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Full Length Article

Evaluation of Citrus Rootstocks against Drought, Heat and their Combined Stress Based on Growth and Photosynthetic Pigments

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Abstract

The change in weather patterns around the world has exposed the living world to a variety of stresses, among which heat and drought stress are alarming ones. In citrus, the rootstocks control several morphological, physiological, genetic and production traits of scion cultivars. The current study was conducted to evaluate ten modern citrus rootstocks including two local commercial rootstocks (Rough lemon and Sour orange) against heat and drought stress based on visual assessments, plant growth, dry matter yield, chlorophyll *a*, *b* and carotenoid contents. The level of heat and drought were control, moderate and severe stress. All plants at severe combined heat and drought stresses died within five days. Brazilian sour orange showed no change in leaf color under heat and drought stress and regarded as tolerant rootstock. Sovage citrange and Yuma citrange (trifoliate citrus) were sensitive rootstocks and shed their leaves. Plant growth attributes e.g., plant height and root length higher in Brazilian sour orange and low in Sovage citrange. Brazilian sour orange showed high shoot and root dry matter yield. The contents of chlorophyll *a*, *b* and carotenoids, were high in Brazilian sour orange followed by Keen sour orange but low in Sovage citrange and Yuma citrange. It is concluded that out of ten rootstocks Brazilian sour orange was proved as tolerant and Sovage citrange as the most sensitive rootstock against drought and heat stress. Brazilian sour orange can be used as rootstock for commercial scion cultivars to evaluate growth and productivity in water deficit and high temperature subtropical areas for sustainable citrus industry.

Keywords: *Citrus*; Abiotic stress; Scion; Production; Climate resilience; Visual scores

Introduction

Citrus ranks second after grapes in term of area, production and yield making it the most important fruit crop of Pakistan. Genus *Citrus* comprises more than 10 cultivated species among which sweet orange contribute 60% fresh and processed fruit around the globe (Nawaz *et al.*, 2017). Rootstock in citrus trees influences the morphological, biochemical, physiological and genetic characteristics of grafted scion cultivar through rootstock scion interaction pathway. Tree productivity and fruit/juice quality is also affected by rootstocks. There is always need of new rootstocks with improved tolerance/resistance to abiotic and biotic stresses to sustain the profitability of citriculture. The shift in climatic patterns enforces to look for rootstocks with ability to face certain biotic and abiotic stresses. Rootstocks sensitivity against abiotic stresses (drought, cold and salinity) and biotic stresses such as citrus greening (major disease of entire Florida citrus industry) and other major diseases have been observed (Chung and Brlansky, 2005).

Global climate changes effect water availability and increase in temperature and are responsible for drought and heat stress which alter the plant morphology, physiology, anatomy, and genetic expression of plants (Chaves *et al*., 2009; Nawaz *et al.*, 2013). Heat and drought have negative effect on respiration, photosynthesis, membrane stability, water relations, levels of hormones, primary and secondary metabolites and limit crop growth and productivity around the world (Bhanu *et al*., 2018; The *et al*., 2019).

Plant roots and shoot under water stress send signal to the aerial part of the plant leaves show symptoms due to closure of stomata. Abiotic stresses also increase the level of ABA hormone in root hair tip. Abiotic stresses such as heat and drought limit growth and adversely effect on crop production. Resulting in the abnormal physiological processes that influence one or a combination of biological and environmental factors (Fathi and Tari, 2016).

Drought/water stress reduces plant photosynthetic rate by damaging photosynthetic apparatus and reducing chlorophyll contents (Noctor *et al*., 2002; Zhang *et al.*, 2018). Chlorophyll and carotenoid contents are linked to stress tolerance in plants (Baker and Rosenqvist, 2004; de Matos Nunes *et al*., 2014). Resistant cultivars show elevated chlorophyll contents when exposed to water stress

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(Zaefyzadeh *et al*., 2009). Water/drought stress reduces chlorophyll *a* and *b* contents in wheat (Liu *et al*., 2006) and limits the plant morphology, dry matter yield and production. Drought stress causes a large decline in the chlorophyll *a* content followed by chlorophyll *b* and total chlorophyll contents (Wessjohann *et al*., 2018). Active oxygen species reduce the chlorophyll contents under drought stress due to damage to thylakoid lamella (Nyachiro *et al*., 2001). Severity and duration of drought upregulates the chlorophyll contents within plant body (Kyparissis *et al*., 2007).

Extreme high temperature affects plant photosynthetic functions by altering structural organization and reducing rate of chemical reactions. Changes in the thylakoid membrane alter physicochemical and functional organization due to high temperature (Berry and Bjorkman, 2003). Photosystem-II, the most delicate component of the photosynthetic system reduces photosynthesis (Berry and Bjorkman, 2003). Heat stress alters the function of oxygen evolving system, resulting in the release of functional manganese ions from the complex (Allakhverdiev *et al*., 2008). Rice mutant stay-green characteristic under heat stress had higher carotenoids and chlorophylls than sensitive wild plants (Panigrahy *et al*., 2011). Plants exposed to heat reduce the chlorophyll biosynthesis in leaves (Nguyen *et al*., 2009).

