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Analysis of Screening Tools for Drought Tolerance in Chickpea (*Cicer arietinum* L.) Genotypes

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ABSTRACT

Background: Chickpea is the third most significant food legume worldwide. While deeper rooting can increase water extraction, as it has been hypothesised for almost three decades. Therefore, understanding the regulation of leaf water losses in plant canopy, first when there is no water limitation and secondly when plants are progressively exposed to water deficit, are likely to be equally critical to roots for achieving high chickpea yield under terminal drought. Present study was carried out with an aim to evaluate the use of physiological traits such as canopy temperature depression (CTD) and chlorophyll content to determine drought tolerance of chickpea genotypes under varying environmental conditions.

Methods: Trials were conducted in 2009-2010 and 2010-2011 growing seasons at Gachsaran agricultural research station situated located in south-western of Iran. Chickpea genotypes were planted in two sets (each set had 3 replicates) by using a RCBD under two supplementary irrigation and rain-fed conditions. CTD measurements were made by infrared thermometer which was focused to 10:1 meter and at late morning to early afternoon cloudless periods (11:00 to 13:00 hours). Leaf chlorophyll content was measured at flowering and grain filling stages by using of a Minolta SPAD meter on 5-8 flag leaves per plot.

Result: The CTD results in emergence of fifty percent of inflorescence stage and CHL in grain filling stage had high significant differences. The significant and positive correlation of DI, K_2STI, Ys, GMP, STI, MP, Yp, K_1STI showed that these indices were more effective in identifying high yielding genotypes under both conditions and result showed that CTD and CHL have played important roles to search physiological basis of grain yield of chickpea and CTD and CHL can successfully use as a selection criterions in breeding programs.

Key words: Canopy temperature, Chickpea, Chlorophyll content, GGE Biplot, Warm climate.

Abbreviations: CTD: Canopy temperature depression, CHL: Chlorophyll content, SEN: Sensitive genotypes to drought stress with low yield, TOL: Tolerant genotypes to drought stress with acceptable yield.

INTRODUCTION

Pulses, besides being an indispensable component of vegetarian diet, play a vital role in sustaining long term productivity of soil through biological nitrogen fixation. After dry beans and peas, chickpea is the third most significant food legume worldwide. It is grown on 12.4 million hectares, generating 11.3 million tons at an average output of 910 kg/ha, according to FAOSTAT information in 2012-2013. In chickpea manufacturing and productivity, climate change is a significant challenge (Yadav *et al.*, 2020). The United Nations General Assembly declared the year 2016 as International year of pulses with the record production of pulses in India of 17.56 Mt. Also, India emerged as the largest chickpea producer in the world with the production of 7.8 Mt (Kumar *et al.* 2018; Gaur *et al.* 2016). Among the major chickpea producer countries, India, Pakistan, Turkey and Iran, most growing areas are classified as arid or semi-arid (Kalefetoglu and Ekmekci, 2009). In these regions, chickpea is generally grown under rain-fed conditions either on stored soil moisture in subtropical environments with summer-dominant rainfall or on current rainfall in winter-dominant Mediterranean-type environments. In both environments, non-irrigated chickpea plantations suffer yield losses from terminal drought (Yadav *et al.*, 2006). Pulses have both environmental and nutritional benefits, they are often recommended in sustainable diets (Chaudhary *et al.*, 2018). More critically for yield determination, however, are the reported effects of decreasing grain number when heat stress occurs before or around anthesis and reduced grain

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weight when it occurs during grain filling (Dolferus *et al.*, 2011; Derying *et al.*, 2014).

For crop species like chickpea that face terminal stress conditions, water availability during the grain filling period is critical. While deeper rooting can increase water extraction, as it has been hypothesised for almost three decades (Kashiwagi *et al.*, 2005), water availability during the grain filling period could also be explained by a more conservative use of water earlier during the cropping cycle. Therefore, understanding the regulation of leaf water losses, first when there is no water limitation and secondly when plants are progressively exposed to water deficit, are likely to be equally critical to roots for achieving high chickpea yield under terminal drought.

Chickpea productivity is constrained by several abiotic stresses (Gaur *et al.*, 2008) and temperature is one of the most important determinants of crop growth over a range of environments (Summerfield *et al.*, 1990) and may limit chickpea yield (Basu *et al.*, 2009). The effects of heat stress during the vegetative and reproductive growth stages using agronomic, phenological, morphological and physiological assessment has been studied in various crops such as bread wheat (Mohammadi *et al.*, 2012; Basu *et al.*, 2014), durum wheat (Karimizadeh and Mohammadi, 2011, Karimizadeh *et al.*, 2011), rice (Weerakoon *et al.*, 2008) and cotton (Cottee *et al.*, 2010) while only limited research has been conducted in chickpea (Wang *et al.*, 2006). Among cool season legumes, chickpea is less sensitive to high temperature (McDonald and Paulsen, 1997). Although chickpea is exposed to warm temperatures (>30°C) in certain regions, limited yield loss was found at 308°C, which is higher than other cool season legumes such as field peas, faba bean and lentil (Summerfield *et al.*, 1984; McDonald and Paulsen, 1997; Patrick and Stoddard, 2010). Therefore, a base level of heat tolerance is found in chickpea. However, there is no clear evidence to show the mechanism of heat tolerance.

