Original Research Article

Impact of Drought on Chlorophyll, Soluble protein, Abscisic acid, Yield and Quality Characters of Contrasting Genotypes of Tomato (*Solanum lycopersicum*)

- The MS requires major revision:

1. Material and methods must be expanded, more details on the methodologies used needed. Authors refer their readers to other's works … all methodologies must be expanded and cover the details
2. Table and graphs titles must be self-explanatory, footnotes must be added under table/graph to explain the acronyms
3. The presentation of **Results** must be completely revised:
    a. ANOVA table missing, adding the ANOVA can help seeing possible TXG interactions
    b. Include statistics for treatments averages, as of now, your tables / graphs doesnot show if the treatments were effective or not
    c. For each variable, first present the effect of treatments, the overall variation of cultivars, and then the interactions (if there is any).
    d. once results presented, include relevant discussions. In the discussion, focus on the subjects that are relevant to your objectives
4. A proofreading at the end needed, some sentences are hard to follow .
5. other comments are in the text.

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Abstracts

Impact of drought stress on chlorophyll, chlorophyll fluorescence (Fv/Fm), chlorophyll stability index (CSI), soluble protein, abscisic acid (ABA), yield and quality of tomato (*Solanum lycopersicum*) genotypes was investigated for the assessment of drought tolerance under field conditions in rainout shelter. The drought condition was created first day from transplanting based on Irrigation water (IW) /: Cumulative Pan Evaporation (CPE) of soil. Experiment was laid out with 10 genotypes by adopting FRBD with three replications and two treatments ~~viz.,~~*of*

1 IW~~/~~:CPE and 0.5 IW~~/~~:CPE. The result revealed that the reductions in chlorophyll content, Fv/Fm, chlorophyll stability index (CSI), soluble protein and yield were noticed at drought condition (0.5 IW/CPE). The genotypes LE 114, LE 57, and LE 118 which showed significantly less reduction in the above parameters during drought were considered as drought tolerant. ~~However, the~~ ABA content and quality characters ~~like~~ such as total soluble solids (TSS), lycopene content were increased under drought condition. Genotypes LE 1 and LE 125 which recorded the lowest chlorophyll content, Fv/Fm, CSI, soluble protein and higher ABA content ultimately poor yield were considered as drought susceptible.

Key-words:

Drought; Tomato; Chlorophyll; Chlorophyll Fluorescence; Soluble protein; CSI; ABA; TSS

1. Introduction

Drought is the major inevitable and recurring feature of semi-arid tropics and despite our improved ability to predict their onset, duration and impact, crop scientists are still concerned about it as it remains the single most important factor affecting the yield potentials of crop species. It is one of the serious environmental factor affecting plant growth, yield, and quality. It induces various physiological and biochemical adaptations in plants. Drought is one of the most important factors for yield reduction in the majority of the cultivated areas, affected 40 to 60% of the world's agriculture lands [1].

Water deficit leads to the perturbation of most of the physiological and biochemical processes and consequently reduces plant growth and yield [2]. Gladden *et al*. [3] showed that water deficit earlier in the growth of tomato caused a significant reduction in leaf chlorophyll content. Abdellah *et al*. [4] recorded the highest reduction in the chlorophyll content in susceptible wheat cultivar under water stress of 30% FC. Water stress reduced the total

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chlorophyll content significantly in different genotypes of moth bean and reduction was more pronounced in late flowering genotypes [5]. Sanadhya *et al*. [6] reported that the water stress reduced the chlorophyll content and hill activity with increased levels of stress in mung bean.

There was a reduction of only 1.3% and 2.2% in Fv/Fm under moderate and severe stress compared to control in *Withania somnifera* [7]. Chlorophyll fluorescence emission well on the level of water stress and, thus, can be used to identify elevated drought tolerance in tomato for selection of resistant genotypes [8]. Decreased chlorophyll content and chlorophyll stability index under both moisture stress and temperature stress wereas found by Sairam *et al*. [9] in wheat.

Daniel and Triboi [10] showed that heat stress decreased the duration of soluble protein accumulation in terms of days after anthesis but not in terms of thermal time. Few studies have investigated the combined influence of drought and heat stress on nitrogen metabolism. Abdellah *et al*. [4] reported that the increased ABA content was observed in wheat cultivar by water stress (30% FC) over control. Under intense water stress, the concentrations of ABA in plants increases, which trigger a number of processes starting from decrease in turgor pressure, decline in cellular expansion and stomatal closure to reduce water loss in leaves [11].

Meenakumari *et al*. [12] studied the physiological parameter governing drought tolerance in maize and recorded more than 80 per cent reduction in yield in highly susceptible lines while in relatively tolerant genotypes reduction was up to 50 per cent. Manojkumar *et al*. [13] reported that water stressed tomato plants showed significant difference in the TSS level at different irrigation levels. As the irrigation frequency increased TSS level decreased. Maximum per-cent

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TSS was observed under IW/CPE ratio of 0.60 (6.10%) and the minimum was recorded at the IW/CPE ratio of 1.20 (4.80%). The fruit quality improvement was observed under water deficit condition in tomato as a result of the synthesis of ascorbic acid, citric acid and malic acid [14].