Observed shift in climate reaching both extremes with high degree of unpredictability; water shortage and fluctuating temperature stresses are inevitable, which threaten agriculture and plant productivity around the globe. It is prerequisite for survival of citrus industry to evaluated suitable rootstocks with ability to adjust in changing climate. The objective of present study was the evaluation of citrus rootstocks against drought, heat and their combined effects based on visual changes and photosynthetic pigments.

Materials and Methods

Experiment Details and Treatments

Experimental material: A field experiment was executed during September 2016 to February 2017 at Institute of Horticultural Sciences, University of Agriculture Faisalabad, Pakistan. The plant material included seedlings of ten citrus rootstocks *i.e.,* Brazilian sour orange (*Citrus aurantium*), Sour orange (*C. aurantium*), Chakotra (*C. maxima*), Yuma citrange (*Citroncirus spp*.), Keen sour orange (*C. aurantium* L.), Gadha dahi (*C. aurantium*), Sovage citrange (*C. sinensis* x *Poncirus trifoliata),* Rough lemon (*C. jambhiri*), Bitter sweet orange (*C. sinensis*) and *C. obovoideae*. Rootstocks seeds were sown in greenhouse media containing soil, silt and sand (1:1:1) and germinated in growth chamber at 32±2°C, 16 h of light and relative humidity oscillating between 80–95%. During growth, the pots were regularly irrigated with tap water to keep 75% field capacity and fertilized at weekly interval with nutrient solution [1.0 g L^{-1} $Ca(NO_3)_2$, 0.4 g L^{-1} KNO_3, 0.6 g L^{-1} $MgSO_4$ and 0.4 g L^{-1} $NH_4H_2P0_4$ (MAP)].

Treatments: To evaluate heat and drought stresses, rootstock seedlings were subjected in controlled growth chamber for 15 days at 38°C and 46°C for heat treatment, 50% and 25% field capacity for drought treatment and 50% field capacity at 38°C and 25% field capacity at 46°C for the combined heat and drought stress. For heat treatment temperature was increased just for day time (12 h) and decreased at 32°C for the night. Prior to imposition of stresses for 15 days to citrus rootstocks another group was maintained at 32°C with well-watered plant for control comparison. Thereby, four major experimental groups were established as control, drought stress, heat stress and combined drought and heat stress.

Visual Assessment, Growth Attributes and Dry Matter Yield under Stress Effects

Leaf and plant were visually observed like a leaf necrosis or chlorosis on rootstock seedlings at 5^{th}, 10^{th} and 15^{th} days after stresses treatment. Plant height and root length were measured after uprooting the plants. Plant materials *i.e*., shoot and root parts were harvested after 15 days to stress and dried in an oven at 60°C for 5 days to determine individual shoot and root dry matter yield per plant.

Leaf Photosynthetic Pigments Contents

Leaf sample of 500mg for each treatment was homogenized by adding 80% acetone (v/v) and filtered through filter paper. Spectrophotometer absorbance at 663, 645 and 480 nm was recorded for chlorophyll *a*, chlorophyll *b* and carotenoids, respectively. Chlorophyll *a*, *b* and carotenoids was worked out as under,

$$\text{Chl } a \text{ (mg/g fresh weight)} = [12.7 \text{ (O.D 663)}–2.69 \text{ (O.D 645)}\times V/1000\times W]$$

$$\text{Chl } b \text{ (mg/g fresh weight)} = [22.9 \text{ (O.D 645)}–4.68 \text{ (O.D 663)}\times V/1000\times W]$$

$$\text{Carotenoids (mg/g fresh weight)} = \text{Acar} / \text{Em}\times 100$$

$$\text{Acar} = \text{O.D } 480 + 0.114 \text{ (O.D 663)} – 0.638 \text{ (O.D 645)}$$

Where

V= Volume of sample, W= Weight of fresh tissue, Em = 2500

Experimental Design and Data Analysis

Experiment was laid out according to factorial under RCBD with ten rootstocks, three stresses, three level of each stresses and three replications of each level. One plant per pot was considered as an experimental unit. The mean data were statistically analyzed using SAS-JMP-pro-13 statistical software. The significance of differences between variables at p<0.05 was checked and mean difference was analyzed by Tukey's test. Simple correlation coefficients among photosynthetic pigments and shoot dry matter yield were determined by Statistix 8.1. Linear regression and correlations between shoot dry matter yield and plant visual health index was also determined.