Deviation of temperature of plant canopies in comparison to ambient temperature, also known as CTD (Canopy temperature depression = Air temperature-Canopy temperature), has been recognized as indicators of overall plant water status (Blum *et al.*, 1982) and used in such practical applications as evaluation of plant response to environmental stress, irrigation scheduling (Wanjura *et al.*, 1995), dry matter accumulation (Basu *et al.*, 2014), cultivar comparison for water use (Pinter *et al.*, 1990) and tolerance to heat (Devasirvatham *et al.*, 2010; Amani *et al.*, 1996) and drought (Talebi *et al.*, 2013; Zaman-Allah *et al.*, 2011; Royo *et al.*, 2002; Rashid *et al.*, 1999). High CTD has been used as a selection criterion to improve tolerance to drought and heat (Amani *et al.*, 1996; Blum 1996; Pinter *et al.*, 1990; Fischer *et al.*, 1998; Karimizadeh and Mohammadi, 2011; Karimizadeh *et al.*, 2011; Karimizadeh *et al.*, 2012; Mohammadi *et al.*, 2012) and has been associated with yield increase among wheat (*Triticum aestivum* L.) cultivars at CIMMYT (Fischer *et al.*, 1998). The suitability of CTD as an indicator of yield and stress tolerance, however, must be determined for individual environments. The physiological basis of drought tolerance among chickpea genotypes was associated with improved chlorophyll content from heading onwards, as well as more leaf chlorophyll retention during grain filling (Macil *et al.*, 2017), greater thermo stability of membranes indicated by electrolyte leakage (Balota *et al.*, 1993) and chlorophyll fluorescence and cooler canopies, which were associated with increased stomatal conductance (Amani *et al.*, 1996). According to Wery *et al.* (1994), critical temperature during the reproductive phase which includes flowering, filling and enlargement seeds chickpea is evaluated with 30°C. Recorded temperatures showed that critical temperature was exceeded only during the pods maturity phase duration. This reveals that this chickpea culture did not suffer from thermal stress (Basu *et al.*, 2009).

In this study, canopy temperature depression and chlorophyll content were used simultaneously to determine the relationships of CTD and chlorophyll content with drought/ heat indices, grain yield and yield components in eight chickpea lines in Gachsaran semi-warm condition of Iran.

MATERIALS AND METHODS

Plant material

Trials were conducted in 2009-2010 and 2010-2011 growing seasons at Gachsaran agricultural research station situated at 710 meters altitude above sea level with longitude 50° 50′ east and latitude 30° 20′ north is located in south-western of Iran. Soil texture of experimental site is silty clay loam and 20 years average of rainfall was 438 mm. In this study, eight chickpea genotypes by names FLIP 03-71C (G1), FLIP 98-106C (G2), FLIP 99-34C (G3), AZAD (G4), FLIP 03-71C (G5), FLIP 03-152C (G6), FLIP 88-85C (G7) and FLIP 01-52C (G8) were planted in two sets (each set had 3 replicates) by using a randomized complete block design under two supplementary irrigation and rain-fed conditions. Plots were planted at a seeding rate of 50 seed per m^2 by WINTERSTEIGER AG trial drilling machine on 28 November 2009 and 30 November 2010. Plot size was containing five rows (6 m long) with row differences of 25 cm. Fertilizers were applied 80 kg ha^{-1} of nitrogen and 80 kg ha^{-1} of phosphorus as 40.40.0 composes fertilizer at planting time. No disease was shown during growth period and weed control was made by hand. After physiological maturity, plots were harvested by WINTERSTEIGER AG trial thrasher/ harvester machine. Regional climatic data during growth seasons (Mean of November 2009 to June 2010 and November 2010 to June 2011) were relatively alike: average monthly temperature and rainfall according to months (November to June) are in Fig 1 and 2. Total rain amount were 401 and 417 mm in 2009-2010 and 2010-2011 growing seasons respectively, although, from emergence of eighty percent of inflorescence to completing of 50 percent anthesis, rain amount were zero for 21 and 32 days respectively. Twice irrigation for trial under supplementary irrigation condition at 20 February and 16 March in 2010 and 28 February and 22 March 2011 were conducted.