Tomato (*Solanum lycopersicum*) is one of the most popular and widely grown vegetables in the world. Considering the potentiality of this crop, there is plenty of scope for its improvement, especially under the drought situation. Some of the ada~~o~~ptive mechanisms of plants to drought stress, which do not decrease~~s~~ plant yield to a greater extent, assume greater importance. There are several physiological and biochemical traits contributing to the drought tolerance of horticultural crops. However, a large number of tomato genotypes have not been screened for drought tolerance or exploited for their cultivation under drought situation and field condition.

To breed drought tolerant genotypes, it is necessary to identify physiological traits of plants, which contributes to drought tolerance. Therefore, the present investigation was carried out to study the chlorophyll characters, soluble protein and ABA to facilitate the screening and selection of tomato genotypes for drought tolerance.

2. Materials and Methods

The study was undertaken to find out effect of drought on chlorophyll characters, soluble protein, ABA, yield and quality in tomato in the field experiment at Rainout Shelter of Crop Physiology Department, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu. The experiment was conducted with ten~~10~~ tomato genotypes *viz.*, LE 1, LE 27, LE 57, LE 114, LE 118, LE 125, CO 3, PKM 1, TH CO 2 and TNAU TH CO 3 and two treatments *viz.*, 1.0 IW/CPE and 0.5 IW/CPE with three replications. Seeds of selected genotypes were sown in

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trays filled with vermicompost for nursery. Twenty five days old seedlings were transplanted and drought was imposed at first day after transplanting onwards based on IW/CPE, 0.5 IW/CPE for drought stress and 1.0 IW/CPE for control were maintained by irrigation the field at regular interval based cumulative pan evaporation. Crop was supplied with fertilizers and other cultivation operations including plant protection measures as per recommended package of practices of Tamil Nadu Agricultural University, Coimbatore. All the observations were recorded on third leaf from top at 60 DAT. The experiment was laid out in factorial randomized block design with three replications.

2.1. Chlorophyll characters→ **more information is needed for all measurements methods, DO NOT refer to other's work, include as much details on the methods as you can .**

Total chlorophyll content use regular font, not bold was estimated following the method suggested by Arnon [15] and expressed as mg g^{-1}. This is not adequate, explain how … **Chlorophyll fluorescence** measurements were recorded using Plant Efficiency Analyzer (Hansatech, UK) following the method advocated by Lu and Zhang [16].

Measurements were made on intact leaves, which were dark adapted for 30 min prior to measurement. The minimal fluorescence level (F_0) with all PS II reaction centers open was assessed by measuring the modulated light, which was sufficiently low (< 0.1 µmol m^{-2} s^{-1}) not to induce any significant variable fluorescence. The maximal fluorescence level (Fm) with all PS II reaction centers closed were determined by a 0.8 s saturating pulse at 8000 µmol m^{-2} s^{-1} in dark-adapted leaves [17]. Using light and dark fluorescence parameters, the maximal efficiency of PS II photochemistry in the dark adapted state, Fv/Fm = (Fm-Fo) / Fm [18] was calculated. Provide more details about the fluorescence measurement method

Estimation of **CSI** was carried out based on the protocol of Koleyoras [19] How? and expressed in terms of per-cent by using following formula. use Word Equation for formulas

$$\text{Chlorophyll stability index (CSI)} = \frac{\text{Total chlorophyll content (Treated)}}{\text{Total chlorophyll content (Control)}} \times 100$$

2.2. Estimation of protein and ABA content

Soluble protein content regular font of leaf was estimated as per the method of Lowry *et al.* [20] how ? and expressed as mg g^{-1} fresh weight. Quantification of **abscisic acid** was done by using the instrument HPLC cyber lab with the column of RP 18 (4.6 mm ID x 250 mm) and mobile phase of acetonitrile (60) and water (40) by adopting the protocol of Krochko *et al.* [21]. Leaf samples were extracted using 80 per cent chilled methanol following series of steps?? and finally partially purified methanolic extracts were filtered through 0.52 µm Millipore filters and injected into 20 µL injector loop fitted over the Cyber lab RP protected by guard column. Leaf collection/ sampling method, freezing, thawing … describe the method clearly

A volume of 20 µL of sample was injected into HPLC. The elution was carried out by a binary gradient of 60 per cent HPLC grade acetonitrile for 20 minute at the flow rate of 1 mL min^{-1}.

The column elutes were passed through an UV detector set at 254 nm and the ABA were estimated measuring the peak area and comparing with standard curve of hormones. The peak areas were measured and ABA concentration quantified using the standard curve obtained from ABA.