Results

Visual Assessment of Stress Effects

Plant leaf color, leaf margins and growth were observed at 5, 10 and 15 days after stresses. Brazilian sour orange showed no symptoms of chlorosis or leaf margin curling under moderate and severe drought stress but sovage citrange indicated necrosis on leaf blade after 5 days of drought stress (Fig. 1). Brazilian sour orange showed tolerance against moderate heat stress while Sovage citrange emerged as the most sensitive and dropped leaves after 6 days of stress application. All the plants at severe stress of heat burn due to extreme heat (46°C) stress (Fig. 2). Under combined heat and drought stress again Brazilian sour orange was observed as tolerant and sovage citrange as sensitive to moderate combined stress. However, all the plants at severe stress burn due to severe heat and drought stress (Fig. 3).

Plant Growth under Drought, Heat and Combined Stress Effects

Plant height: Drought stress affected significantly on plant height of all rootstocks. Brazilian sour orange was considered as tolerant because of maximum plant height under moderate (14.2 cm) and severe (9.8 cm) drought stress. Chakotra, *C. obovoideae* and Yuma citrange attained minimum plant height under moderate stress. However, under severe drought stress the Sovage citrange was with minimum plant height (5.03 cm) (Fig. 4). Plant height differed significantly among citrus rootstocks, treatments and their interaction. Plant height under control and moderate stresses was high in Brazilian sour orange (17.0 and 15.3 cm, respectively). Sovage citrange proved as sensitive under moderate stress with minimum plant height (Fig. 4). All rootstock plants exposed to severe heat stress (46°C) for continuous 15 days did not survive. Brazilian sour orange (15.0 cm) and Keen sour orange (14.6 cm) showed maximum plant height at moderate combined drought and heat stress. Minimum plant height was recorded in Sovage citrange (8.2 cm) under moderate combined stress. All the rootstock plants did not survive at severe stress of drought plus heat (Fig. 4).

Root length: The interaction of rootstocks and drought stress treatments had significant effect on root length of plants under drought. Brazilian sour orange yielded maximum root length under moderate (21 cm) and severe (18.5 cm) drought stress. Similarly, Sovage citrange showed maximum root length (26 cm) under control. However, the smallest roots under moderate and severe stress were recorded in Sour orange (12 cm) and Sovage citrange (10 cm), respectively (Fig. 5). Heat stress levels significantly affected the root length in all rootstocks. Sovage citrange showed small roots under control (15 cm) and moderate (8.5 cm) stress. However, Brazilian sour orange (17.2 cm) followed by Bitter sweet orange (16.9 cm) produced long

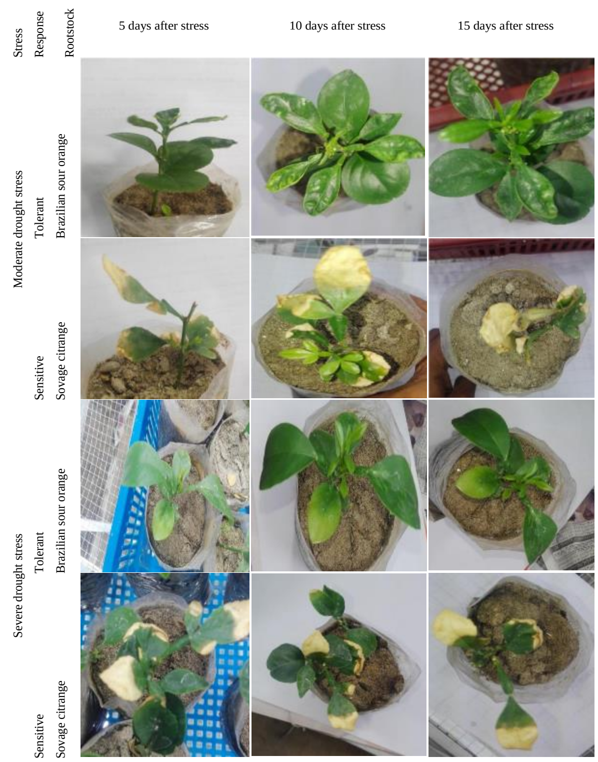

Fig. 1: Visual assessment of Citrus rootstock seedling against drought stress after 5^{th}, 10^{th} and 15^{th} day

