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RESEARCH ARTICLE

Chlorophyll stability index : A rapid method of screening chilli accessions to drought under tropical conditions

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SUMMARY

Chilli is one of the important spices as well as vegetable crops of India and worldwide, which is predominantly grown as a rainfed crop in southern states of India, but now-a-days there is a drastic rise in temperature which affects the availability of water for irrigation. Chlorophyll plays a vital role in photosynthesis. But, they are highly thermosensitive in nature and degradation occurs when subjected to high temperature. So, for this changing environment there is anurgent need breed heat tolerant varieties. Chlorophyll stability index (CSI) is a function of temperature and is inversely related to the degree of stress conditions imposed on the plants. So the stability of chlorophyll pigments can be correlated with drought tolerance. So, CSI can be a rapid screening method of chilli genotypes for drought tolerance or susceptibility. We observed that, among genotypes used in the study M106 showed higher mean CSI value of 88.41 per cent followed by F02 (86.56%) and M08 showed least CSI value of 42.47 per cent. We can deduce that,M 106 can be used infuture breeding programmes for evolving drought tolerant chilli varieties, so that we can bring more acreage under chilli cultivation under rain fed cultivation in India.

Key Words : Chlorophyll stability index, Rapid method, Screening chilli accessions

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Chilli is one of the important spice as well as vegetable crops of India and worldwide. Majority of chilli is produced in the states of Gujarat, Punjab, Odisha and North Eastern states.But, it is predominantly grown as a rainfed crop in southern states of India such as Andhra Pradesh, Maharashtra, Karnataka, Tamil Nadu and some parts of Kerala. Though India is the leading producer of chilli, the average yield is very low (1.11 t/ha dry chilli) as compared to developed countries like USA, China, South Korea, Taiwan etc., where the average yield ranges from 3-4 t/ ha. There is vast scope to increase the production and promote export besides meeting our domestic requirements.

Global warming has resulted in long-term

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fluctuations in climatic parameters, encompasses high and low temperature, heavy rainfall, long-lasting drought and high evapo-transpiration. Frequency and intensity of extreme weather events may significantly affect agriculture at larger scale. Water is recognized as an essential resource for economic and social development and also a key input in agricultural production, which influences number of physical, physiological and biochemical processes of the plant. Water deficit leads to the perturbation of most of the biological processes of the plant (Farooq *et al*., 2008) and consequently reduces plant growth and yield almost by 50 per cent (Wang *et al*., 2003; Allahmoradi *et al*., 2011; Hemaprabha *et al*., 2004 and Mollasadeghi *et al*., 2011).

Understanding abiotic stress responses in plants is an important and challenging topic in plant research which includes abiotic stresses such as drought, low or high temperature, salinity, flooding, shade (Bohnert and Sheveleva, 1998) that causes heavy losses worth hundreds of million dollars. Among them, water stress is the most widely operative stress that causes heavy loss in productivity of crops every year after disease infestation (Mahajan and Tuteja, 2005; Anjuum *et al*., 2011 and Hammad and Ali, 2014). High temperature is one the major factor that influence water stress conditions. Under high temperature stress the cell generally shrinks and reduces its volume making its solutes more viscous which leads to excessive generation of reactive oxygen species (ROS) and results in oxidative stress and this can be detrimental to photosynthetic mechanism (Hasanuzzaman *et al*., 2013 and Hoekstra *et al*., 2001). In India, it is observed that the annual mean temperature has increased at the rate of 0.42°C (Arora *et al*., 2005). Even a change in small magnitude at critical stages of chilli can cause yield reduction.

Adequate irrigation is necessary to realize maximum yield potential of any crop plant. But in India, chilli cultivation is confined to warm and semi-arid regions, where water is often a limiting factor for production. Where in, the number of drought period, a complex combination of stresses involving both moisture stress and high temperature and erratic south west monsoon in recent times are major constraints in chilli production (Tas and Tas, 2007). Plant response to heat stress varies with the species and growth stage as well as the duration of water limitation (Farooq *et al*., 2009 and Aslam *et al*., 2014). Drought in chilli is damaging the crop due to rapid expansion of area inplaces reeling under repeated water stress across the world (Postel, 2000). Identification and breeding varieties that have greater ability to use limited water and tolerate high temperatures is imperative and thereby enhancing the productivity of the crop (Chaves *et al*., 2003). The response mechanism of plants towards drought stress is highly complex. Therefore, development of drought-tolerant chilli varieties should be mandatory objective in the future research. The physiological screening and the cultivar identification are establishing theoretical and practical foundation for breeding chilli for drought tolerance. Leaves are the most sensitive parts of plant to drought and at the same time, they are the most essential organ for the manufacture of food by the process known as photosynthesis.