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The total weight of fruits harvested from each plant of all picking was added and average yield per plant was worked out and expressed in gram per plant. Later the **yield per hectare** was calculated and expressed as tonnes per hectare. Combine in one paragraph

2.3. Quality characters

Drop of juice extracted from cut fruit was used to determine **TSS** with the help of Hand Refractometer (0 to 32°Brix) at room temperature and the value was noted in °Brix. Lycopene content of fruit was extracted by using petroleum ether and OD of the extract was measured at 503 nm in UV-VIS-spectrophotometer using petroleum ether as a blank [22].

Lycopene content of the sample was calculated by using the following formula and expressed in mg 100 g^{-1}.

$$\text{Lycopene} = \frac{3.1206 \times \text{OD of sample} \times \text{volume made up} \times \text{dilution}}{\text{Weight of sample} \times 1000} \times 100$$

The data on various parameters were analyzed statistically as per the procedure suggested by Gomez and Gomez [23]. Wherever the treatment differences are found significant, critical differences were worked out at five per cent probability level and the values were furnished?? and discussed.

3. Results and Discussion

3.1. Impact of drought on chlorophyll characters

The intensity of the greenness in terms of **chlorophyll content** of the plant had influenced the photosynthetic rate and thereby the efficiency of the plant for increased biomass

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production. Ma *et al.* [24] reported a highly significant correlation of chlorophyll in terms of SPAD values/ readings with photosynthetic rate in soybean and Kapotis *et al.* [25] in weed species (*Amaranthus viltus* L.). Chlorophyll content in terms of SPAD values can be used for evaluation for the response of plant species to the drought and heat stresses in the field [26]. First presnt the results, then disccuss, dont start with discussion,. In the present study, the adverse effect of drought on greenness of the leaf could be observed through about 23.48 per cent reduction in mean total chlorophyll content. Rephrase, not clear, simply report the findings (apperantly reduction in SPAD due to drought? , then explain why, and what other people found The reduction of chlorophyll content under drought might be due to the fact that drought stress blemishes the chlorophyll content through causing internal modification in the thylakoid membrane. Similar to this finding, Ghaffari *et al.* [27] stated that the tolerant sunflower line had higher chlorophyll than the susceptible line under drought. Among the genotypes, highest reduction of total chlorophyll content was recorded in the genotype LE 1 (34.76%) followed by LE 125 (33.10%) and CO TH 2 (31.65%) under drought **(Table 1.).** The present study also indicated the ability of the genotypes LE 57 (18.79%), LE114 (19.65%) and LE 118 (21.37) in maintaining total chlorophyll content under drought (0.5 IW/CPE) by showing less reduction. Therefore, these genotypes were able to endure drought injury better than the sensitive lines. These findings are in agreement with the earlier findings of Petcu *et al.* [28] in sunflower.

A considerable reduction in **chlorophyll fluorescence (Fv/Fm)** was observed ~~under~~ due to the drought treatment ~~condition~~. ~~The~~ A possible reason for this effect is that the drought stressed plants have lower capacity for the use of transported electrons and their electron transport chain is more reduced at any light condition [29].

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For the treatments, ~~lesser~~ smaller mean fluorescence value (0.63) was registered by 0.5 IW/CPE with the reduction of 25.88 per cent than 1.0 IW/CPE (0.85). Relating to the genotypes, LE 57 was significantly superior chlorophyll fluorescence value (0.74) followed by LE 118 and LE 27 while the lowest was recorded by LE 125 (0.47). The genotype, LE 57 proved its supremacy with less reduction (20.69%) of Fv/Fm followed by LE 118 (20.69%) **(Table 1).** The high Fv/Fm ratio indicates that genotype has more efficient in protecting their photosynthetic apparatus under drought. This result is in agreement with Mishraa *et al.* [8] in tomato. Lower values of Fv/Fm ratio under drought, indicated an injury to electron transfer system in photo system II, causing an imbalance between generation and utilization of electrons, resulting changes of quantum yield efficiency [30].

Table 1. Effect of …. and … (water treatments) ~~water deficit~~ on total chlorophyll content and Fv/Fm of tomato genotypes at 60 days after transplanting

Genotypes	Total chlorophyll content (mg g^{-1})		Chlorophyll fluorescence (Fv / Fm)	
	1.0 IW/CPE	0.5 IW/CPE	1.0 IW/CPE	0.5 IW/CPE
LE 1	2.555	1.667	0.83	0.57
LE 27	2.932	2.284	0.87	0.67
LE 57	2.895	2.351	0.93	0.74
LE 114	2.932	2.356	0.81	0.56
LE 118	2.944	2.315	0.87	0.69
LE 125	2.007	1.878	0.75	0.47
CO 3	3.291	2.371	0.84	0.62
PKM 1	3.011	2.402	0.82	0.61
THCO 3	3.005	2.227	0.89	0.69
COTH 2	3.425	2.341	0.90	0.67
Mean	**2.900**	**2.219**	**0.85**	**0.63**
	G	**T**	**G**	**T**
SEd	**0.0241**	**0.0108**	**0.007**	**0.003**
CD (0.05)	**0.0487**	**0.0218**	**0.015**	**0.007**

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What's SEd. CD? What's G and T?