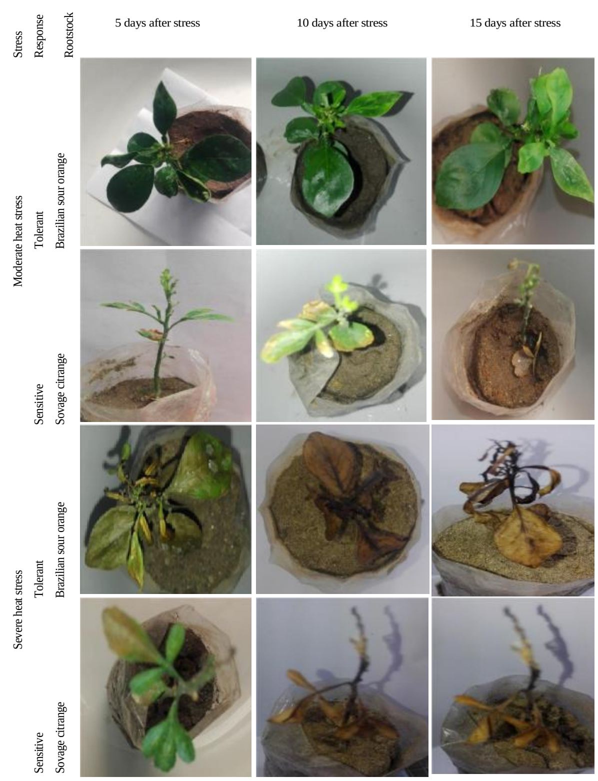

Fig. 2: Visual assessment of Citrus rootstock seedling against heat stress after 5^{th}, 10^{th} and 15^{th} day

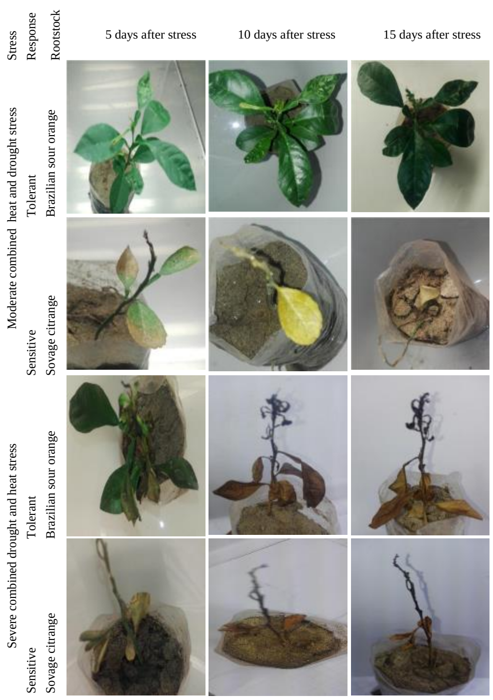

Fig. 3: Visual assessment of Citrus rootstock seedling against combined drought + heat stress after 5th, 10th and 15th day

roots under moderate stress. All the plants at severe stress did not survive at extreme high temperature (46°C) (Fig. 5). Combined heat and drought stress significantly affected the root length in all rootstocks. Under control, maximum root length was observed in Brazilian sour orange (22.4 cm) and minimum in *C. obovoideae* (17.4 cm) which was statistically at par with Sour orange (17.8 cm). Similarly, Brazilian sour orange under moderate stress produced the longest roots (16.9 cm); however, sovage citrange produced the smallest roots (7.9 cm). The combined severe drought and heat stress resulted in plants mortality after 6 days to stress (Fig. 5).

Dry Matter Yield under Drought, Heat and Combined Stress Effects

Shoot dry matter: Bitter sweet orange (1.15 g/plant) followed by Chakotra and Brazilian sour orange (1.13 g/plant) showed high dry matter contents while it was low in Sour orange (0.98 g/plant) under control level. Brazilian sour orange yielded high shoot dry matter per plant under moderate (1.07 g/plant) and severe (0.72 g/plant) stress. Sovage citrange proved sensitive with low dry matter yield under moderate (0.68 g/plant) and severe (0.48 g/plant) drought stress (Fig. 6). Shoot dry matter was significantly affected by heat stress (p<0.05). Brazilian sour orange showed high shoot dry matter yield under control (1.21 g/plant) and moderate (0.93 g/plant) drought stress. Dry

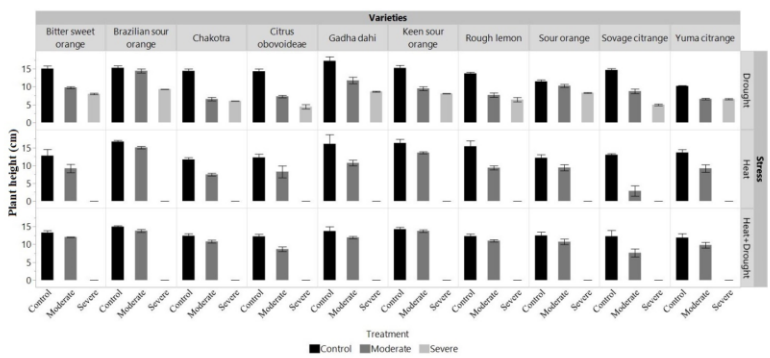

Fig. 4: Effect of drought, heat and combined drought + heat stress on plant height (cm)

