

INFLUENCE OF BRASSINOLIDES ON PLANT PHYSIOLOGY AND YIELD OF CANTALOUPE UNDER HIGH TEMPERATURE STRESS

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ABSTRACT

Cantaloupe is a high value crop growing in greenhouse. Economical crop yield reduction due to heat stress, caused by global warming is an emerging issue of cantaloupe. High temperature significantly decreases the plant performances and finally effect on the crop yield. Brassinosteroids (BRs) regulates cellular and physiological processes and environment response to abiotic stress of plants. To find out the best concentration of BRs for alleviate the high temperature stress of cantaloupe, plant physio-chemical behaviors and yield responses were evaluated after the application of four concentrations of exogenous BRs (24-epibrassinolide, EBR) to cantaloupe plants grown under 47 ± 3 °C. Chlorophyll a, chlorophyll b, total chlorophyll content, rates of photosynthesis and transpiration, stomatal conductance, intercellular CO₂ concentration, proline and malondialdehyde contents, electrolyte leakage, catalase and peroxidase activity, fruit position, fruit yield, total soluble solid content and fruit firmness were significantly differed according to the type of cultivar and EBR concentration. EBR concentration at 0.1 - 0.2 mg L⁻¹ given significant results in order to reduce the impacts of high temperature under greenhouse environment. Higher EBR concentrations than 0.2 mg L⁻¹ recorded negative effect on evaluated physiological and biochemical characters. Furthermore, EBR enhanced the early fruiting of cantaloupe.

Key words: 24-epibrassinolide, fruit position, heat stress, malondialdehyde, proline

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تأثير البراسينوليد على فيزيولوجيا النبات وإنتاجية الشمام تحت ضغط درجات الحرارة العالية

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المستخلص

الشمام هو محصول عالي القيمة ينمو في البيوت المحمية. يعد التخفيض الاقتصادي في غلة المحاصيل بسبب الإجهاد الحراري الناجم عن الاحتباس الحراري من قضايا الشمام الناشئة. تؤدي درجة الحرارة المرتفعة إلى انخفاض معنوي في أداء النبات وتؤثر في النهاية على إنتاجية المحصول. البراسينوستيرويدات (BRs) تنظم العمليات الخلوية والفسولوجية والاستجابة البيئية للإجهاد اللاأحيائي للنباتات. لمعرفة أفضل تركيز من BRs للتخفيف من إجهاد درجة الحرارة المرتفع للشمام ، تم تقييم السلوكيات الفيزيائية والكيميائية للنبات واستجابات الغلة بعد تطبيق أربعة تركيزات من BRs الخارجية (24-epibrassinolide، EBR) لنباتات الشمام التي تزرع تحت 47 ± 3 درجات مئوية. الكلوروفيل أ ، الكلوروفيل ب ، محتوى الكلوروفيل الكلي ، معدلات التمثيل الضوئي والنتح ، التوصيل الشعري ، تركيز ثاني أكسيد الكربون بين الخلايا ، محتويات البرولين ومالونديالدهيد ، تسرب المنحل بالكهرباء ، نشاط الكاتلاز والبيروكسيداز ، موضع الفاكهة ، محصول الفاكهة ، إجمالي المحتوى الصلب الذائب وثبات الفاكهة. تختلف اختلافاً كبيراً وفقاً لنوع الصنف وتركيز EBR. تركيز EBR عند 0.1 - 0.2 مجم لتر - 1 مع إعطاء نتائج مهمة من أجل تقليل تأثيرات درجات الحرارة المرتفعة تحت بيئة الدفيئة. سجلت تركيزات EBR الأعلى من 0.2 مجم L⁻¹ تأثيراً سلبياً على الصفات الفسولوجية والكيميائية الحيوية المقيمة. علاوة على ذلك ، عزز EBR الإثمار المبكر للشمام.