Measurement of canopy temperature and Chlorophyll content

CTD measurements were made by infrared thermometer (Model 8866, JQA Instrument, Inc., Tokyo, Japan) which was focused to 10:1 meter and at late morning to early afternoon cloudless periods (11:00 to 13:00 hours). The study results of Summy *et al*. (2015) revealed that the crop moisture relation parameters had direct bearing on grain yield formation *via* yield components. Therefore, the measurement of CTD and chlorophyll fluorescence during midday hours which is simple and rapid could be utilized in chickpea genotypes for crop improvement programmes of drought tolerance. As a similar to method of Fischer *et al.* (1998), the data for each plot were the mean of four readings, taken from the same side of each plot at an angle of approximately 45° to the horizontal in a range of directions such that they covered different regions of the plot and integrated many leaves. Also, measurements were at different two periods on 12th March (emergence of fifty percent of inflorescence), 20th April (completing of anthesis, watery ripe, clear liquid). Variance analysis of all agronomical traits and CTD measurements on each growth stage were carried out and the significance of cultivar mean square determined by testing against the error mean square. Leaf chlorophyll content was measured at flowering and grain filling stages by using of a Minolta SPAD meter on 5-8 flag leaves per plot. Genotypes means over all dates were compared by the least significant difference method at P<0.05 by Genstat 12 statistical packed program. Correlations between two traits were evaluated by MINITAB 14.

Drought indices

Drought is a polygenic stress and is considered as one of the most important factors limiting crop yields around the world. As climate change leads to increasingly hotter and drier summers, the importance of drought constraints on yield and yield components has increased in World (Karimizadeh *et al.*, 2012; 2016a; 2016b). Nine drought tolerance indices including stress susceptibility index (SSI), relative drought index (RDI), stress tolerance index (STI), geometric mean productivity (GMP), tolerance (TOL), mean production (MP), drought resistance index (DI), yield stability index (YSI), stress susceptibility percentage (SSPI), were calculated (Fischer and Maurer, 1978; Fischer *et al.*, 1998; Fernandez,1992; Rosielle and Hamblin, 1981).

Drought tolerant indices were calculated on the basis of grain yield of genotypes (Table 2). To investigate suitable stress resistance indices for screening of cultivars under

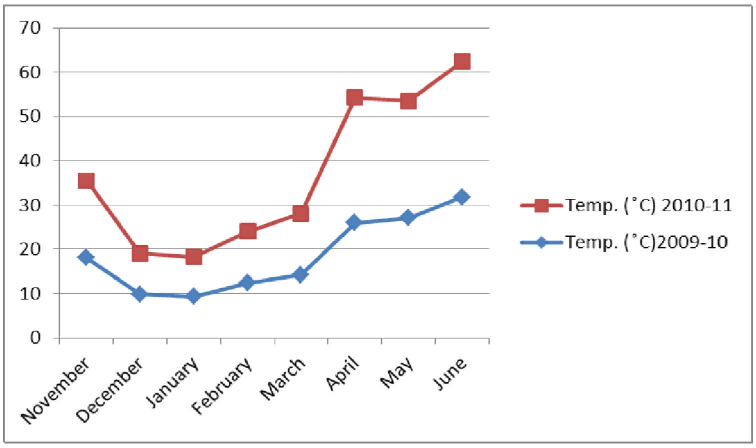

Fig 1: Temperature diagram of Gachsaran region during two years (2009-2010 and 2010-2011).

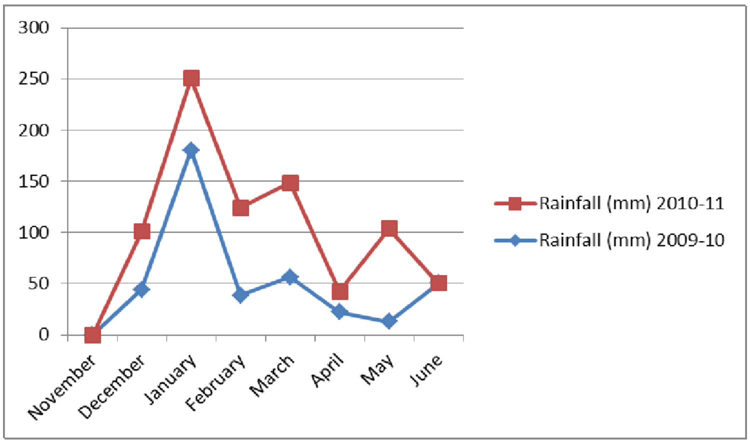

Fig 2: Rainfall diagram of Gachsaran region during two years (2009-2010 and 2010-2011).

drought condition, grain yield of cultivars under both non-stress condition (supplementary irrigation) and stress condition (rain-fed condition) were measured for calculating different sensitivity and tolerance indices.

RESULTS AND DISCUSSION

The result of combined analyses of variance revealed significant differences among genotypes for grain yield, days from sowing to flowering and maturity, plant height, agronomic score, 100-seed weight, number of filled pods per plant, number of empty pods per plant, chlorophyll content and canopy temperature depression (Table 1).