Treatments averages were different? Not seen in the table

Chlorophyll Stability Index (CSI) is an indicator of the stress tolerance capacity of the plants and is a measure of integrity of membrane [31]. A higher CSI helps the plants to withstand stress through better availability of chlorophyll, leading to increased photosynthetic rate, more dry matter production and higher productivity. Kilen and Andrew [32] observed a high correlation between CSI and drought tolerance in corn.

Drought condition aggravates chlorophyll degradation in later part of growth due to loss of membrane compartmentation. Membrane stability index decreased significantly under water stress condition over control in wheat varieties [33].

In the present study also corroborates the earlier findings with 18.49% reduction of CSI in drought (0.5 IW/CPE) compared to 1.0 IW/CPE. The primary effect of drought at the cellular level is to affect the integrity of membrane which in turn leads to disruption of cellular compartment ultimately destruction chlorophyll contents. The earlier findings of Fariduddin *et al.* [34] confirm the present study.

The lowest reduction of CSI was observed in the genotypes LE 114 (14.68%) followed by LE 118 (15.46%) while the highest reduction was showed by LE 125 (24.73%) and CO TH 2 (24.29%) under drought condition (**Table 2.**). The ability of the genotype maintained the higher CSI under drought is a desirable character for tolerance. Maintenance of CSI at drought condition by the genotype might be due to high membrane stability. Beena *et al.* [35] reported that high membrane stability index and chlorophyll stability index were recorded in tolerant inbred lines of rice than in susceptible lines under water stress condition.

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3.2. Impact of drought on soluble protein

The **soluble protein** content of the leaf, being a measure of RuBP carboxylase activity was considered as an index for photosynthetic efficiency???. Rubisco enzyme forms nearly 80 per cent of the soluble proteins in leaves of many plants [36]. Diethelm and Shibles [37] opined that the rubisco content per unit leaf area was positively correlated with that of soluble protein content of the leaf. The amount of rubisco in leaves is controlled by the rate of synthesis and degradation. Even under drought stress the rubisco holo enzyme is relatively stable with a half-life of several days [38].

However, drought stress in tomato[39], ~~Arabidopsis~~arabidopsis[40] and rice [41] leads to a rapid decrease in the abundance of rubisco small subunit (*rbc*S) transcripts, which may indicate decreased synthesis. In the present study also confirms the earlier findings with 32.28 per cent reduction of soluble protein content under drought. The reduction of soluble protein content might be due to the degradation of available soluble protein in plant and reduction of synthesis of new protein.

Among the genotypes, CO TH 2 (15.63) and TH CO 3 (15.18) registered highest soluble protein content at under 1.0 IW/CPE ratio level. During drought (0.5 IW/CPE), LE 57 recorded significantly superior soluble protein content (11.99), however the genotype LE 118 proved its endurance to water deficit with less reduction (19.48%) and LE 125 showed highest reduction of 52.66 per cent.

Biochemical limitations of photosynthetic carbon fixation by the inhibition of rubisco activity play an important role mostly under conditions of prolonged or more severe drought [42, 43]. Maintenance of soluble protein content by the genotypes could be attributed to higher

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rubisco activity leads to more carbon fixation and ultimately to higher photosynthetic efficiency under drought is one of the important traits for drought tolerance.

Table 2. Effect of water deficit on CSI and soluble protein content of tomato genotypes at 60 days after transplanting

Genotypes	Chlorophyll stability index (%)		Soluble protein content (mg g^{-1})	
	1.0 IW/CPE	**0.5 IW/CPE**	**1.0 IW/CPE**	**0.5 IW/CPE**
LE 1	79.0	65.5	10.85	6.51
LE 27	83.3	70.2	13.98	10.72
LE 57	84.6	69.5	15.03	11.99
LE 114	83.8	71.5	13.43	10.19
LE 118	85.4	72.2	14.58	11.74
LE 125	79.9	63.9	11.07	5.24
CO 3	83.0	66.4	11.55	8.69
PKM 1	82.4	66.9	11.33	7.69
THCO 3	79.5	63.0	15.18	8.46
COTH 2	80.7	61.1	15.63	8.58
Mean	**82.2**	**67.0**	**13.26**	**8.98**
	G	**T**	**G**	**T**

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SEd	**0.52**	**0.23**	**0.137**	**0.061**
CD (0.05)	**1.06**	**0.47**	**0.278**	**0.124**

3.3. Impact of drought on ABA content

It was found a significant per cent increment of **ABA content** in leaf under drought condition (39.45%) over control. The increment of ABA content under drought condition was reported by several workers [4, 11, 44]. Accumulation of ABA under drought condition is a favourable mechanism for drought tolerance through reducing transpiration rate by closing of stomata. However, complete closure of stomata leads to increment of leaf temperature which produces reactive oxygen species ultimately death of the plant.