الكلمات المفتاحية 24-epibrassinolide، موضع الفاكهة، الإجهاد الحراري، malondialdehyde، البرولين

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INTRODUCTION

Cantaloupe (*Cucumis melo* var. *cantaloupensis*) is a popular economical greenhouse fruit crop (56, 6). Even though it is a warm climate crop, high environmental temperatures due to global warming and extreme temperature conditions effect on crop production (18, 41). High greenhouse temperature disturbs the balance between photosynthesis and respiration. When the temperatures increase beyond the optimum, the physiological activities of plants negatively affected followed by proteins include enzymes inactivation. Maximum temperature limits for melon crop identified as 39°C - 45°C (28). It was reported that cantaloupe can tolerate in higher temperature than 45 °C. Heat stress due to high temperature throughout the cropping season decline biomass production, plant physiological and biochemical processes and finally, crop yield (39, 17, 3). Reactive oxygen species (ROS) produce when plants opened to heat stress conditions (5, 15). When production of ROS exceeded, lipid peroxidation in biological membranes occurs and decompose polyunsaturated fatty acid. This generates Malondialdehyde (MDA) (9, 44, 29). Proline is an osmolyte and it act in protective mechanism for oxidative stress (40). Proline buffer cellular redox potential and adjust osmotic potential. Proline protect Photosystem II and the chlorophyll from high MDA content (40, 48). Heat stress alters the structures of plant cell membrane proteins by increasing the kinetic energy of molecules and losing the chemical bonds, which leads to increase of loss of electrolytes. This expressed by the value of electrolyte leakage (EL) (22, 8). Hormones are important elements in the process of plant tolerance development for heat stress (23). Brassinosteroids (BRs) are important natural plant hormones. 24-epibrassinolide (EBR) is an important naturally occurring derivative of BRs (23, 7). BRs regulate plant physiological processes such as cell division, cell differentiation, cell wall metabolism, plant organ development, vegetative and reproductive development, photosynthesis, ethylene biosynthesis, gene expression and response to temperature stress (45, 50). Exogenous foliar spray of BRs will increase the heat tolerance capacity of plants

in future sustainable crop production, towards global warming (10, 20) through improving the proline and antioxidant enzymes i.e. peroxidase (POD) and catalase (CAT) activities, while significantly decreased the peroxidation of membrane lipids or MDA content (33, 49). It has been found that exogenous BRs increase ethylene production and increase femaleness of cucumber (49). According to the observations on tomato, EBR pretreatments reduced the inhibition of photosynthesis by high temperature (35). EBR can effectively alleviated the down regulation of PSII activity under high growth temperatures by increasing activity of the ROS hunting system and increasing carboxylation efficiency (1). BRs can act in very small concentrations on plants (42). Exogenous application of BRs can be harmful if exceed a certain concentration level (46). In this previous experiment, it was identified that some cantaloupe cultivars can tolerate in tropical greenhouse at 47±3 °C, with yield reduction than the ambient temperature (3). There were no reported data on BRs application on cantaloupe beyond the 45 °C temperature. This experiment was conducted to investigate the plant physiological and biochemical responses of cantaloupe for different EBR concentrations under extreme high temperature. And also, to find out most efficient EBR concentration for foliar application to enhance the stress tolerance of cantaloupe grown in tropical greenhouse. Findings from this study with the foliar application of EBR on plants in high temperatures, will be a good changing fact for safe and economic agricultural practices towards global warming.

MATERIALS AND METHODS

Experimental site, Planting Materials and Crop Establishment: Cantaloupe cultivars, Himalai-99 and Glamour, which identified as most tolerant cultivars in our previous experiment were used in this study. Seeds of four cantaloupe cultivars were from commercial sources. This research study was conducted in greenhouse at Faculty of Agriculture, Universiti Putra Malaysia (2.9917 °N, 101.7163 °E). Temperature controlled by exhaust and axial fans. Data on atmospheric temperature, humidity and CO₂ in every 5