In this table, environment that defined as combination of year × condition and the genotypes showed high significant difference at 0.01 probability level for all studied traits except agronomic score (Table 1). Thus, indirect selection for a drought prone environment based on the results of optimum conditions will not be efficient. These results are in agreement with those of Kanouni *et al.* (2012) and Ghiabi *et al.* (2013). Genotype × environment (GE) interaction showed non-significant difference, also this GE interaction cannot use for determining genotypic stability. The estimates of stress tolerance attributes indicated that the identification of drought-tolerant genotypes based on a single criterion was contradictory. For example, according to STI and GMP and MP genotypes G1, G2, G3 and G4 were found drought tolerance with highest STI and grain yield under irrigation (non-stressed) condition, while genotypes G5, G6 and G7 were displayed the lowest amount of for these indices under irrigation condi-tion (Table 2). Similar results were reported by Mohammadi *et al.* (2012) in bread wheat and Naghavi *et al.* (2013) in maize data. Pireivatlou *et al.* (2010) was also noted that STI can be a reliable index for selecting high yielding genotypes. The greater the TOL value, the larger the yield production under supplementary irrigation conditions and the smaller the TOL value, the larger the yield production under rain-fed conditions. For TOL and SSI indices, the desirable drought tolerant genotypes for supplementary irrigation were G2 and G3. Suggest that selection based on TOL will result in reduced yield under well-watered conditions. Similar results were reported by Farshadfar and Elyasi (2012) and Talebi *et al.* (2009). YSI index selected the genotypes G4, G3 and G1 as the most relatively tolerant genotypes while for RDI the genotypes G4, G8 and G3 were the most relative tolerant. The smaller the SSPI value, the larger the yield production under rain-fed conditions, base on SSPI index, genotypes G8, G6 and G5 were relatively tolerant genotypes. According to K_1STI and K_2STI the genotypes G1, G2, G3 and G4 were the most relative tolerant genotypes (Table 2). DI selected the genotypes G4, G3, G1 and G2 as the best, while the genotypes G5, G6, G7 and G8 as the worst relatively tolerant

Table 1: Combined analysis of variance for supplementary and rain-fed conditions at 2 years for all traits.

Sources	DF	GY	DF	DM	PH	AS	100 SW	FP no.	EP no.	Chl.	CTD
Environment†	3	12528681**	1439**	1044**	354.6**	1.01^{ns}	187.3**	128.3**	4.3**	634**	78.4**
Error 1	8	89542	0.59	1.8	15.1	0.47	5.73	3.259	0.02	8.7	3.1
Genotype	7	799431**	15**	8.4**	55.2**	1.11^{ns}	8.37**	31.84**	2.18*	107**	4.3**
GE Interaction	21	59093^{ns}	0.96 ^{ns}	1.6 ^{ns}	13.9^{ns}	0.51 ^{ns}	1.77 ^{ns}	0.089^{ns}	0.02 ^{ns}	1.2^{ns}	0.02^{ns}
Error 2	56	48747	1.91	1.3	8.9	0.61	2.57	4.719	0.86	15.	1.56
Total	95	-	-	-	-	-	-	-	-	-	-
%CV	-	12.58	1.41	0.85	5.61	19.84	5.42	5.66	35.5	5.45	4.17

†Environment = Year × Condition.

Deg. F: Degree of freedom; GY: Grain yield; DF: Days to flowering; DM: Days to maturity; PH: Plant height; AS: Agronomic score; 100 SW: 100 seed weight; FP no.: Full pods number; EP: Empty pods number; Chl.: Chlorophyll content; CTD: Canopy temperature depression.

Table 2: Drought indices of eight chickpea genotypes under supplementary and rain-fed conditions.

Gen code	Ys	Yp	TOL	MP	GMP	SSI	STI	RDI	YSI	DI	SSPI	K_1STI	K_2STI
G1	1275.5	2679.7	1404.2	1977.6	1848.7	1.422	0.22	1.00	0.48	0.54	29.5	1.270	1.268
G2	1276.2	2710.8	1434.6	1993.5	1859.9	1.437	0.22	0.99	0.47	0.53	30.2	1.299	1.270
G3	1329.6	2752.3	1422.8	2040.9	1913.0	1.403	0.24	1.01	0.48	0.57	29.9	1.340	1.378
G4	1366.8	2552.9	1186.1	1959.8	1867.9	1.261	0.23	1.12	0.54	0.65	24.9	1.152	1.457
G5	883.5	2022.2	1138.6	1452.8	1336.7	1.528	0.12	0.92	0.44	0.34	23.9	0.723	0.609
G6	951.5	2061.4	1109.9	1506.4	1400.5	1.462	0.13	0.97	0.46	0.39	23.3	0.751	0.706
G7	934.5	2092.9	1158.4	1513.7	1398.5	1.502	0.13	0.94	0.45	0.37	24.4	0.775	0.681
G8	1042.2	2152.0	1109.8	1597.1	1497.6	1.400	0.14	1.02	0.47	0.45	23.3	0.819	0.847

Ys: Yield in stress condition; Yp: Yield in non-stress condition; TOL: Tolerance index stress; MP: Mean production; GMP: Geometric mean productivity; SSI: Susceptibility index; STI: Stress tolerance index; RDI: Relative drought index; YSI: Yield stability index stress; DI: Drought resistance index; SSPI: Stress susceptibility percentage index; K_1STI: Modified stress tolerance index in moderate and severe stress.