Among the genotypes, the elevation in ABA was less in LE 114 (24%) under drought, whereas nearly double fold increment of ABA content was observed in LE 125 and LE 1 **(Fig. 1).** ABA synthesized in response to drought stress, is known to induce stomatal closure which leads to reduced transpirational water loss [45]. In the present study, LE 1 and LE 125 showed higher ABA content which ultimately recorded less transpiration rate by closing of stomata. However, the genotype LE 114 showed a moderate increment of leaf ABA content leads to partial closure of stomata with maintains the photosynthetic rate and leaf temperature. Hence, both the physiological characters are important for drought tolerance. The present study in agreement with earlier findings of Wang and Huang [46], who reported that highly significant negative correlation between ABA content and leaf water potential, stomatal conductance, transpiration rate and net photosynthetic rate.

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Fig 1. Effect of water deficit on ABA content (nmol g^{-1}) of tomato genotypes at 60 days after transplanting

1-graph titles are under the graph

2- change the line graph to bar chart and add the statistics that shows significant differences

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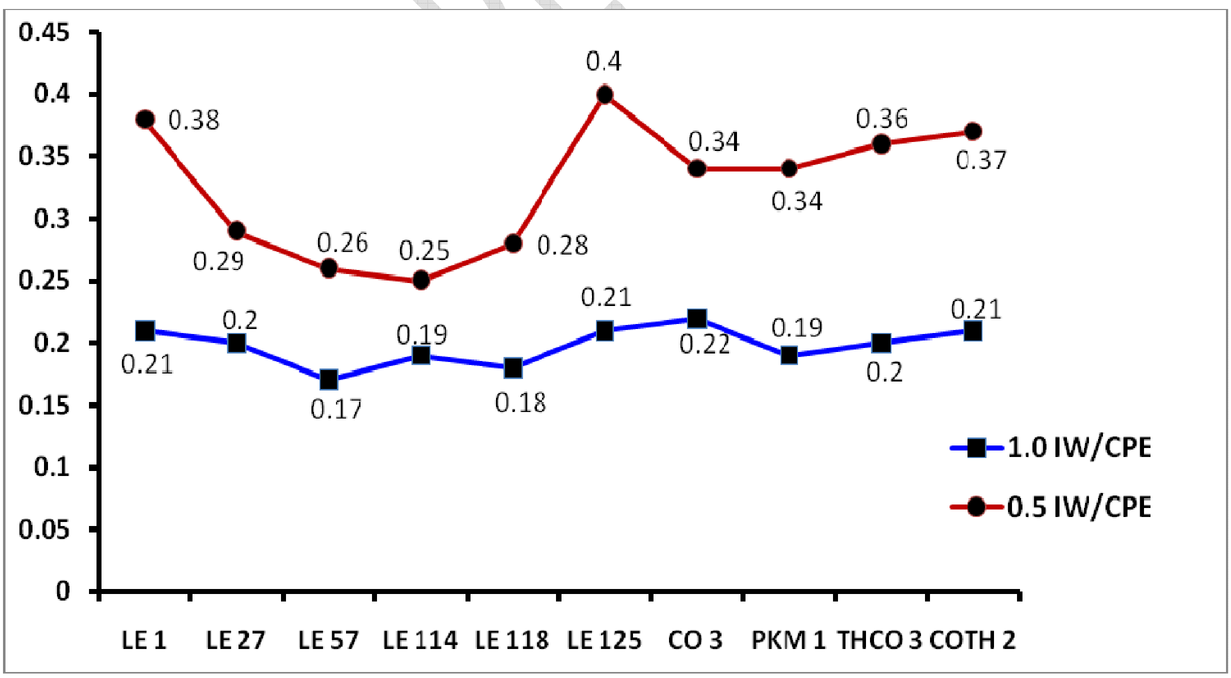

3.4. Impact of drought on yield characters

Comparing two treatments, plants received 1.0 IW/CPE ratio recorded higher average fruit yield of 62.32 than drought imposed plants (29.92) (**Table 3**). At 0.5 IW/CPE ratio level, LE 57 showed its supremacy of higher fruit yield of 54.94 which was on par with LE 118 (50.06), LE 114 (42.17) and LE 27 (40.17) while the lowest was recorded by LE 125 (10.95) and LE 1 (12.71). Drought stress resulted in the overall yield loss of tomato fruits up to 52 per cent under field condition. The highest yield loss of 83.18 and 81.51 per cent were shown by LE 125 and LE 1 respectively.

A significantly lesser reduction of 32.49 per cent was exhibited by LE 118 followed by LE 57 (33.13%) and LE 114 (38.55) showing their tolerance nature to drought stress. Therefore, it could be clearly revealed that water deficit as the result of drying soil caused a major adverse effect on yield and yield components even in tolerant genotypes. The reduction in fruit yield and related parameters under drought probably due to reduction of water content in plant which disrupting leaf gas exchange properties which limited the source size and activity (photosynthesis) and partitioning of photo assimilates to fruits. The present study confirms the early findings of Farooq *et al*. [47] and Manjunatha *et al*. [48]. Izzeldin *et al*. [49] also explained that the impact of drought before the time of flowering affects the reproductive system with the increasing sterility of flowers, so that flowering and fruiting will fail if the water shortage is prolonged.