minutes were monitored by data logger. The plants were fertigated five times per day (0800, 1030, 1200, 1430 and 1700 hours) using Dosatron fertigation system (Model: D25RE2, 0.2 – 2%, 2.5 m³/h, USA). Modified Cooper formulation was used as the fertilizer source. Seeds were sown in nursery trays filled with decomposed coco-peat and maintained in shade house. 10 days old seedlings were transplanted into 16 ×16 cm sized polybags which contained coco-peat. Plant main stem was pruned at 6 feet height and only one fruit per plant was maintained by pruning other side branches after first fruit set. Crop was maintained in maximum 47 ± 3 °C day-time temperature. 24-Epibrassinoloid was dissolved in 1% ethanol and then brought into distilled water (55). Four concentrations of 24-epibrassinolide (EBR); 0, 0.1, 0.2 and 0.3 mg L⁻¹ applied foliar on 15 days after transplant (DAT), 50ml per plant. RCBD design was used with four replicates with 2 sub samples. Data were recorded on 3rd mature leaf at 7 days after treatment. Fruits were harvested at the commercial harvesting stage of each cultivar (60 - 70 DAT). The Total Soluble Solid (TSS) were measured for each sample of fruit in three replications using digital refractometer (Atago Co. Ltd., Japan). Firmness was measured by texture analyzer.

Physiological measurements: Chlorophyll Content was determined according to the method described by Coombs et.al. (1986) with use of four leaf-discs of 4 cm², collected from the fully opened third young leaf. Chlorophyll was extracted with use of acetone. After pigments totally absorbed in to the acetone solution, absorbance was measured at 647nm and 664nm with use of spectrophotometer (UV-3101PC UV-VIS-NI, Shimdsu, Japan). Chlorophyll content was calculated with use of following formulas.

$$\text{Chlorophyll a} = 13.19 (A_{664}) - 2.57 (A_{647})$$

$$\text{Chlorophyll b} = 22.1 (A_{647}) - 5.26 (A_{664})$$

$$\text{Total chlorophyll content} = 3.5 (\text{chl a} + \text{chl b})/4$$

A₆₄₇ and A₆₆₄ = absorbance of the solution at 647nm and 664nm respectively
13.19, 2.57, 22.1 and 5.26 = absorption coefficients

4.5 = volume of solution taken from the original solution for the analysis (ml)

$$4 = \text{total leaf disc area (cm}^2\text{)}$$

Leaf gas exchange; photosynthesis rate (PR), stomatal conductance (SC), intercellular CO₂ concentration (C_i) and transpiration rate (TR), were measured by using portable photosynthesis system (Li 6400, Li-Cor, USA). Data obtained between 0830-1030 hours of the day and repeated in three growth stages.

Biochemical measurements: Analysis of MDA was done according to the method described by Madhava Rao and Sresty (41). 1g of fresh leaf was homogenized with 0.1% trichloroacetic acid (TCA) and homogenate was centrifuged for 10 min at 15,000 rpm. 0.5ml of supernatant was added to thiobarbituric acid + 20% TCA and kept in water bath at 90 °C for 20 minutes. Reaction was stopped in an ice bath and centrifuged for 5 min at 10,000 rpm. Absorbance of supernatant was measured at 532 nm and 600 nm. MDA concentration was calculated by using extinction coefficient of 155 mM⁻¹cm⁻¹.

Proline content of third fully mature leaf was determined according to Bates et al., 1973, (19). The 0.5g leaf samples were homogenized in 10 cm³ of aqueous solution of 3% sulphosalicylic acid and centrifuge at 1500ppm for 10 min. Then supernatant, acid ninhydrin and glacial acetic acid added 1:1:1 and incubated in water bath for 1 h at 100°C and reaction was concluded in an ice bath. Then add 4 cm³ of toluene and mixed vigorously. Proline content was measured by a spectrophotometer at 520 nm using toluene as a blank and calculated as micrograms per gram FW against standard proline. Nine young leaf discs, 1cm² each from 3rd matured leaf were used to measure EL according to Ahammed et al. (2). Leaf discs were washed 3 times with deionized water to remove surface- adhered electrolytes and they were placed in closed vials containing 10 mL of deionized water. Samples were incubated at 25°C on a rotary shaker. After 24 hours, electrical conductivity (EC1) of the solutions were recorded. Then samples were autoclaved at 120°C for 20 minutes. After cooled to 25°C, final electrical conductivity (EC2) was measured and EL was calculated as follows
$$\text{EL} = (\text{EC1} / \text{EC2}) \times 100$$

