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Interactive effect of elevated carbondioxide and high temperature on quality of hot chilli (*Capsicum chinense* Jacq.)

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Abstract: *The study was conducted during 2012-15 in Carbondioxide Temperature Gradient Tunnels (CTGTs) and in ambient condition to assess the interaction effect of elevated CO_2 and temperature in two cultivars of hot chilli (*Capsicum chinense *Jacq.). The treatments consisted of field (ambient CO_2 and ambient temperature), CTGT I (380 ppm CO_2 and ambient temperature), CTGT II (550 ppm CO_2 with 2^0 C elevation of ambient temperature) and CTGT III (750 ppm CO_2 with 4^0 C elevation of ambient temperature). The temperature elevation was maintained from flower bud initiation to maturity stage. The results revealed that CTGT II had greater impact on quality parameters* viz. *fruit diameter AND capsaicin content. On the other hand at further higher CO_2 and temperature in CTGT III, most of the quality parameters showed a declining trend indicating the deleterious effect of higher CO_2 and high temperature. However, some degree of tolerance were exhibited by cv. Manipur compared to cv. Assam under elevated carbondioxide and temperature conditions. The study therefore, indicates differential responses of* Capsicum chinense *Jacq. genotypes under different microclimatic conditions.*

INTRODUCTION

Global warming has been largely derived from anthropogenic CO_2 emission, which have raised atmospheric CO_2 from pre-industrial levels of 280 ppm to the latest record of 401.57 ppm in October 2016 (Anonymous, 2016). The predicted CO_2 value is between 700 to 1000 ppm by the end of this century (Meehl, *et al.*, 2007). According to ACIA report (2004), a rapid warming is expected to continue in the future, with more than 6 °C warming expected throughout the twenty ûrst century under some scenarios. Elevated temperature is becoming an increasingly significant abiotic stress in the scenario of global climate change, as temperature is predicted to increase more than 5.8°C by the end of this century (Solomon *et al.*, 2007).

Therefore, the present investigation was made to study the interactive effect of elevated CO_2 and high temperature stress on the quality of hot chilli (*Capsicum chinense* Jacq.). This study was done to understand the differential response of two germplasms of *Capsicum chinense* Jacq., commonly cultivated in Assam and Manipur, to the interaction of elevated CO_2 and high temperature stress. Such study may serve as the basis in determining the cultivation of these germplasms to suit in the changing environment. Since the increase in ambient temperature in near future is likely to affect the quality of a plant. Therefore, it is important to investigate the impacts of climate change on some quality parameters of hot chilli.

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MATERIAL AND METHODS

The experiment was conducted with two germplasms of hot chilli (*Capsicum chinense* Jacq.) which were collected from local races of Manipur and Assam. They were used to study the effect of high CO_2 and temperature on the water status and to identify the genotype for suitability under the present scenario of climatic change. The experiment was laid out in Completely Randomized Design (CRD) having 2 factors with three replications. Two races were considered for the study. The experiment included four treatments *viz.* T_1 = Field (Ambient CO_2 condition and temperature), T_2 = CTGT I (380 ppm CO_2 and ambient temperature), T_3 = CTGT II (550 ppm CO_2 and 2^0C higher than ambient temperature from flower bud initiation till maturity) and T_4 = CTGT III (750 ppm CO_2 and 4^0C higher than ambient temperature from flower bud initiation till maturity). The CO_2 was maintained through out the entire crop growth period from 9 a.m. to 2 p.m. regularly. The elevation of temperature was maintained from flower bud initiation to maturity stage for a particular period of time through Infra Red Heater regulated by SCADA software.