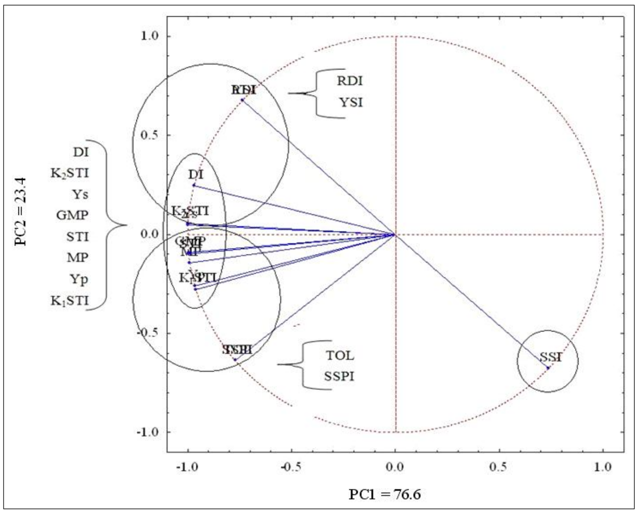

Fig 3: Grouping and biplot of drought tolerance indices.

genotypes. Majidi *et al.* (2011) reported that GMP, STI and HM indices were similarly able to separate drought sensitive and tolerant genotypes of safflower in both mild and intense water stress environments. Talebi *et al.* (2009) also reported that cultivars producing high yield in both drought and well watered conditions can be identified by STI, GMP and MP values. Selection based on a combination of indices may provide a more useful criterion for improving drought tolerance of chickpea but study of correlation coefficients are useful in finding the degree of overall linear association between any two attributes. Thus, a better approach than a correlation analysis such as biplot is needed to identify the superior genotypes for both rain-fed and supplementary irrigation environments. Principal component analysis (PCA) revealed that the first PCA explained 81.25% of the variation, thus, the first dimension can be named as the yield potential and drought tolerance. Considering the high and positive value of this biplot, genotypes that have high values of these indices will be high yielding under rain-fed and supplementary irrigation environments. The second PCA explained 18.68% of the total variability and correlated positively with TOL and SSI. Therefore, the second component can be named as a stress-tolerant dimension and it separates the stress-tolerant genotypes from supplementary irrigation tolerant ones. Thus, selection of genotypes that have high PC1 and low PC2 are suitable for both rain-fed and supplementary irrigation environments (Fig 3). Therefore, genotype G4 was superior genotypes for both environments with high PC1 and low PC2. Genotypes G1, G2 and G3 with high PC2 were more suitable for supplementary irrigation environment than for rain-fed environment. Farshadfar and Elyasi (2012), Sabaghnia *et al.* (2014), Hamayoon *et al.* (2011) in chickpea and Karimizadeh and Mohammadi (2011), Karimizadeh *et al.* (2012), Sio-Se Mardeh *et al.* (2006) and Golabadi *et al.* (2006) obtained similar results in multivariate analysis of drought tolerance in different crops.

For these eight genotypes, the crop cycle was reduced from 131 days (emergence to physiological maturity) in TOL group, to an average of 136 days in the SEN group in rain-fed condition. A comparison of performance on a per day basis between TOL and SEN groups showed that, under drought stress, total yield and yield per day at maturity had 37.7% and 32.6% differences, respectively. The crop cycle was reduced from 139 days (emergence to physiological maturity) in TOL group, to an average of 141 days in the SEN group in supplemental irrigation condition. A comparison of performance on a per day basis between TOL and SEN groups showed that, total yield and yield per day at maturity had 28.4% and 29.7% differences respectively. A comparison of other trait performance between TOL and SEN groups showed that, pods per plant, empty pods number and filled pods number had 25.4%, 22.7% and 48.9% differences respectively in rain-fed condition but these traits had 12.5%, 27.8% and 17.3% differences respectively in supplementary irrigation condition (Table 3).

Table 3: Performance and morphological/physiological traits for the mean of three drought tolerance (TOL) and four drought sensitive (SEN) chickpea genotypes over 4 environment.