Enzyme extraction for POD and CAT analysis was done according to the Poli et al. (38). 0.1 g of leaf sample was mixed with 5 ml extraction buffer containing 0.1M phosphate buffer (pH 7.5) containing 0.5 mM EDTA and 1% (w/v) of polyvinylpyrrolidone (PVP). The enzyme extract was centrifuged for 20 min at 15000 g at 4°C. The supernatant was collected and stored until assay for enzyme activity. POD assay was done according to Mardinata et al. (32). For assay, a 3 ml of reaction mixture consist of 2.83 mL of 0.1 M sodium phosphate buffer (pH 7.0), 28 µL of guaiacol, 30 µL of 30% H₂O₂ and 100 µL of enzyme extract was used. The increase in absorbance at 420 nm was measured for 2 min. The POD activity was measured and expressed as mmol of H₂O₂ reduced per minute per gram FW.

The CAT activity was assayed according to Poli et al., (38) by measuring the ability of the enzyme extract to decompose H₂O₂. The reaction mixture of 3 ml consisted of 0.05 ml extract, 1.5 ml phosphate buffer (100 mM buffer, pH 7.0), 0.5 ml H₂O₂, and 0.95 ml distilled water. A decrease in the absorbance was recorded at 240 nm for 2 minutes. The CAT activity was expressed as mmol of H₂O₂.

Leaf nutrient content

The third mature leaves were harvested and dried 72 hours in 70°C with use of forced oven. Then, 0.25 g of sample mixed with 5 ml of concentrated sulfuric acid (H₂SO₄) in digestion tubes and kept for 24 hours for digestion. Then, 2 ml of 50% H₂O₂ was added and heated at 285°C for 45 minutes in block digester. The mixture was left to cool and added of 2 ml H₂O₂ followed by heating in block digester. Cooling tubes, adding H₂O₂ and heating for 45 minutes were repeated until totally transparent solution developed. The final samples were diluted with distilled water and volumed up to 100 ml. Then filtered using Whatman No. 4 filter paper before the concentration was analyzed. The concentration of K, Ca and Mg were measured using Atomic Absorption Analyzer (AAS) and calculated the each element %.

Statistical analysis

Analysis of variance (ANOVA) was done using statistical software R-studio 1.4.1106. The least significant difference (LSD) at 5% significant level was used for mean separation

and Ls-means was used where the interaction effect was significant. For analyze the correlation between parameters, Pearson Correlation method was used.

RESULTS AND DISCUSSION

Maximum temperature was controlled at 50 °C. Minimum and maximum atmospheric [CO₂] was recorded as 429 and 499 ppm at 1400 and 0700 time of the day.

Chlorophyll content and leaf gas exchange

EBR significantly reduced the leaf **Chl-a** and **Chl-total** contents and treatments had a significant interact effect (Table 1). Chl-a reduced by 16% and Chl- total reduced by 11% at 0.1 mg/L EBR. **Chl-b** didn't have either significant in interaction effect or cultivar effect. Compare to the control, Chl-b was increased by 6-8% at 0.1mg/L and 1% at 0.2mg/L, but not significantly differ with control. Chlorophyll is vastly sensitive to stress conditions which decrease total chlorophyll a, b, and carotenoid contents in leaves. BRs protect the chlorophyll and increase chlorophyll content under different stresses conditions (32, 4). Data relevant to PR, SC, Ci and TR presented in Table 1, had a significant effect by the treatments and significant interaction between the treatments. Compare to the control, application of 0.1 mg/L EBR had increased the PR, SC and TR by 9-13% and 0.2 and 0.3 mg/L made reduction of the PR by 10-7% and 33% respectively and significantly. Each cultivar not significantly differ between control and 0.1 mg/L, 0.1 and 0.2 mg/L. At 0.3 mg/L, Ci was increased by 3% in each cultivar. 0.1mg/L EBR BRs exhibit multiple actions on essential processes of leaf gas exchange. The high temperature inhibited the photosynthesis rate due to photosynthetic apparatus disruption. BRs regulate plant growth and development and alleviate high temperature stress (14). Application of EBR increase quantum yield of PSII photochemistry, electron transport rate and PR and it was observed in soybean (37). *Cucumis sativus* at 27 °C to three leaf stage plants 0.1 mg/L EBR foliar application recorded most effective for stimulated PR. 0.1mg/L EBR recorded a higher quantum yield of PSII electron transport due to a significant increase in the photochemical quenching. Particularly the capacity of CO₂ assimilation in