Carbondioxide Temperature Gradient Tunnel (CTGT): The controlled environment for the experiment was created in the carbondioxide temperature gradient tunnel (CTGT), of the Department of Crop Physiology, AAU, Jorhat (Make: Genesis Technologies, Maharashtra). Three numbers of CTGTs were used for carrying out the experiments. The structure consisted of a rectangular (10 m length, 2.5 m breadth, 2 m height) block, fabricated by metallic sheet pipe. They were covered with poly carbonate sheet (100 micron gauge) that showed more than 85% transmission of light. The tunnel was divided into four compartments of 2.5 meter in length and each chamber was fitted with 4 RTD temperature sensors (one RTD sensor/compartment). Humidity transmitters were placed in each chamber to get data through four core shielded cable. Infrared heaters were mounted inside the CTGT. For recording the ambient data, temperature sensor and humidity transmitter were placed outside the chamber. The humidity sensors enable measurement of humidity in the range of 0-100% relative humidity. Air compressor was used to maintain uniformity of CO_2 gas at set concentration.

CO_2 and temperature control and monitoring

The system for monitoring and controlling the CO_2 in CTGTs was fully automatic and the desired CO_2 level could be maintained. Data logger, SCADA software and PC were used to monitor and control appropriate CO_2 levels in each chamber. The SCADA software facilitated to set different concentration of CO_2 in different CTGC chamber at a time. Most control actions are performed automatically by Remote Terminal Units ("RTUs") or Data logger. The command to cut and pass CO_2 is given by software to the solenoid valves.

Method of planting and aftercare

Fine soil and organic matter were collected, drenched with 0.1% Captan and covered with polythene sheet. Furadon 3G @ 3g/m^2 was mixed thoroughly and covered for two more days. Fine and sterilized field soil were used to germinate the Bhoot Jolakia (hot chilli) seeds of both the cultivars in the earthen pots (28 cm height and 30 cm in diameter) filled with a mixture of soil and organic matter (50:50). Seeds were put in a container and captan @ 2.5 g/kg seed was added to it. The fungicide was mixed thoroughly with seeds by agitating them for five minutes.

Fine and sterilized field soil were used to germinate the bhoot jolokia seeds of different lines in the earthen pots filled with a mixture of soil and organic matter (50:50). Seedlings of 3-4 leaf stage were transplanted in earthen pots of 40 cm height and diameter which were filled with topsoil, sand and cow dung (1:1:1). The pots were fertilized @ 120: 60: 60:: N: P: K kg/ha. Half of N, full P and full K were applied as basal dose and rest half N was top dressed in 2 split doses at 30 and 60 days after transplanting. The experimental site and pots were kept free from weeds by periodic hand weeding. Irrigation and pesticide application were done as and when required.

Plant sampling: Five pots of each cultivar were kept in each treatment for studying the various parameters. Sampling was done at flowering stage.

Only the physiologically active leaves were used for the study.

Observations recorded

Capsaicin content: The capsaicin content of chilli fruits was determined colorimetrically using the method used by Quagliotti (1971). The absorbance was read at 650 nm using a UV-VIS Spectrascan. The standard graph was prepared using 0-200 µg capsaicin (standard solution) simultaneously. The estimation was done in duplicate and the mean was expressed as percentage of capsaicin in moisture free sample.

$$\text{The formula used for the capsaicin estimation}: \%\,capsaicin = \frac{\mu g\ capsaicin}{200}$$

The capsaicin contents were converted to Schoville Heat Units by multiplying the pepper dry weight content in grams of capsaicin per gram of pepper by the coefficient of heat value for capsaicin; which is 1.6 ´ 10^7 (Todd *et al*., 1977).

Carotenoid content: The carotenoid content was estimated according to Rodrigue Amaya (1999). The absorbance was recorded at 450 nm in a UV-VIS Spectrascan and total carotenoid content was calculated as µg g^{-1} using the following formula.

$$Carotenoid\ content = \frac{Absorbance \times Volume\,(ml) \times 10^4}{Absorbance\ coefficient\,(2592) \times sample\ weight}$$

Fruit diameter

At the time of harvest, five fruits were collected randomly from each pot and diameter was calculated using slide calipers and average was recorded.