Traits	Rain-fed condition		% difference	Supplementary irrigation		% difference
	TOL	SEN		TOL	SEN	
Grain yield (kg ha^{-1})	2186.6	1588.2	37.7	2673.9	2082.1	28.4
Number of pods in 1 m^2	1495	1106	35.2	1687	1456	15.9
Pods per plant	28.1	22.4	25.4	31.6	28.1	12.5
Number of empty pods	2.2	4.9	22.7	1.8	2.3	27.8
Number of filled pods	25.9	17.4	48.9	29.8	25.4	17.3
Days to flowering	98	95	3.2	99	100	1.0
Days to maturity	136	131	3.8	139	141	1.4
Pods filling period	38	33	15.2	45	43	4.7
Plant height	54	44	22.7	58	54	7.4
Agronomic score	4.2	3.5	20.0	4.6	4.2	9.5
Yield per day (kg ha^{-1}d^{-1})	16.1	12.1	32.6	19.2	14.8	29.7
100 kernel weight (g)	32.7	27.6	18.5	34.1	32.3	5.6

Differences in yield and morpho-physiological traits for the TOL versus SEN genotypes was apparently associated with differing performance during grain filling, as reflected by the difference in grain filling rate, rather than parameters up until anthesis (Table 3).

The physiological data indicate similar contrasts between TOL and SEN genotypes for photosynthetic chlorophyll content and canopy temperature again with differences most pronounced from flowering onwards (Table 4). Result showed that chlorophyll content values in 50% flowering stage were highest value ratio grain filling stage in both conditions. The reducing of chlorophyll content in supplemental irrigation condition was clearer than rain-fed condition (Table 4). Differences between chlorophyll content values in 50% flowering stage in supplemental irrigation were smaller than rain-fed condition. One of the most important factors can be impact of irrigation water on creating of mild environment and therefore difference between chlorophyll content of leaves in TOL and SEN groups were decreases. This result is consistent with the reports of Talebi *et al.* (2013) and Karimizadeh *et al.* (2011).

The CTD measurements were taken at the stages of 50% flowering and watery ripe, clear liquid (grain filling stage). Genotypic differences were detected at the first stage for both supplementary irrigation and rain-fed condition on chickpea genotypes. CTD values changed between 4.6°C (SEN genotypes) and 6.2°C (TOL genotypes) and between 7.2°C (SEN genotypes) and 6.9°C (TOL genotypes) in rain-fed and supplemental irrigation conditions respectively. At grain filling stage, CTD changed between 3.1°C (SEN genotypes) and 3.9°C (TOL genotypes) and between 6.3°C (SEN genotypes) and 5.9°C (TOL genotypes) in rain-fed and supplemental irrigation conditions respectively (Fig 4 and 5).

High temperature after flower opening decreases chickpea seed yield by reducing the number of seeds per plant and weight per seed (Wang *et al.*, 2006). In chickpea, Summerfield *et al.* (1984) suggested that the longer the exposure during reproductive development to a high day

Table 4: Physiological parameters for the mean of four drought tolerant (TOL) and four drought sensitive (SEN) chickpea genotypes measured on leaves at two phenological stages.

Condition	50 per cent flowering stage	Grain filling stage
Rain-fed condition		
Canopy temperature depression		
TOL	6.2	3.9
SEN	4.6	3.1
Chlorophyll content (SPAD)		
TOL	66.7	58.1
SEN	61.9	51.6
Supplemental irrigation condition		
Canopy temperature depression		
TOL	6.6	5.9
SEN	7.2	6.3
Chlorophyll content (SPAD)		
TOL	59.8	56.5
SEN	61.1	50.8

temperature of 35°C, the lower the yield. Most chickpea genotypes do not set pods when temperatures reach >35°C (Basu *et al.*, 2009). However, there is considerable variation among genotypes for response to high temperature. The period of anthesis and seed set are clearly critical stages for exposure to heat stress (Gross and Kigel 1994). Nayyar *et al.* (2005) suggested that the development of male (pollen, anthers) and female (stigma-style, ovary) parts are the most sensitive organs to abiotic stress in reproductive biology. Therefore, pollen viability, stigma receptivity and ovule viability are useful indicators of sensitivity to abiotic stress (Nayyar *et al.*, 2005). However, the effect of stress on either male or female organs depends upon the stage of sporogenesis (micro or mega). Due to heat stress, meiosis and pollen development are the most affected part in micro-sporogenesis. Megaspore formation in the ovule and fertilisation are the most important events in mega-

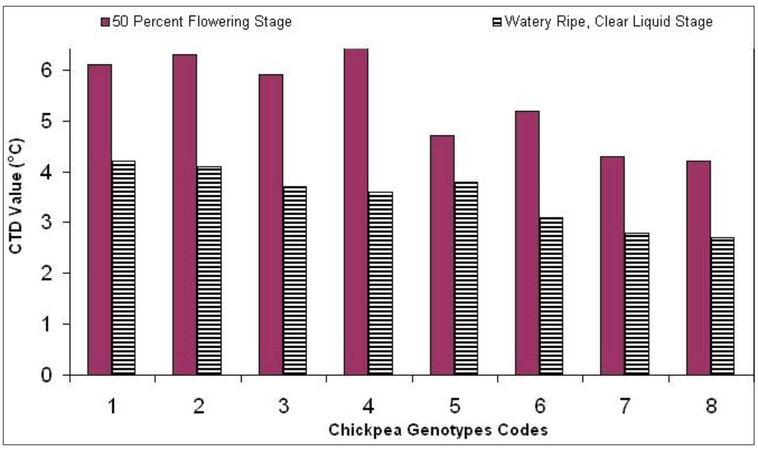

Fig 4: Canopy temperature depression values of chickpea genotypes in rain-fed condition.