the Calvin cycle which were mainly attributed to an increase in the initial activity of Rubisco in photosynthesis mechanism (52). Microscopic analyses showed that the

application of EBR maintains the typical shape of the chloroplasts and promotes the formation of grana in the stressed cucurbit plants (53).

Table 1. Interaction effect of EBR on chlorophyll-a, total chlorophyll content and leaf gas exchange and effect of EBR on chlorophyll-b of each cultivar.

Factors	Chlorophyll content (mg cm ⁻²)			Leaf gas exchange			
	Chl-a	Chl-b	Chl-total	PR ($\mu\text{Mm}^{-2}\text{s}^{-1}$)	SC ($\text{mMm}^{-2}\text{s}^{-1}$)	Ci ($\mu\text{Mm}^{-2}\text{s}^{-1}$)	TR ($\text{mMm}^{-2}\text{s}^{-1}$)
Himalai -99							
0	4.64a	1.86a	4.64a	28.6c	1.42c	305cd	14.e
0.1	4.01c	1.98a	4.01b	31.4b	1.56a	302d	16.4c
0.2	3.92d	1.88a	3.92d	25.8e	1.23d	305cd	17.8a
0.3	3.04g	1.70b	3.04g	19.6f	0.82e	316a	12.1f
Glamour							
0	4.65a	1.85b	5.68a	28.7c	1.55ab	306c	15.6d
0.1	4.14b	2.00a	5.36b	32.6a	1.63a	307bc	17.0b
0.2	3.86e	1.96a	5.10e	26.8d	1.45bc	310b	16.1c
0.3	3.75f	1.75c	4.81f	19.2f	0.81e	315a	11.6g

Note: Chl – Chlorophyll, PR - photosynthesis rate, SC - stomatal conductance, Ci - Intercellular carbon dioxide concentration, TR - transpiration rate

Correlation analysis

Ch-a and Ch-b significantly correlate by 97 and 79% to the total chlorophyll content and by 57 and 72% to PR (Table 2). Total chlorophyll content 66% correlate to the PR. Data reveals that PR highly, positively and significantly correlate with the SC and TR. TR highly correlate with PR, SC and Ci. Ci negatively correlated with all the compared analyzed parameters. In the present study, EBR accompanied by the increasing PR, SC

and high TR. The TR was correlated with the photosynthesis (1), which could be due to the carbon dioxide availability. PR significantly correlated with SC, TR and Ci. PR extrovertly correlate with Ci and in EBR-treated plants, when PR increases, TR and SC increases and Ci decreased. It was likely due to 0.1 - 0.2 mg/L EBR make open stomata. The significance of this correlation depend on the type of the plant and type of the BRs (11).

Table 2. Pearson correlation between chlorophyll content and leaf gas exchange

	PR	Chl-a	Ch-b	Ch-tot	SC	Ci
Chl-a	0.67**	-				
Chl-b	0.77**	0.38*	-			
Chl-tot	0.77**	0.98**	0.56**	-		
SC	0.96**	0.7**	0.78**	0.8**	-	
Ci	-0.82**	-0.62**	-0.68**	-0.71**	-0.80**	-
TR	0.8*	0.44*	0.76**	0.56**	0.81**	-0.78**

Note: Chl – Chlorophyll, PR - photosynthesis rate, SC - stomatal conductance, Ci - Intercellular carbon dioxide concentration, TR - transpiration rate