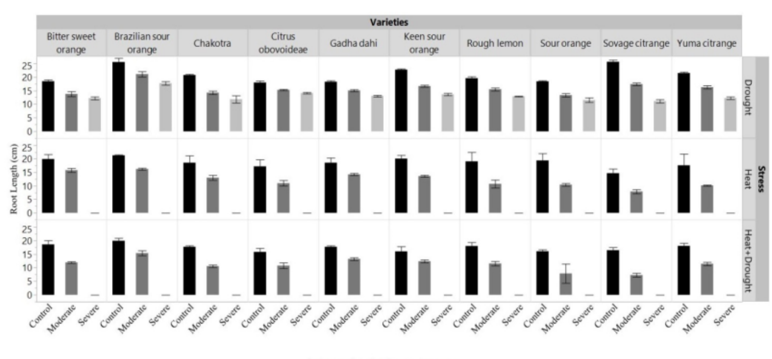

Fig. 5: Effect of drought, heat and combined drought + heat stress on root length (cm)

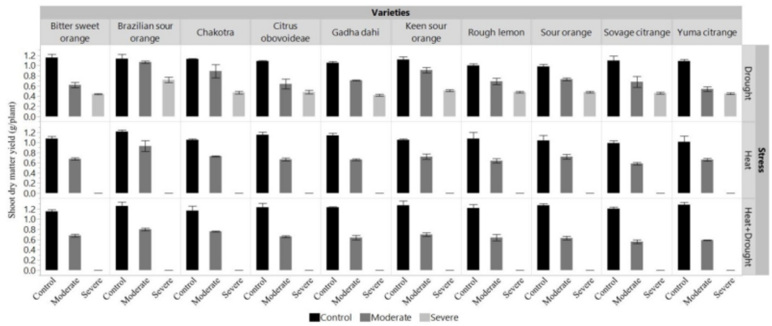

Fig. 6: Effect of drought, heat and combined drought + heat stress on shoot dry matter yield (g/plant)

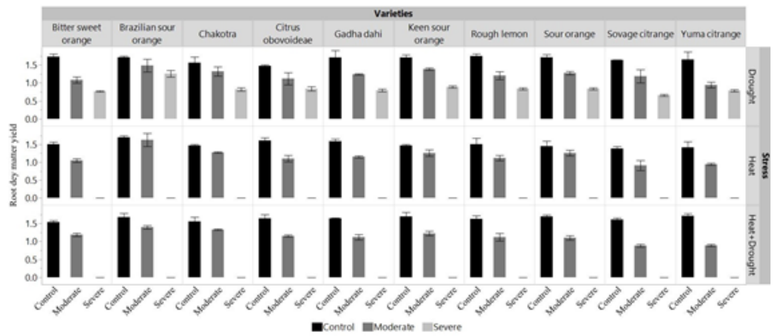

Fig. 7: Effect of drought, heat and combined drought + heat stress on root dry matter yield (g/plant)

matter was low in Sovage citrange under control (0.98 g/plant) and moderate (0.58 g/plant) stress. The combined severe drought and heat stress resulted in plants mortality after 6 days to stress (Fig. 6). High dry matter yield per plant was examined under moderate combined drought and heat stress in Brazilian sour orange (0.80 g/plant) followed by Chakotra (0.78 g/plant); however, low dry matter yield was recorded in Sovage citrange (0.560 g/plant) which was statistically at par with Yuma citrange (0.59 g/plant). All plants at severe combined stress showed mortality after 6 days to stress (Fig. 6).

Root dry matter yield: Brazilian sour orange had high root dry matter yield under moderate (1.49 g/plant) and severe (1.26 g/plant) drought stress. Low root dry matter was observed in Sovage citrange (0.65 g/plant) and Gadha dahi (0.73 g/plant) under severe stress which were statistically similar with each other. Yuma citrange produced low root dry matter yield (0.94 g/plant) under moderate stress (Fig. 7). Heat stress significantly (p<0.05) affected the root dry matter yield. Dry matter yield was high in Brazilian sour orange under control (1.70 g/plant) and moderate (1.63 g/plant) heat stress. Sovage citrange yielded low dry matter under moderate heat stress (0.912 g/plant) which was statistically at par with Yuma citrange (0.945 g/plant). All plants no mortality after 6 days due to severe heat stress (Fig. 7). Combined heat and drought exhibited significant (p<0.05) effect on root dry matter yield. High root dry matter yield was observed under control in Yuma citrange (1.72 g/plant) followed by Sour orange (1.70 g/plant); however, Brazilian sour orange yielded high root dry matter under moderate (1.40 g/plant). Low root dry matter was weighed in Sovage citrange (0.88 g/plant) which was statistically at par with Yuma citrange (0.89 g/plant). All plants at severe combined stress showed no mortality after 6 days to stress (Fig. 7).