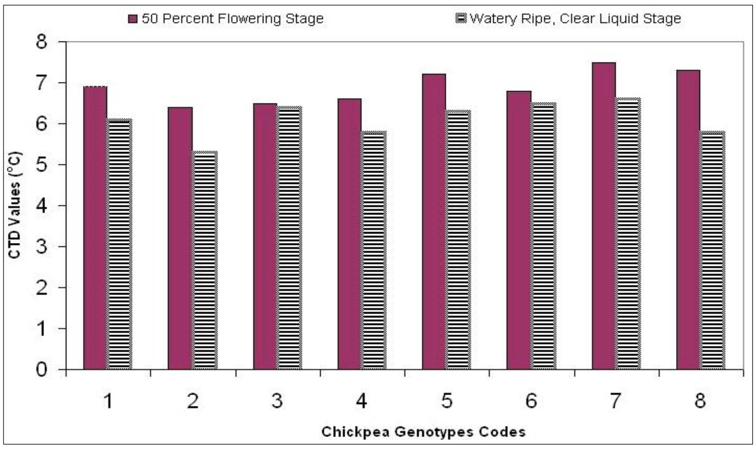

Fig 5: Canopy temperature depression values of chickpea genotypes in supplementary irrigation condition.

sporogenesis under high temperature stress (Gross and Kigel,1994).

Devasirvatham *et al.* (2010) believes that although classification of heat responses of chickpea has been documented (Upadhyaya *et al.*, 2011), there has been little attempt to extrapolate these findings across the world's chickpea production areas. The determination of a heat response phenotype through screening is vital if the genetic control of heat tolerance in chickpea is to be understood and significant progress made through plant breeding. Clearly, the research under high temperature stress shows that early phenology is the most important mechanism and pod set the primary yield component to be considered in heat tolerance breeding.

In drought sensitive chickpea parent HC1 relative stress injury (RSI) was recorded 16.4% under irrigated whereas 31.3% under drought condition. Whereas, in drought tolerant ICC 4958, RSI was recorded 16.3% under irrigated and 20.9% under drought conditions (Yadav *et al.*, 2015). Also in tolerant parent ICC 4958, there is increase in CTD from -1.1 under irrigated condition to 0.9 under drought conditions is less as compared to HC 1 from -0.4 in irrigated condition to 2.6 in drought conditions. In best yielding progeny lines, CTD ranges from -1.97 to 0.50 under drought condition (Yadav *et al.*, 2015). Rees *et al.* (1993) reported that CTD values have been changed between 3.54 and 5.10°C before anthesis, 3.16 to 4.61°C after anthesis in bread wheat. Reynolds *et al.* (1997) reported that CTD average values of heat stress tolerant genotypes in bread wheat were respectively 7.4, 9.0 and 6.5°C before heading, at heading and grain filling periods. These values were respectively 7.1, 7.9 and 5.7°C at the same periods in susceptible genotypes. In this study, it has been shown similar to this situation; for instance, CTD values have been observed such as 6.9, 9.1 and 5.3°C in G6 before heading, at heading and grain filling periods respectively in rain-fed condition. It has understood that this genotype have cooler plant canopy than the other cultivars. Also, Barma *et al.* (1997) showed that CTD values could have been changed -2.4 and -5.5°C sometimes. The rankings of the indices based on the power

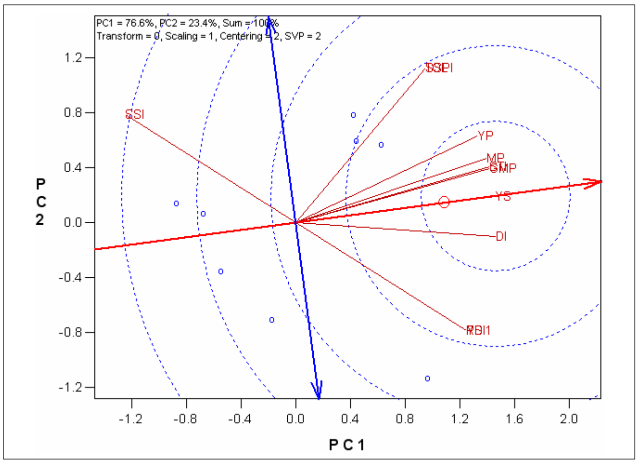

Fig 6: Ranking indices based on both discriminating ability and representativeness.

of representation are shown in Fig 3. The results showed that the Ys Index showed the best representation power and the DI, STI and GMP indices had the lowest angle with the Ys Index and showed the next best rankings to evaluate diversity and power of representation (Fig 6).