Biochemical changes

Data relevant to MDA, proline, EL and CAT given a significant interaction effect between cultivars and EBR concentrations. POD activity expressed a significant effect by the EBR concentration and significant difference between cultivars, where no interaction effect (Table 3). Compare to the control, application of 0.1 mg/L EBR increase the proline content

by 35 and 27% in Himalai-99 and Glamour. At 0.2mg/L, increment was 51 and 121%. MDA content was decreased by 34-26% at 0.1 mg/L and 32-12% at 0.2 mg/L. But increased by 33-17% at 0.3mg/L which means plants get stressed at higher EBR concentrations. EBR reduced the ROS and increase MDA in heat stressed plants (37, 41) The decrease in MDA content after EBR treatment is an indicator for

EBR induced efficient scavenging of ROS (25). Reduction of MDA content with temperature and EBR treatment interaction was observed by Jin et al. (24), in fig plants at 40 °C compared to 28 °C. Thussagunpanit et al. (46), recorded that EBR treatment significantly reduced the MDA content of rice at 40 °C and it facilitated by protective mechanism of EBR on chlorophyll content and oxidative stress. Compare to the control, application of 0.1 mg/L EBR increase the proline content by 35 and 27% in Himalai-99 and Glamour. At 0.2mg/L, increment was 51 and 121%. Compared to the control, maximum reduction of EL for each cultivar (43% in Himalai-99 and 63% in Glamour), was recorded at 0.2mg/L. Same effective concentration of EBR (0.2 mg/L) for reduction of MDA and EL in abiotic stress was observed by Li and Feng (31), on 1 year old sorbifolia seedlings and Dou et al. (12) on 2 weeks old cotton seedlings. Khan et al. (26), recorded significant reduction of EL in application of EBR on both thermo-tolerant and thermo-sensitive cultivars of okra. EBR act on a membrane in deferent ways; (i) influence on the fatty acid composition, (ii.) modify cell

membrane properties (iii) improve the cell membrane functions under hostile environmental temperatures, (iv) subpress the excess ROS and reduce the damage of constituent membrane elements, (v) defends cell membranes by neutralize free radicals leads to reduce lipid peroxidation (28). Compared to the control, POD of Himalai-99 increased by 12, 31 and 20 % at 0.1, 0.2 and 0.3 mg/L EBR. In Glamour, it was 2, 10 and 20%. POD activity in control was comparatively lower in Himalai-99 and it may be because it is more heat resistant. Yun-ying et al. (54), reported that EBR increase the POD activity and also improve the expression of POD isozymes by inducing new native bands of isozymes in rice seedlings which protect the plants from heat stress. Compared to the control, CAT activity was increased by 60 – 47% at 0.1 mg/L and it was 34-24% at 0.2mg/L. In heat stressed environment, EBR significantly and markedly increase the proline, CAT and POD to protect the cell functions from ROS (42, 51).

Table 3. Effect of EBR on plant biochemical changes

Cultivars in EBR mg/L	MDA (µmol/g FW)	Proline (µmol/g FW)	EL (%)	POD (µg g ⁻¹ FW min ⁻¹)	CAT (U mg ⁻¹ Protein)
Himalai-99					
0	1.07 ^c	8.31 ^{de}	61.8 ^b	12.30 ^b	7.57 ^f
0.1	0.71 ^f	11.23 ^{bcd}	40.0 ^d	13.73 ^{ab}	12.09 ^b
0.2	0.73 ^f	12.51 ^{ab}	35.3 ^e	16.08 ^a	10.16 ^d
0.3	1.42 ^a	9.01 ^{cde}	36.7 ^e	14.69 ^{ab}	6.20 ^g
Glamour					
0	1.10 ^c	6.68 ^e	67.9 ^a	14.70 ^b	9.04 ^e
0.1	0.81 ^e	8.46 ^{de}	35.1 ^e	15.01 ^{ab}	13.25 ^a
0.2	0.97 ^d	14.80 ^a	25.3 ^e	16.19 ^a	11.16 ^c
0.3	1.28 ^b	11.74 ^{bc}	48.0 ^c	16.14 ^a	6.20 ^g