In leaves, chlorophyll plays a vital role in the photosynthesis that helps in conversion of carbon dioxide and water into oxygen and glucose. Amount of chlorophyll content is correlated with dry matter production of a plant. The amount of solar radiation absorbed by a leaf is largely a function of the foliar concentrations of photosynthetic pigments and, therefore, low concentrations of chlorophyll can directly limit photosynthetic potential and hence, primary production (Curran *et al*., 1990 and Filella *et al*.,1995). Besides, chlorophyll pigments are highly thermo sensitive in nature and degradation occurs if the amount of light absorbed is above the capacity of the chloroplasts in many crops including, wheat (Sarker *et al*., 1999), grass *Eragrostiscurvula* (Colom and Vazzana, 2003), cattail (*Typhalatifolia*) (Li *et al*., 2004) and turfgrasses (Jiang and Huang, 2001). This may result in photo-oxidative destruction of the photosynthetic apparatus (Long *et al*., 1994 and El-Sharkawi and Salama, 1977). In this changing environment, there is an urgent need to evolving heat tolerant varieties. As an adaptive measure thermostability of chlorophyll, cell membranes and photosynthesis are, therefore, increasingly being used to screen genotypes for heat tolerance (Sadalla *et al*., 1990). Use of crop varieties that are tolerant to drought stress is an effort to reduce losses from drought (Abdulmalik *et al*., 2012 and Sumartini *et al*., 2013). Screening for drought tolerance is still complicated process due to lack of fast, reproducible, simple and convenient parameters and the inability to routinely create defined and repeatable water stress conditions, where a large number of genotypes can be screened efficiently.

The conventional approach of breeding for drought resistance has been based primarily on pod yield, a complex trait which integrates a number of local adaptation factors this might be confined or suitable to a given location and not across the wider locations because of large genotype × environment interactions for yield (Branch and Hildebrand, 1989; Jackson *et al*., 1996 and Araus *et al*., 2002). In order to improve drought tolerance and screening the existing germplasm for future drought resistance breeding programmes, understanding genotype and phenotype response and also their interaction to array of changing environmental conditions especially drought could be a better outreach to develop or induce drought tolerance in new or conventional germplasms, respectively. With inadequate time, spaceand funds,it becomes difficult to test all the available genetic material in the field for heat stress. It is, therefore, necessary that, some criterion should be established for carrying out rapid screening of a large number of provenances, even in the nursery stage, for selecting the most promising ones for field trials.

Chlorophyll stability index (CSI) is a function of temperature and is inversely related to the degree of stress conditions imposed on the plants. So CSI can be correlated with drought tolerance (Koleyoreas, 1958; Ali and Patil, 1986 and Pawar *et al.,* 2004). Higher the CSI value indicates lower effect on chlorophyll content of plants. A higher CSI value helps plants to withstand stress through better availability of chlorophyll (Yadav *et al*., 1991). This also leads to increased photosynthetic rate, more dry matter production and higher productivity (Madhanmohan *et al*., 2000). CSI can be considered asan important parameter and could be used successfully for rapid screening of germplasmor cultivars for drought tolerance (Yadav *et al.*,1991). CSI indirectly indicates how well chlorophyll can perform under stress. Previous studies revealed that, there is a significant and positive correlation with CSI under moisture stress conditions (Reddy *et al.*, 2003).

Hence, CSI serves as a potent character for rapid screening of genotypes for drought tolerance in chilli with less affected by environmental variations (Misra, 1980). CSI can be taken as a valid criterion for making an initial selection of the drought hardy genotypes in the nursery stage; thus, narrowing the number of available provenances to a few likely promising ones for further intensive field trials. Thus, CSI plays asignificant role in developing well-adapted productive genotypes and in directing the breedtag of interracial and interspecific hybrids towards adaptation to wider localities.