Selected stresses affected plants growth, visual health index and dry matter yield of all rootstocks in given order from tolerant to sensitive; Brazilians sour orange> Keen sour orange> Gadha dahi> Bitter sweet orange> Sour orange> Rough lemon> *C. obovoideae*> Chakotra> Yuma citrange> Sovage citrange.

Correlation among Shoot Dry Matter and Visual Health of Plant

Shoot dry matter significantly correlated with visual health of the citrus rootstocks. A strong positive correlation was observed between visual health index and shoot matter yield content on drought, heat and their combined stress (Fig. 8).

Photosynthetic Pigments Contents under Stress Effects

Chlorophyll *a*: Rootstocks, treatments and their interaction differed significantly (p<0.05) based on chlorophyll *a* content under drought stress. Brazilian sour orange under moderate (3.26 mg/g) and severe (2.65 mg/g) stress had high amount of chlorophyll *a*. Gadha dahi yielded low chlorophyll *a* content under control (3.26 mg/g). Keen sour orange also showed high chlorophyll *a* content under

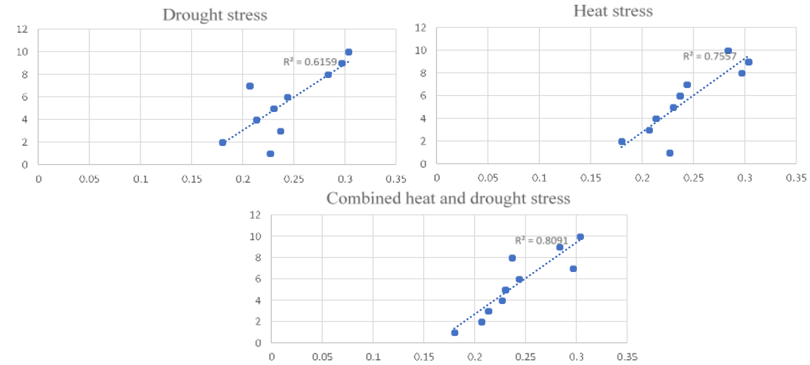

Fig. 8: Correlation of visual plant health and shoot dry matter yield under drought, heat and their combined stress

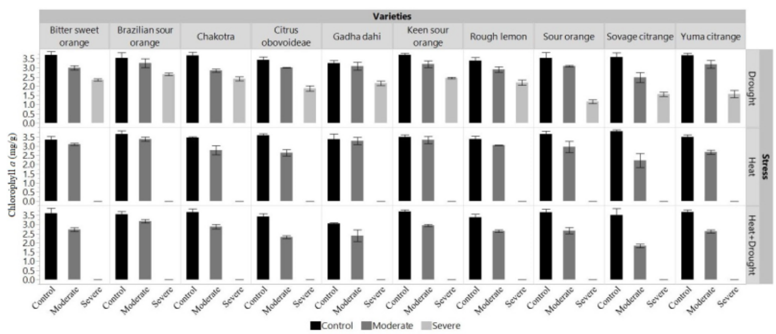

Fig. 9: Effect of drought, heat and combined drought + heat stress on leaf chlorophyll *a* content

control (3.71 mg/g). Sovage citrange was with low chlorophyll *a* content under moderate (2.48 mg/g) and severe (1.57 mg/g) stress (Fig. 9). Under heat stress chlorophyll *a* content varied significantly (p<0.05) in rootstocks. Plants died five days after severe heat stress (46°C). Brazilian sour orange showed high (3.4 mg/g) content under moderate stress followed by Keen sour orange (3.35 mg/g). Sovage citrange showed higher contents under control (3.82 mg/g) condition and lower under moderate (2.23 mg/g) stress (Fig. 9). Combined application of drought and heat stress significantly (p<0.05) affected the chlorophyll contents. Higher chlorophyll *a* content was observed under control condition in Keen sour orange (3.71 mg/g) followed by Yuma citrange and Chakotra (3.68 mg/g). Brazilian sour orange observed higher in contents under moderate (3.18 mg/g) and low in Sovage citrange (1.84 mg/g) (Fig. 9).

Chlorophyll *b* content: Drought stress significantly (p<0.05) affected the citrus rootstock for chlorophyll *b* contents. Brazilian sour orange displayed high amount of chlorophyll *b* contents under moderate (1.18 mg/g) and severe (1.07 mg/g) stress. Yuma citrange indicated high content under control (1.40 mg/g) condition. Sovage citrange yielded low chlorophyll *b* contents on moderate (0.57 mg/g) and severe (0.38 mg/kg) stress (Fig. 10). Chlorophyll *b* differed significantly in rootstocks and treatments interaction. High chlorophyll *b* contents were observed in Brazilian sour orange (1.19 mg/g) under moderate stress and low in Sovage citrange (0.53 mg/g). All the plants at severe stress were burnt due to severe heat stress (Fig. 10). Rootstocks varied significantly (p<0.05) for chlorophyll *b* content under combined (heat+ drought) stress. Under control (Fig. 10) *C. obovoideae* (1.39 mg/g) followed by Bitter sweet orange (1.37 mg/g) yielded high chlorophyll *b* contents (0.138 mg/kg) while under moderate stress Brazilian sour orange produced (1.11 mg/g) chlorophyll *b* contents. However, this was low in Sour orange (0.65 mg/g) under control and Sovage citrange (0.63 mg/g) under moderate stress. Under combined severe heat and drought stress all plants were burnt after 5 days of stress (Fig. 10).