Note: MDA - Malondialdehyde, EL - electrolyte leakage, POD – peroxidase content, CAT – catalase activity

Leaf nutrient content

Leaf calcium and potassium contents were significantly affected by EBR application and not either significantly differ between cultivars or interaction effect (Table 4). Compared to the control, leaf calcium content was 81% higher in F3 and 55% higher in F4. Leaf potassium content was increased with application of EBR at 0.2 and 0.3 mg/L and at

F3, increment was 31%. EBR application was significantly affected on Mg content and there was a significant difference between cultivars. But there was no interaction effect on leaf Mg content. K, Ca and Mg are three essential minerals of cantaloupe to develop the membrane structure and membrane functions of fruits and postharvest quality improvement (30). Giri et al. (16) observed the reduction of

root nutrition absorption in elevated temperature in tomato. They and Hellal (21) highlighted that moderate heat stress has no

significant effect on nutrient absorption rate where severe heat stress significantly decreased the root nutrient absorption rate.

Table 4. Leaf nutrient content

Factor	Leaf nutrient content (%)		
	Ca	K	Mg
Cultivar			
Himalai-99	0.230a	0.497a	0.081a
Glamour	0.209a	0.451a	0.073b
	ns	ns	*
EBR concentration mg/L			
0	0.154d	0.427bc	0.065c
0.1	0.183c	0.417c	0.068bc
0.2	0.281a	0.557a	0.097a
0.3	0.239b	0.496ab	0.077b
	*	*	*

Notes: Ca – Calcium, K – Potassium, Mg – Magnesium

Fruiting node in main stem

Fruiting node in high temperature with application of EBR had a significant effect from EBR concentration and observed a significant interaction effect (Figure 1). In our previous experiment (3) , we observed an increment of fruiting node in high temperature than the ambient temperature by around two folds. Compare to the control, in the application of EBR, earliest fruit set or the lowest node number of Himalai-99 recorded at

0.1mg/L and it was 38% reduction. In Glamour, lowest node number was recorded at 0.2mg/L and it was 64% reduction. EBR increase cucurbit sex expression and decrease the time to appear female flower. On the other hand, EBR increase Ethelene production and early fruiting and fertilization believed facilitated by Ethelene (36). EBR increase fruit set of cucurbits and after anthesis, EBR activating ovary cell division and duplication (13).

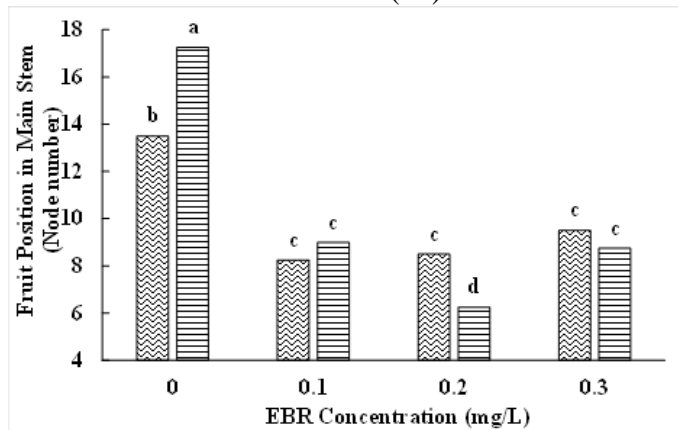


Figure 1. Interaction effect of EBR on fruit position in main stem

Fruit yield and quality

Data grafted in Figure 2 had a significant effect from EBR concentration, significant difference between cultivars and significant interaction effect between treatments. Compare to the control, maximum yield increment was observed at 0.1 mg/L by 29-22%. At 0.2 and 0.3 mg/L, increments were

23-14 and 10-12%. This yield response depend on the developmental stage of EBR application and EBR concentration (34). Foliar application of EBR increase crop yield by facilitating/increasing the crop growth, photosynthesis, early fruiting, biomass partitioning and fruit growth rate (11, 47).