RESULTS AND DISCUSSION

A significant difference was recorded in capsaicin content among the cultivars in both the years (Table 1). A maximum (17% and 12%) per cent increase was recorded in cv. Manipur in the first and second year respectively. Elevated CO_2 and temperature brought about a significant difference in capsaicin content in both the years. The increase in capsaicin content ranged from 27% (CTGT II) to 38% (CTGT III) and 36% (CTGT II) to 44% (CTGT III) in the first and second year respectively. Plants under both the CTGTs significantly improved the quality of *Capsicum* in terms of capsaicin content. Temperature stress altered the capsaicin content in the *Capsicum* fruits. The stress induced adverse effect of this parameter was significantly ameliorated under CTGT. In the present study, it clearly indicates the sustenance of stimulatory effect of CO_2, may have helped to maintain the quality of fruits in *Capsicum*. Pungency, the unique characteristics of *Capsicum spp*, is due to the biosynthesis of capsaicinoids and has driven its rapid domestication in different parts of the world (Walsh and Hoot, 2001). Capsaicin, a volatile phenolic amine, is a very stable molecule and is responsible for the pungency commonly associated with chili peppers (Heiser, 1976). A widely used heat measurement of chile peppers is the Scoville Heat Unit (SHU) (Scoville, 1912). This measurement is the highest dilution of a chile pepper extract at which heat can be detected by a taste panel. When some chilies, such as the haberno, are ground into a powder, capsaicin can be detected by taste at dilutions up to 1 ppm. In the present investigation, plants grown at elevated CO_2 and temperature recorded higher capsaicin content. Elevated CO_2 might have contributed to high concentration of carbon based secondary metabolites (capsaicin). Alternatively high temperature may influence the activity of capsaicin synthase the enzyme responsible for capsaicin synthesis. High temperature stress triggered the synthesis of capsaicin in green and ripe fruits. Plants under both the CTGTs significantly improved the capsaicin content of *Capsicum*. The study indicates that stimulatory effect of CO_2 help to maintain the quality of *Capsicum* fruits. Higher capsaicin content was recorded in cv. Manipur compared to cv. Assam in the present experiment. The amount of capsaicinoids in a chile pepper pod is dependent on the genetic makeup of the plant and the environment where it is grown (Zewdie and Bosland, 2000). Plants might have improved carbon use efficiency resulting from increased photosynthesis, light use efficiency and decreased dark respiration at that modified environment.

There was a significant difference in carotenoid content among the cultivars in both the years (Table

2). In cv. Assam a decrease of 11% and 13% over cv. Manipur was noted in the first and second year respectively. Similarly, a significant difference in carotenoid content was noted amongst the treatments in both the years. Stress induced reduction in carotenoid content over ambient ranged from 39% (CTGT II) to 44% (CTGT III), although there was no significant difference among the two treatments in the first year. In the second year, a stress induced reduction in carotenoid content ranged from 37% (CTGT II) to 51% (CTGT III). In the interaction effect of treatments and cultivars, a non significant difference was recorded in the first year. A maximum decrease of 19% and 44% was recorded in cv. Manipur in CTGT II and CTGT III respectively whereas; cv. Assam recorded a maximum per cent decrease of 53% and 56% in CTGT II and CTGT III respectively in the second year.

A reduction in carotenoid content was recorded due to elevated CO_2 and temperature in the present study. Carotenoids not only play a role as accessory light-harvesting pigments but also protect photosynthetic systems as non-enzymatic antioxidant compounds against reactive oxygen species generated during stress. Greater destruction of leaf carotenoids on exposure to higher temperature and greater sunlight has been reported by Young and Britton (1990). Thus at CTGT III may lead to lower carotenoid content by decreased protection of the photosynthetic apparatus. A higher carotenoid accumulation under stress condition was found in cv. Manipur compared to cv. Assam. Similarly, genetical difference in carotenoid accumulation has also been reported by Kopsel (2004).