In GGE biplot methodology, the yield and stability of the genotypes are examined by an average tester coordinate (ATC). The mean yield of the genotypes is estimated by their projections on the ATC × axis. The average location, as the virtual location, is shown by a circle and indicates the positive end of the ATC × axis. According to the ATC figure, the length of the average location vector was adequate to select genotypes based on mean yield. In the GGE model, G4 was the least stable genotype which had variable performance across test locations, while G6, G3 and G1 were the most stable genotypes. The performance of genotype G6 close to ATC axe was stable, whereas this genotype showed low mean yields (Fig 7).

In different crops, as well as in chickpea, differential genotypic response to drought stress, as a result of variation in physiological parameters has been reported (Gunes *et al.*, 2006; Gunes *et al.*, 2008). In this study, we tried to explain

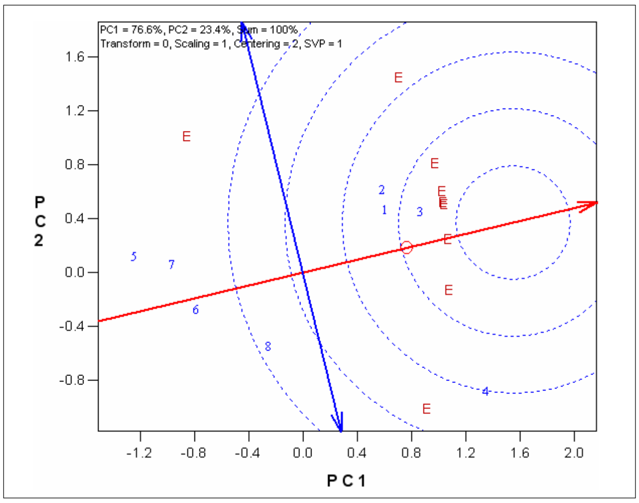

Fig 7: Ranking chickpea genotypes based on yield and stability in four environments.

the responses of the genotypes and discussed some physiological parameters that were affected by drought stress. These parameters were also evaluated as drought tolerance selection criteria. Drought stress can also alter the tissue concentrations of chlorophylls and carotenoids (Jaleel *et al*., 2008; Kalefetoglu and Ekmekci, 2009). While increased chlorophyll and carotenoid content under drought stress may be related to a decrease in leaf area, it also can be a defensive response to reduce the harmful effects of drought stress (Farooq *et al*., 2009). The photochemical efficiency was recorded in the range of 0.654 to 0.770 for highest yielding 20 progeny lines. Increase in CTD might have occurred due to decreased transpiration resulting from stomatal closure. The photosynthetic efficiency, transpiration and the values of relative stress injury declined in chickpea under drought conditions (Kumar *et al*., 2012). The total chlorophyll content significantly decreased in all genotypes under drought stress, but the reductions were not as great in tolerant genotypes. Higher level of carotenoid concentration in drought-tolerant genotypes has also been reported (Deng *et al*., 2003; Kalefetoglu and Ekmekci, 2009). Chlorophyll content was greater at 10 and 20 t. ha of biochar compared with control at all measurement dates in the winter sowing. This response could partly be attributed to the effect of biochar on plant nutrient status (Lusiba *et al*., 2016). Drought-tolerant genotypes accumulated more carotenoids than susceptible genotypes. Accumulation of carotenoids for osmotic regulation in drought-stressed leaves in many crops has been reported (Khan *et al*., 2001; Gunes *at al.,* 2008).

CONCLUSION

The results of calculated gain from indirect selection in moisture stress environment would improve yield in moisture stress environment better than selection from non-moisture stress environment. Breeders should, therefore, take into account the stress severity of the environment when choosing an index. Estimating yield from a small number of short-term CTD measurements seems much more dubious, however, since short-term CTD and transpiration rate are related to temporally variable environmental properties including irradiance, air temperature, wind speed and vapour pressure deficit. If suitable days are used for CTD measurement in terms of sufficiently high irradiance, sufficiently low wind speed, no rainfall and sufficient vapour pressure deficit to permit transpiration, fairly consistent rankings for genotypes can be obtained; however, measurements should be made in as short a time as possible. Unless one has high confidence in weather stability, it is doubtful whether readings from different days can be combined without introducing a large error from genotype × environment interaction. Based on empirical comparisons under our conditions, CTD data from days in which mean solar irradiance was <500 w m^{-2} or mean wind speed was >4 m per s were unsuitable for estimating yield or ranking genotypes. In this study, positive correlation among CTD, chlorophyll content, YS, YP, TOL, MP and grain yield showed that CTD and chlorophyll content can be favourite indices in plant breeding. Finally, our data suggest that it is important that measurements are made in as little time as possible to reduce potentially large errors from a changing environment. In our experience, the traditional handheld infrared thermometer (IRT) is not well suited to this requirement.

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