MATERIAL AND METHODS

An experiment replicated thrice was conductedat College Orchard, Horticulture College and Research Institute (HC and RI), Tamil Nadu Agricultural University (TNAU), Coimbatore in May 2018. In the present investigation, twenty-three genotypes of chilli were usedfor estimating chlorophyll stability index (CSI).

The list of genotypes used for study ispresented in Table A.

Table A : Following genotypes were used in this study

Genotype name	Genotype name
M08	LCA 206
F702	CA166
F704	CA41
F01	F701
F06	Male
M105	Jayanthi KD/PY
M106	Parthaman
TA/CA/17	PLR 1
CA173	F02
LCA235	F101
V. S. Ramachandran	M412
Rajamani	

Chlorophyll stability index (CSI):

Leaf samples were selected randomly from the plants and homogenized in a mortar using acetone. The extract was centrifuged at 5000 RPM for 5 min. Absorbance of the supernatant was recorded at 663 and 645 nm, spectrophotometrically. Chlorophyll (Chl) content was determined following the method described byArnon (1949). The chlorophyll stability index (CSI) was determined according Murthy and Majumdhar (1962).

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

Based on CSI values plants can be classified into four different groups	
More than 85 per cent	Highly drought tolerant
75 to 85%	Moderately drought tolerant
65 to 75%	Drought susceptible
Less than 65%	Cannot survive under drought

RESULTS AND DISCUSSION

The chlorophyll stability index is an indicative of the maintenance of photosynthetic pigments under drought situation. Higher the CSI value indicates that stress had no much impact on chlorophyll content of plants and better tolerance to heat stress. Data analysis showed that, among the genotypes M106 and F02 maintained a good mean CSI valueof 88.41 and 86.56 per cent, respectively. On the otherhand, the genotype CA166 followed by M412, LCA 206 and Rajamani (79.56, 78.34, 77.60 and 76.63%, respectively) was found torank under moderate category, indicating the moderately tolerant natureas far as the moisture stress is concerned. The susceptible genotype V.S. Ramachandran followed by LCA 235, CA173, TA/CA/17 showed the lowest mean value of 74.35, 74.07, 68.11 and 66.46 per cent CSI at vegetative stages, respectively. Among all the genotypes studied, M08 shoed least CSI value of 42.47 (Table 1).

In general, the tolerant genotype, namely M106 and F02 have shown least reduction in CSI when compared to M08 (Fig. 1). The high chlorophyll stability index indicates that, the plant can withstand stress by maintaining more chlorophyll due to less degradation of chlorophyll as a result of environmental stress, which leads to increase photosynthetic rate and more dry matter production (Madhanmohan *et al.*, 2000).

Hence, CSI is a promising criterion for selection of drought tolerant chilli. Genotypes M106 and F02 are highly suitable forregions that experience frequent and prolonged heat stress and can also beused as parents in the future breeding programme for transfer of heat

Table 1: Mean values of chlorophyll stability index (CSI) of genotypes

Genotypes	CSI (%)
M08	42.47
F702	83.68
F704	45.42
F01	46.90
F06	60.25
M105	43.48
M106	88.41
TA/CA/17	66.46
CA173	68.11
LCA235	74.07
V S Ramachandran	74.35
Rajamani	76.63
LCA 206	77.06
CA166	79.56
CA41	80.72
F701	60.65
Male	81.76
Jayanthi KD/PY	83.16
Parthaman	62.99
PLR 1	64.67
F02	86.56
F101	83.97
M412	78.34