3.5. Impact of drought on quality characters

Plants imposed with 0.5 IW/CPE ratio recorded higher Total Soluble Solids (TSS: ^{o}Brix)

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brix value (3.01) than 1.0 IW/CPE ratio (2.89). Among the genotypes, TH CO 3 recorded higher average brix value of 4.00 than the rest of the genotypes. At 0.5 IW/CPE ratio condition, the highest TSS value was recorded by TH CO 3 (4.1) followed by CO TH 2 (3.9), PKM 1 (3.6) and CO 3 (3.4) while the lowest was registered by LE 125 (2.2). Regarding treatments, plants imposed with 0.5 IW/CPE ratio recorded higher lycopene content (3.23) than 1.0 IW/CPE ratio (3.02). With respect to the genotypes, CO 3 recorded significantly higher average lycopene content (4.69). Hence, the present study indicated that the quality parameters like TSS and lycopene increased slightly under drought compared to control.

Present study corroborates with early findings of Ali *et al.* [50] in tomato. Nahar *et al.* [51] also explained that the fruit quality improvement under water deficit condition in tomato might be due to the synthesis of ascorbic acid, citric acid and malic acid. In the present study, LE 118, LE 57 and LE 27 showed their primacy with highest increment of TSS and lycopene content. This finding was strongly supported by Tambussi *et al*. [52] and it was also explained that the increase in lycopene and TSS might be an effective strategy to protect membranes from oxidative damage in water stressed condition.

4. Conclusion

Water stress causes detrimental effects on plant activities, which are likely to alter the yielding potential of the crops. Hence, to identify the physiological parameters, which get altered under drought conditions is pre-requisite to evaluate drought tolerant varieties. It is concluded that the tomato genotypes LE 118, LE 57 and LE 114 were identified as the most tolerant lines to drought stress imposed provided with Rainout shelter. As the genotypes LE 125 and LE 1 recorded significantly lesser yield under the same condition, these two genotypes were considered as susceptible to water deficit.

Table 3. Effect of water deficit on yield and quality of tomato genotypes under two treatments of ….

Genotypes	Estimated fruit yield (tonnes ha^{-1})		TSS (° Brix)		Lycopene (mg 100 g^{-1})	
	1.0 IW/CPE	**0.5 IW/CPE**	**1.0 IW/CPE**	**0.5 IW/CPE**	**1.0 IW/CPE**	**0.5 IW/CPE**
LE 1	68.74	12.71	2.5	2.7	2.21	2.39
LE 27	71.20	40.17	2.5	2.6	2.52	2.73
LE 57	82.16	54.94	2.4	2.6	2.46	2.68
LE 114	68.62	42.17	2.4	2.5	2.82	2.88
LE 118	74.15	50.06	2.4	2.5	2.85	2.95
LE 125	65.10	10.95	2.2	2.2	2.13	2.67
CO 3	41.04	22.74	3.3	3.4	4.54	4.84
PKM 1	38.98	20.94	3.5	3.6	3.78	4.05
THCO 3	54.33	22.38	3.9	4.1	3.35	3.53
COTH 2	58.85	22.13	3.8	3.9	3.54	3.55
Mean	**62.32**	**29.92**	**2.89**	**3.01**	**3.02**	**3.23**
	G	**T**	**G**	**T**	**G**	**T**
SEd	**0.960**	**0.429**	**0.03**	**0.01**	**0.048**	**0.022**
CD (0.05)	**1.943**	**0.869**	**0.05**	**0.02**	**0.097**	**0.044**

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References

1. Mollasadeghi V, Valizadeh M, Shahryari R, Imani AA. Evaluation of end drought tolerance of 12 wheat genotypes by stress indices. Middle-East J. Sci. Res. 2011; 7(2): 241-247.

2. Boutraa T, Akhkha A, Shoaibi AA, Alhejeli AM. Effect of water stress on growth and water use efficiency (WUE) of some wheat cultivars (*Triticum durum*) grown in Saudi Arabia. Journal of Taibah University for Science**.** 2010; 3: 39–48
3. Gladden L, Wang A, Hsieh YC, Tsou I. Using deficit irrigation approach for evaluating the effects of water restriction on field grown tomato (*Lycopersicon esculentum*). African Journal of Agricultural Research. 2012; 7(14): 2083- 2095.
4. Abdellah A, Boutraa T, Alhejely A. The rates of photosynthesis, chlorophyll content, dark respiration, proline and abscicic acid (ABA) in wheat (*Triticum durum*) under water deficit conditions. Int. J. Agric. Biol. 2011; 13(2): 215-221.
5. Garg BK, Burman U, Kathuja S. Effect of water stress on moth bean (*Vigna aconitifolia* (Jacq) Marechal) genotypes. Indian J. Plant Physiol. 2004; 9: 29-35.
6. Sanadhya D, Kathuria E, Kakralya BL, Malik CP. Influence of plant growth regulators on photosynthesis in mung bean subjected to water stress. Indian J. Plant Physiol. 2012; 17(3&4): 241-245.
7. Shah S, Saravanan R, Gajbhiye NA. Leaf gas exchange, chlorophyll fluorescence, growth and root yield of Ashwagandha (*Withania somnifera* Dunal.) under soil moisture stress. Indian J. Plant Physiol. 2010; 15(2): 117-124.
8. Mishraa KB, Iannaconeb R, Petrozzab A, Mishraa A, Armentanob N, Vecchiab GL, Trtilek M, Cellini F, Nedbala L. Engineered drought tolerance in tomato plants is reflected in chlorophyll fluorescence emission. Plant Sci. 2012; 182: 79-86.
9. Sairam RK, Desmukh PS, Shukla DS. Tolerance of drought and temperature stress in relation to increased antioxidant enzyme activity in wheat. J. Agron. Crop Sci. 1997;