Carotenoids contents: Brazilian sour orange produced the highest carotenoids under moderate (1.23 mg/g) and severe (0.84 mg/g) stress of drought. Yuma citrange (1.39 mg/g) followed by Keen sour orange (1.38 mg/g) produced higher carotenoids under control drought stress. Sovage citrange showed lower carotenoids under moderate (0.69 mg/g) and severe (0.38 mg/g) stress. Chakotra yielded low carotenoids under control (1.14 mg/g) stress (Fig. 11). As regards as heat Brazilian sour orange produced the highest (1.14 mg/g) carotenoid under moderate stress followed by Keen sour orange (1.12 mg/g) and under control higher in Yuma citrange (1.37 mg/g). Sovage citrange revealed lower in carotenoids under moderate (0.604 mg/g) heat stress. All the plants under severe stress did not survive (Fig. 11). Combined application of heat and drought stress significantly affected the carotenoids in selected citrus

Table 1: Correlation among photosynthetic pigments and shoot dry matter yield under drought, heat and their combined stress

Parameters	Stress	Chlorophyll *a*	Shoot Dry matter	Carotenoids
Shoot dry matter	Drought	0.48*		
	Heat	0.4086		
	Combined	0.68*		
Carotenoids	Drought	0.71**	0.69**	
	Heat	0.68*	0.65*	
	Combined	0.9**	0.56	
Chlorophyll *b*	Drought	0.76**	0.34	0.53**
	Heat	0.5292	0.1798	0.4932
	Combined	0.88**	0.69*	0.8*

(*: Significant, **: Highly significant)

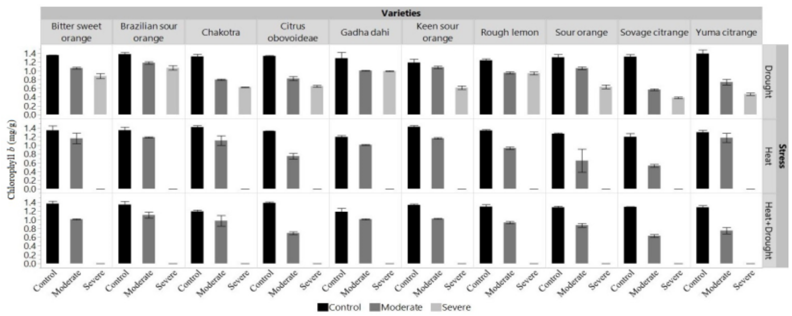

Fig. 10: Effect of drought, heat and combined drought + heat stress on leaf chlorophyll *b* content

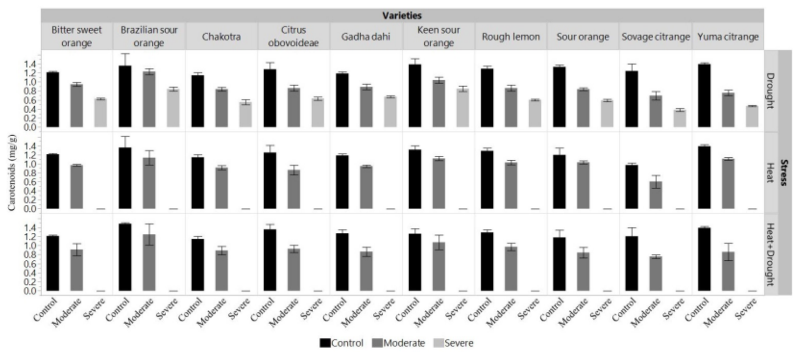

Fig. 11: Effect of drought, heat and combined drought + heat stress on leaf carotenoids content

rootstocks. The rootstock with high carotenoid contents under control (1.48 mg/g) and moderate (1.25 mg/g) stress was Brazilian sour orange. Low carotenoid contents were observed under moderate in Sovage citrange (0.76 mg/g) rootstock (Fig. 11).

Based on the photosynthetic pigments, all rootstocks behaved in the order from tolerant to sensitive; Brazilians sour orange> Keen sour orange> Gadha dahi> Sour orange> Bitter sweet orange> Rough lemon> *C. obovoideae*> Chakotra> Yuma citrange> Sovage citrange.