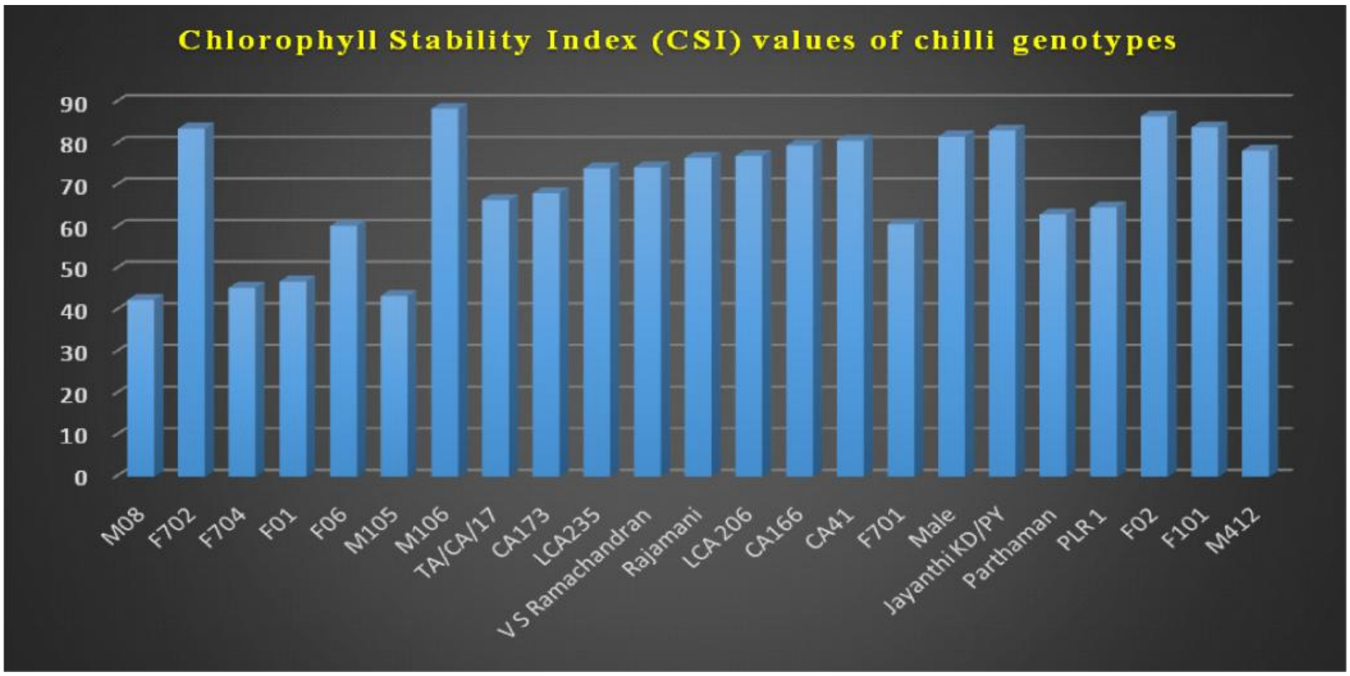

Fig. 1 : Mean CSI values are depicted in clustered column

tolerant traits to the susceptible high yielding genotypes.

Conclusion:

Temperature strongly influences the development of flowers and fruits of chilli (Mansour-Gueddes *et al*., 2010). The optimum temperature for chilli pepper cultivation is in the range of 21-33°C. Extreme conditions affect the seed germination, fruit setand fruit size (Erickson *et al*., 2001). High temperature that occurs frequently after an thesis of chilli pepper strongly affects the reproduction and yield. Besides, it also reduces number of branches, shoot dry weight and number of fruits. Drought is a worldwide problem, constraining global crop production seriously and recent global climate change has aggravated this situation and has more serious impact (Pan *et al*., 2002). Drought has significant impact on plants complex physio-chemical process, in which many biological macro and micro molecules such as nucleic acids, proteins, carbohydrates, lipids, hormones, ions, free radicals and mineral elementsare involved.

Lack of accurate screening indicator is a limiting factor for developing drought tolerant chilli cultivars. Though several researchers have been continuously working to identify various physiological markers and incorporating them through breeding several cultivars, not much success has been achieved so far. As an alternative option, CSI can be used as a rapid screening method for drought in chilli. From the present investigation, it is found that cultivar M 106 has greater tolerance to drought, which seemed to be due to better leaf water balance, chlorophyll stability as well as higher accumulation of proline, phenols and potent antioxidant like ascorbic acid, which increase the photosynthetic rate, dry matter production and high productivity. This generated information would be handy in future breeding programme to inculcate drought tolerance in chilli, which helps in bringing more acreage under rain fed chilli cultivation in India.

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Conflicts of interest:

The author declaresthat he does not have any competing interests.

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