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178: 171-177.

10. Daniel C, Triboi E. Changes in wheat protein aggregation during grain development: Effects of temperature and water stress. Eur. J. Agron. 2002; 16: 1-12.

11. Thompson S, Wilkinson S, Bacon MA, Davies WJ. Multiple signals and mechanisms that regulate leaf growth and stomatal behaviour during water deficit. Physiol. Plant. 1997; 100: 303-313.

12. Meenakumari SD, Vimala Y, Pawan A. Physiological parameters governing drought tolerance in maize. Indian J. Plant Physiol. 2004; 9: 203-207.

13. Manojkumar G, Singh P, Batra BR. A note on response of tomato to irrigation and fertility levels. Haryana J. Hort. Sci. 1998; 27(3): 215-217.

14. Nahar K, Gretzmacher R. Response of shoot and root development of seven tomato cultivars in hydrophonic system under water stress. Academic J. Plant Sci. 2011; 4(2): 57-63.

15. Arnon DI. Copper enzymes in isolated chloroplasts, polyphenoxidase in *beta vulgaris*. Plant Physiology. 1949; 24: 1-15.

16. Lu C, Zhang J. Effects of water stress on photo system II photochemistry and its thermo stability in wheat plants. J. Exp. Bot. 1999; 50: 1199-1206.

17. Lu CM, Lu QT, Zhang JH, Kuang TY. Characterization of photosynthetic pigment composition, photosystem II photochemistry and thermal energy dissipation during leaf senescence of wheat plants grown in the field. J. Exp. Bot. 2001; 52:1805-1810.

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18. Van Kooten O, Snell JHF. The use of chlorophyll fluorescence nomenclature in plant stress physiology. Photosyn. Res. 1990; 25: 147-150.

19. Koleyoras AS. A new method of determining drought resistance. Plant Physiol. 1958; 33: 232-233.

20. Lowry OH, Brought NTR, Farr LA, Randall RJ. Protein measurement with folin phenol reagent. J. Biol. Chem. 1951; 193: 265-275.

21. Krochko JE, Abrams GD, Loewan MK, Abrams SR, Cultler AJ. ABA-8-hydroxylase is a cytochrome P450 monoxygenase. Plant Physiol. 1998; 118: 849-860.

22. Ranganna S. Handbook of analysis and quality control for fruit and vegetable products. 1986. 2nd Ed. Tata Mc Graw Hill Publication Co. Ltd, New Delhi, India.

23. Gomez KA, Gomez AA. Statistical procedures for agricultural research. 1984. 2nd Ed. John Wiley and sons, NewYork, USA, pp. 680.

24. Ma BL, Morrison MJ, Voldeng HD. Leaf greenness and photosynthetic rates in soybean. Crop Sci. 1995; 35: 1411-1414.

25. Kapotis G, Zervoudakis G, Veltsislas T, Salahas G. Comparison of chlorophyll meter readings with leaf chlorophyll concentration in *Amaranthus viltus*: Correlation with physiological processes. Russ. J. Plant. Physiol. 2003; 50: 395-397.

26. Hawkins TS, Gardiner ES, Comer GS. Modeling the relationship between extractable chlorophyll and SPAD-502 readings for endangered plant species research. J. Nature Conservation. 2009; 17: 123-127.

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27. Ghaffari M, Toorchi M, Valizadeh M, Shakiba MR. Morpho-physiological screening of sunflower inbred lines under drought stress condition. Turk. J. Field Crops. 2012; 17(2): 185-190.

28. Petcu E, Arsintescu A, Stanciu D. The effect of hydric stress on some characteristics of sunflower plants. Romanian Agric. Res. 2001; 16: 15-22.

29. Dias MC, Bruggemann W. Limitations of photosynthesis in *Phaseolus vulgaris* under drought stress: gas exchange, chlorophyll fluorescence and Calvin cycle enzymes. Photosynthetica. 2010; 48(1): 96-102.

30. Reddy AR, Chaitanya KV, Vivekanandan M. Drought-induced responses of photosynthesis and antioxidant metabolism in higher plants. J. Plant Physiol. 2004; 161: 1189-1202.