Correlation among Shoot Dry Mass and Photosynthetic Pigments

Shoot dry matter correlated significantly with chlorophyll *a* at drought and combined stress, with carotenoids at drought and heat stress and with chlorophyll *b* at combined stress only (Table 1). Carotenoids had significant correlation with chlorophyll *a* at all stresses and with chlorophyll *b* at drought and combined stress. Chlorophyll *a* and *b* also correlated significantly at drought and combined stress (Table 1).

Discussion

In this experiment, citrus rootstocks were subjected to water stress, heat stress and combination of water and heat stress, major factors which are affected due to climate change. However, it is predicted that responses of individual or combined stress or single stress to citrus rootstocks could not be inferred. For this reason, there is a need to understand the nature of responses to multiple stresses in order to develop plants more tolerant to environmental cues in a climate change scenario. In this context, drought and heat represent two stress conditions that are expected to have increased incidence in the next 50–100 years, drastically affecting global agricultural systems (Zandalinas *et al*., 2016).

Shoot and root growth of plants is critical in crop production under water-limited and high temperature environments (Ali *et al.*, 2009; Goufo *et al*., 2017; Huang *et al*., 2018). Increase in root and shoot mass and structure improves the photosynthetic rate which enhances the tolerance of plants against stresses (Habben *et al*., 2014). Drought and heat stress inhibit or limit the plant growth and health which overall reduce the plant fresh and dry weight, moisture content and plant dry matter yield. In this study Brazilian sour orange was proved as a tolerant rootstock based on maximum plant height, root length, shoot dry matter yield and root dry matter yield because of better defense against the drought and heat stress. Sovage citrange was regarded as sensitive rootstock under both stresses.

Citrus rootstocks with high chlorophyll *a* and *b*, and carotenoid contents are regarded as tolerant rootstocks (Homayoun *et al*., 2011). Visual assessments indicated the Brazilian sour orange as tolerant rootstock without changes in leaf green color and leaf necrosis; however, Sovage citrange emerged as the most sensitive, with maximum plant death and leaf shedding during stress treatments. Plants having green tissues (chlorophyll) under drought stress are considered as resistant cultivars.

Water is essential for citrus trees (or for any plant) because it is an integral component of the biochemical reactions that occur within the plant. Average water requirement is half litter per plant which is equal to 75% field capacity of plant soil media (Wright, 2000). Deficiency of water within soil had significant effect on chlorophyll contents, damages leaf chloroplast limiting photosynthesis within plant body. Drought sensitive cultivars have decreased chlorophyll *a* and *b* pigments, while no significant difference is observed in tolerant/resistant cultivars (Zaefyzadeh *et al*., 2009). Brazilian sour orange showed least reduction in chlorophyll *a* and *b*, and carotenoid contents under drought stress and hence is considered as tolerant rootstock. Sovage citrange significantly decreased the chlorophyll pigments and was regarded as sensitive citrus rootstock against drought stress.

Optimum temperature requirement for citrus growth is 25–30°C but under warm-subtropical climate during summer leaf temperature raised up to 39 to 41°C adversely affect plant growth and health. Temperature above optimum level drastically affects the photosynthetic rate and photosynthetic apparatus of plants by limiting the rate of chemical reactions. Heat stress cause negative changes in thylakoid membrane's physicochemical properties and functional organization (Berry and Bjorkman, 2003). Heat-tolerant cultivars stay-green have higher chlorophyll and carotenoids contents than sensitive cultivars when exposed to prolonged heat (Panigrahy *et al*., 2011). Brazilian sour orange was considered tolerant, stayed green and showed no significant change in chlorophyll and carotenoids under heat stress. Both trifoliate citrus cultivars Sovage citrange and Yuma citrange were found most sensitive to heat stress. Brazilian sour orange also responded well against combined heat and drought stress, hence, considered as tolerant rootstock.

The correlation coefficient is a statistical tool for analyzing association among traits. Drought, heat and combined stress had a positive correlation between shoot dry mater yield and plant health index which means that better the plant health more will be the dry matter content. Brazilian sour orange yielded high dry matter content. A positive correlation occurred between shoot dry matter content and photosynthetic pigments under drought and combined stress. Plants with high shoot dry matter yield had high chlorophyll *a*, Chlorophyll *b* and carotenoids. Plant growth or crop yield is dependent on the dry matter yield by photosynthesis and amount of photosynthetic pigment (Zheng *et al*., 2009).

Conclusion

Drought and heat stress in citrus diminish the growth by reducing the plant metabolism and affects overall plant chlorophyll and carotenoid contents which are most important regulators of gas exchange in citrus rootstocks. Brazilian sour orange, Sour orange and Keen sour orange showed better performance against high temperature and drought stress. Sovage citrange and Yuma citrange were the most sensitive rootstocks against selected stresses.

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