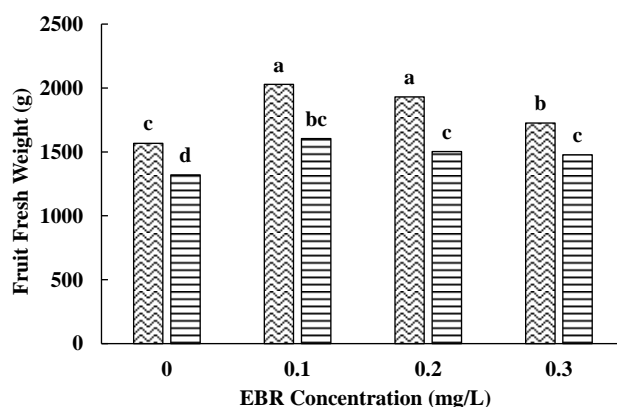
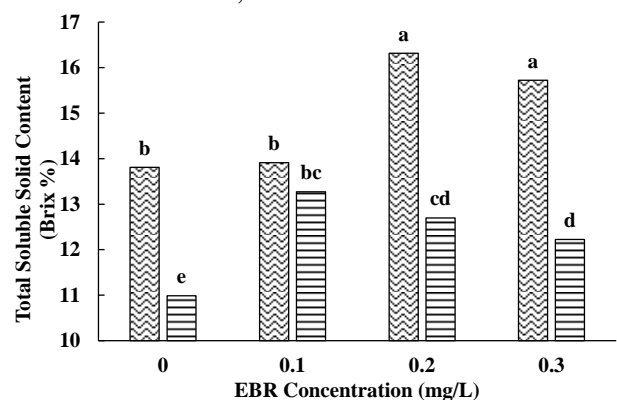


Figure 2. Interaction effect of EBR on fruit fresh weight

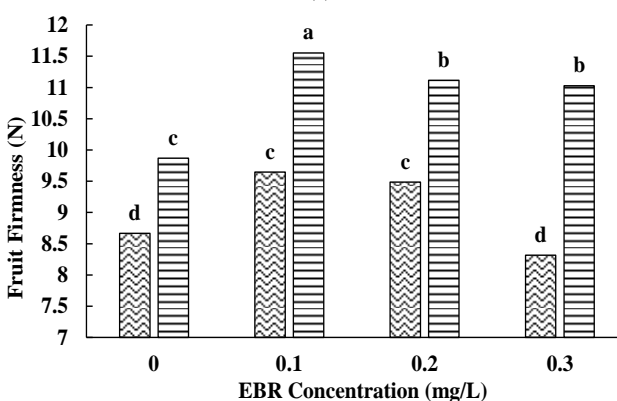
TSS content and firmness values of the fruits were grafted in Figure 3. Brix value of Himalai-99 was significantly higher than the Glamour. In Himalai-99, highest brix value was observed at 0.2mg/L and it was significantly not differed with 0.3 mg/L.

Both cultivars recorded their maximum firmness at 0.1mg/L EBR. In Himalai-99, at

0.1 and 0.2 mg/L firmness was not significantly differed. Fruit firmness is a direct measurement of fruit maturity and fruit quality. EBR application to stressed plants has been increased the plant performances and fruit quality too.



(i)



(ii)

Figure 3. Interaction effect of EBR on fruit TSS and firmness

CONCLUSION

Exogenous application of BRs successfully reduces the plant stress and enhance the cantaloupe plant physiological and biochemical functions to increase the production in elevated temperature in modern

farming. EBR concentration of 0.1-0.2 mg/L is effective to increase plant growth, flowering, fruit setting, leaf gas exchange, antioxidant enzymes, proline content and reduce the malondialdehyde content and electrolyte leakage. Higher EBR concentrations than

0.2mg/L cause complications in all evaluated physiological and biochemical characters. EBR enhanced early fruiting by facilitating either early female flowering or reducing flower abortion or increase fertility or all.

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