicates into one homogenous sample at 10 weeks after transplanting.

acids in plants played an important role in triggering the transition from meristem to shoot growth until mature organs. Various recent studies on the use of gibberellic acids indicated that these plant hormones could increase plant growth and development, improved yield, and increased tolerance to abiotic stresses such as drought, heat and salinity (Pal *et al.*, 2016; Sarwar *et al.*, 2017; Miceli *et al.*, 2019; Zhu *et al.*, 2019).

Fig. 2 - The effect of the ratio of soil+trichocompost+rice husk charcoal on the growth of chili pepper during limited water supply (A = 2:1:1; B = 2:2:1; C = 1:2:1; D = 1:1:2).

Growing media is one of important elements supporting plant growth and development. A good media should have good aeration, be able to hold water, and capable to store nutrients for plants. The mixture of soil+trichocompost+rice husk charcoal with the ratio of 2:1:1 or 1:1:2 produced better plant growth compared to others. The greatest plant height, total leaf area and dry weight of aboveground parts were achieved on plants grown on media with the ratio of 2:1:1 on all biostimulant applications, except Citorin® on 1:1:2 medium composition. Meanwhile, the highest number of productive branches (aged 10 and 14 WAT) was obtained on medium composition of 2:1:1 in all biostimulant applications. Based on these results it can be seen that media with more soil or more rice husk charcoal were preferable to produce better growth of chili pepper treated with biostimulants.

Trichocompost is *Trichoderma*-based fertilizer that function to enhance plant's drought tolerance by improving root development (Shukla *et al.*, 2012), activating antioxidant protection to prevent damage caused by dehydration (Brotman *et al.*, 2013), and delaying changes in stomatal opening, photosynthesis and chlorophyll content due to drought (López-Bucio *et al.*, 2015). *Trichoderma* sp. help plants better resist environmental stress such as drought via reinforcing plant growth and reprograming gene expression in roots and shoots. The tolerance to water deficit was attributed to activation of antioxidant responses and higher activity of ascorbate and glutathione-recycling enzymes (Mastouri *et al.*, 2012). The fungal mycelium secreted different compounds that increase the branching capacity of the root system, thus improving nutrient and water acquisition (López-Bucio *et al.*, 2015).

Good growth performance of chili pepper grown on soil+trichocompost+rice husk charcoal and sprayed with biostimulants was followed by good production in term of fruit number and weight. These result was the consequence of a significant interaction of the two factors. Moreover, plants grown in medium with ratio of 2:1:1 and sprayed with Citorin® produced higher total sugar and chlorophyll content and lower in proline compared to those grown on the same medium but in the absence of biostimulant, as well as plants grown on other media ratios but treated with Hantu®.

Total sugar content in chili pepper grown on soil+trichocompost+rice husk charcoal with ratio of 2:1:1 and treated with biostimulants was higher than those grown on other media but in the absence of biostimulant. The results of this study are in line with study conducted by Martim *et al.* (2009) on grapevines which showed that drought stress could increase respiration rate of plants. Increased respiration rate will lower plant carbohydrates and promote total sugar content which also function as an osmotic adjustment.

Chloroplast contains chlorophyll which is a major component involving in photosynthesis. Decrease in chlorophyll content during drought was an indication of oxidative stress caused by photo-oxidative pigment and chlorophyll degradation (Farooq *et al.*, 2009; Anjum *et al.*, 2011). The increase of chlorophyll content in chili peppers grown on different ratios of soil+trichocompost+rice husk charcoal indicates that the plants were able to survive drought stress condition. The application of biostimulants may thus improve plant physio-biochemical attributes under drought stress. This is in accordance with the results noted on *Triticum aestivum* and *Solanum lycopersicum* (Yasmeen *et al.*, 2013) and *Phaseolus vulgaris* (Rady and Mohamed, 2015; Elzaawely *et al.*, 2017). El-Mageed *et al.* (2017) claimed that the improvement of chlorophyll content due to biostimulant application under drought stress may be attributed to the protection impacts on the photosynthetic systems.

Proline is one of dissolved compounds produced by plants in drought stress condition, which acts as an osmotic adjustment in addition to other compounds such as fructan, trehalose, polyol, polyamine and glycinbetain (Mitra, 2001). As an osmotic adjustment, proline keeps plants to continue to grow even in a low water potential condition. Low proline content in plants grown on media ratio of 2:1:1 and treated with Citorin® indicates that they do not experience stress due to drought. This is in accordance with report by Goñi *et al.* (2018) on tomato grown on limited soil water content and treated with *Ascophyllum nodosum* extract which showed a lower leaf proline content in comparison to untreated plants.

Biostimulants containing bioactive compounds are desirable in today's agriculture because of their capability to enhance nutrient uptake which positively affect overall plant vigor resulting in high quantity and quality of harvest (Parađiković *et al.*, 2017). In our study biostimulant application on chili pepper grown on soil+trichocompost+rice husk charcoal could improve growth, increase nutrient status as well as total sugar and chlorophyll contents, and reduce proline level in leaves. In addition, plant height, number of productive branches, total leaf area, and dry weight of above-ground parts were higher in biostimulant-treated plants. Biostimulant Citorin® might be used to ensure the production of chili pepper by overcoming drought stress and providing good nutrient uptake on medium consists of soil+trichocompost+rice husk charcoal with ratio of 2:1:1.

Further works would be necessary to study the application different concentrations of Citorin® on plants grown on 2:1:1 media composition to find out their effects on the yield.

Acknowledgements

We are very grateful His Magnificence Rector of the University of Jambi in Jambi, Indonesia for the financial support of this study through the Competitive Internal Research Grant under the Professor Acceleration Research Scheme 2019.

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