During the study significant difference in fruit diameter was observed due to the different treatment levels in both the years (Table 3). The per cent increase in fruit diameter over ambient ranged from 33% (CTGT III) to 43% (CTGT II) and 35% (CTGT III) to 39% (CTGT II) in the first and second year respectively. Elevated CO_2 has been found to negate the effect of high temperature on fruit diameter when cultivated in CTGTs. The present study clearly indicates the sustenance of stimulatory effect of CO_2 that may help to maintain the quality

Table 1
Interactive effect of elevated carbon dioxide and high temperature on capsaicin content (SHU in lakhs)

	1st year	*2nd year*	*Pooled*
Treatment			
Field	3.30	3.41	3.35
CTGT-I (Amb)	3.25	3.33	3.29
CTGT-II	4.14	4.50	4.32
CTGT-III	4.49	4.81	4.65
SEd	0.119	0.11	0.155
CD (0.05%)	0.253	0.232	0.358
Cultivar			
cv. 1 (Manipur)	4.10	4.25	4.17
cv. 2 (Assam)	3.49	3.78	3.63
SEd	0.084	0.077	0.11
CD (0.05%)	0.179	0.164	0.253

Treatment x Cultivar	*cv. 1*	*cv. 2*	*cv. 1*	*cv. 2*	*cv. 1*	*cv. 2*
Field	3.51	3.08	3.70	3.13	3.61	3.10
CTGT-I (Amb)	3.45	3.05	3.65	3.00	3.55	3.02
CTGT-II	4.43	3.86	4.54	4.46	4.48	4.16
CTGT-III	5.01	3.98	5.10	4.52	5.06	4.25
SEd	0.169		0.155		0.22	
CD (0.05%)	NS		NS		NS	

Table 2
Interactive effect of elevated carbon dioxide and high temperature on carotenoid content (μg/g)

	1st year	*2nd year*	*Pooled*
Treatment			
Field	12.83	12.16	12.49
CTGT-I (Amb)	13.92	12.91	13.41
CTGT-II	8.53	8.12	8.33
CTGT-III	7.84	6.35	7.09
SEd	0.75	0.75	0.76
CD (0.05%)	1.6	1.6	1.75
Cultivar			
cv. 1 (Manipur)	11.35	10.50	10.93
cv. 2 (Assam)	10.20	9.27	9.73
SEd	0.53	0.53	0.54
CD (0.05%)	1.12	1.12	1.24

Treatment x Cultivar	*cv. 1*	*cv. 2*	*cv. 1*	*cv. 2*	*cv. 1*	*cv. 2*
Field	12.41	13.24	12.90	11.41	12.66	12.33
CTGT-I (Amb)	14.56	13.27	12.31	13.51	13.44	13.39
CTGT-II	9.29	7.91	9.98	6.27	9.57	7.09
CTGT-III	9.15	6.38	6.82	5.87	8.05	6.13
SEd	1.06		1.06		1.07	
CD (0.05%)	NS		2.26		NS	

Table 3
Interactive effect of elevated carbondioxide and high temperature on fruit diameter (cm)

	1st year		*2nd year*		*Pooled*	
Treatment						
Field	2.30		2.23		2.27	
CTGT-I (Amb)	2.22		2.15		2.18	
CTGT-II	3.17		2.98		3.08	
CTGT-III	2.95		2.90		2.93	
SEd	0.105		0.121		0.057	
CD (0.05%)	0.222		0.257		0.131	
Cultivar						
cv. 1 (Manipur)	2.61		2.53		2.57	
cv. 2 (Assam)	2.71		2.61		2.66	
SEd	0.075		0.086		0.04	
CD (0.05%)	NS		NS		NS	
Treatment x Cultivar						
	cv. 1	*cv. 2*	*cv. 1*	*cv. 2*	*cv. 1*	*cv. 2*
Field	2.23	2.37	2.17	2.30	2.20	2.33
CTGT-I (Amb)	2.17	2.27	2.13	2.17	2.15	2.22
CTGT-II	3.17	3.17	2.93	3.03	3.05	3.10
CTGT-III	2.87	3.03	2.87	2.93	2.87	2.98
SEd	0.148		0.171		0.08	
CD (0.05%)	NS		NS		NS	

of *Capsicum* fruits. A differential responses of genotypes has also been noticed in the present study.

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