31. Murthy KS, Majumder SK. Modifications of the technique for determination of chlorophyll stability index in relation to studies of drought resistance in rice. Curr. Sci. 1962; 31: 470-471.

32. Kilen TC, Andrew RH. Measurement of drought resistance in corn. Agron. J. 1969; 61(5): 669-672

33. Gupta NK, Gupta S, Kumar A. Exogenous cytokinin application increases cell membrane and chlorophyll stability in wheat (*Triticum aestivum* L.). J. Cereal Res. Comm. 2000; 28(3): 287-291.

UNDER PEER REVIEW

34. Fariduddin Q, Khanam S, Hasan SA, Ali B, Hayat S, Ahmad A. Effect of 28-homobrassinolide on the drought stress-in-duced changes in photosynthesis and antioxidant system of *Brassica juncea* L. Acta Physiol. Plant. 2009; 33: 889-897.

35. Beena R, Thandapani V, Chandrababu R. Physio-morphological and biochemical characterization of selected recombinant inbred lines of rice for drought resistance. Indian J. Plant Physiol. 2012; 17(2): 189-193.

36. Joseph MC, Randall DD, Nelson CJ. Photosynthesis anmd RUBP-case of polyploidy tall fescue. Plant Physiol. 1981; 68: 894-898.

37. Diethelm R, Shibles R.Relationship of enhancedsink de-mand with photosynthesisandamount and activityofribulose1, 5-bisphosphatecarboxylasein soybean leaves. J.Plant Physiol. 1989; 134: 70-74

38. Webber AN, Nie GY, Long SP. Acclimation of photosynthetic proteins to rising atmospheric CO_2. Photosyn. Res. 1994; 39: 413-425.

39. Bartholomew DM, Bartley GE, Scolnik PA. Abscisic-acid control of rbcSandcabtranscriptionintomatoleaves. PlantPhysiol. 1991;96:291-296.

40. WilliamsSJ,Bulman MP,Neill SJ. Wilt-inducedABAbiosynthesis,gene-expressionanddown-regulationofrbcSmessenger RNAlevelsin*Arabidopsisthaliana*. Physiol. Plant. 1994; 91:177-182.

41. Vu JCV, Gesch RW, Allen LH, Boote KJ, Bowes G. CO_2 enrichment delaysarapid, drought - induced decrease in Rubisco small subunit transcript abundance. J. Plant Physiol. 1999; 155: 139 -142.

UNDER PEER REVIEW

42. Lawlor DW, Cornic G. Photosynthetic carbon assimilation and associated metabolism in relation to water deficits in higher plants. Plant Cell Environ. 2002; 25: 275-294.

43. Medrano H, Escalona JM, Bota J, Gulias J, Flexas J. Regulation of photosynthesis of C_3 plants in response to progressive drought: Stomatal conductance as a reference parameter. Ann. Bot. 2002; 89: 895-905.

44. Unyayar S, Keles Y, Unal E. Proline and ABA levels in two sunflower genotypes subjected to water stress. Bulg. J. Plant Physiol. 2004; 30: 34-47.

45. Schroeder JI, Allen GJ, Hugouvieux V, Kwak JM, Waner D. Guard cell signal transduction. Annu. Rev. Plant Physiol. Plant Mol. Biol. 2001; 52: 627-658.

46. Wang Z, Huang B. Physiological recovery of Kentucky bluegrass from simultaneous drought and heat stress. Crop Sci. 2004; 44: 1729-1736.

47. Farooq M, Wahid A, Kobayashi N, Fujita D, Basra SMA. Plant drought stress: effects, mechanisms and management. Agron. Sustain. Dev. 2009; 29: 185-212.

48. Manjunatha MV, Rajkumar GR, Hebbara M, Ravishankar G. Effect of drip and surface irrigation on yield and water-production efficiency of brinjal (*Solanum melongena*) in saline vertisols. Indian J. Agric. Sci. 2004; 74(11): 583-587.

49. Izzeldin H, Lippert LF, Takatori FH. An influence of water stress at different growth stages on yield and quality of lettuce seed. J. Amer. Soc. Hort. Sci. 1980; 105(1): 68-71.

50. Ali AK, Delbert WH, William OP. Evaluating leaf water potential, stomatal resistance and canopy surface temperature of tomatoes as indices for irrigation timing. Acta Hort. 1980; 100: 181-192.

UNDER PEER REVIEW

51. Nahar K, Ullah SM, Islam N. Osmotic adjustment and quality response of five tomato cultivars (*Lycopersicon esculentum* Mill) following water deficit stress under subtropical climate. Asian J. Plant Sci. 2011; 10: 153-157.

52. Tambussi EA, Bartoli CG, Beltrano J, Guiamet JJ, Araus JL. Oxidative damage to thylakoid proteins in water-stressed leaves of wheat (*Triticum aestivum*). Physiol. Plant. 2000; 108: 398-404.

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