

Original Research Article

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Individual Heat and Combined Heat Drought Stresses in Wheat: Variation in NDVI and Canopy Temperature

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ABSTRACT

The present study was conducted to determine genotypic variations for normalized difference vegetation index (NDVI) and canopy temperature (CT) in wheat under terminal heat and combined heat and drought stresses. Forty-four wheat genotypes were evaluated under late sown with irrigation (heat stress) and without irrigation (combined heat and drought stress) in an environment with average temperatures of 28.6°C during the growing season and 30.62°C during the grain filling stage. Moisture under heat stress range from 14.84 to 19.12 % whereas under combined stress it was 5.08 to 10.49 %. The mean grain yield was 106.7 and 123.4 gm/ plot under heat stress and combined stress respectively. The mean CT 24.1 and 29.4°C was under heat stress and combined stress respectively. The mean NDVI 0.57 and 0.50 under heat stress and combined stress respectively. Overall, under combined stress there was a 13 % decrease in NDVI, 18% increase in the CT and 13.5% increase in the yield than the heat stressed genotypes. The negative correlation between NDVI and yield whereas positive correlation between CT and grain yield under combined stress. The NDVI and CT are the two physiological traits to measure high yielding genotypes under combined stress.

Keywords

Heat stress, Combined stress, NDVI, Canopy temperature

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Introduction

Wheat production is mostly limited by water availability, high temperature and combined heat and drought stresses. As a result of climate change, the frequency and intensity of drought and high temperatures are projected to increase on a global scale in the next decades. Negative changes are expected to be larger in lowland tropical regions (Rosenzweig *et al.*,

2014 and Porter *et al.*, 2014). A large portion of such losses can be avoided through crop improvement. The association between heat and drought stress is poorly understood. Drought stress is often a combination of low water availability and increased temperature resulting from reduced transpirational cooling under limited water conditions. However, there is evidence that the response to drought stress at elevated ambient temperatures is

unique and cannot be extrapolated from the sum of the effects of both stresses (Rizhsky, Liang, and Mittler 2002, 2004; Barnabas, Jaeger, and Feher 2008; Cairns *et al.*, 2013). Three traits that can potentially be used in a wheat improvement program for tolerance to combined stress of heat and drought are canopy temperature and the normalized differential vegetation index (NDVI) and grain yield. Using selection indices like as CT and NDVI in conjunction with GY, will improve selection gains and increase cost efficiency of breeding programs (Neiff *et al.*, 2015).

Canopy temperature (CT) is a useful indicator of crop water status (Jackson *et al.*, 1977) and has potential as a tool for indirect selection of genotypes tolerant to drought and heat stressed environments (Reynolds *et al.*, 2009). Developing cultivars with improved adaptation to drought and heat stressed environments is a priority for plant breeders. Canopy temperature (CT) is a useful tool for phenotypic selection of tolerant genotypes, as it integrates many physiological responses into a single low-cost measurement. Canopy temperature strongly depends on stomatal conductance and the plant's access to water. This makes canopy temperature a potential trait for indirect selection of maize germplasm for improved drought resistance (Garrity and O'Toole 1995). Different vegetative indices have the potential to be used to address wheat improvement; (i) the Normalized Difference Vegetative Index (NDVI) is the ratio between distinctive reflectance characteristics of the crop canopy in the red and near-infrared (NIR) region of the spectrum (Henik *et al.*, 2012).

Grain yield is the most important trait for selection in crop improvement programs. Heritability for grain yield under stress is typically lower than that under optimal conditions, reducing genetic gain from selection under abiotic stress conditions (Weber *et al.*, 2012). The specific objectives

of this study were to identify vegetative indices measured with high throughput devices that could potentially be used as alternative to visual scores taken by breeders.

Materials and Methods

Wheat germplasm and experimental design

The field experiment was carried at new area of ICAR-Indian Agricultural Research Institute, New Delhi, India (28°41' North latitude and 77°13' East latitude, 228 m above mean sea level) Material for the study comprised of 44 genotypes of wheat were evaluated under late sowing with irrigation (heat stress) and without irrigation (combined heat & drought stress) during 2016-17. The experiment was laid out in alpha lattice design with two replications in heat stress and combined stress conditions. The average temperatures of 28.6° C during the growing season and 30.2° C during the flowering period, with temperatures ranging from 24° C at night to 46° C at midday during flowering.

The soil moisture, temperature and precipitation during flowering to maturity stage are shown in Figure 1 and 2. Each genotype was planted with manually with gross plot size of 0.46 × 2.5 m, with rows at 23 cm apart. The experiment was carried out under well-watered and water-deficit conditions. The date of sowing in the month of December 8th, 2016 which was late or delayed sown in order to get terminal heat stress.

In the well-watered treatment (subsequently referred to as heat-stress treatment), plants were continuously irrigated throughout the cropping season. In the drought-deficit treatment (subsequently referred to as combined heat and drought stress treatment). The standard cultivation practices prescribed for wheat under irrigated conditions were followed precisely.

Measurements and analyses

Soil moisture measurements

Soil samples from each experimental unit were collected in aluminium boxes with secure lids every week interval at three depths (15, 30 and 45 cm) by using augers. The samples were weighed immediately and then oven dried at 105 °C for 72 h for determining soil moisture content by gravimetric method and it has correlated with neutron moisture meter readings (Fig. 1).

Weather parameters

The different weather parameters during wheat crop growing season for the period 2016–2017 measured were daily maximum temperature (°C), minimum temperature (°C), RH maximum (%), RH minimum (%), rain (mm), average wind speed (AWS), evapotranspiration and sunshine hours (Fig. 3)

NDVI and canopy temperature measurements

The NDVI measurements were taken three times in season with different crop growth stages viz., anthesis, grain filling, and maturity using a hand-held Ntech 'Greenseeker' (Field portable NDVI sensor). The NDVI was calculated based on wavelengths measured with the multispectral sensor. The canopy temperature was recorded during crop grain filling stage on all replications using the Sixth Sense LT300 IRT (Hand-held IRT, Mikron M90, Mikron Infrared Instrument Company Inc., Oakland, NJ, USA). Two NDVI and CT measurements were taken per plot, one on each bed. The two measurements were then averaged to obtain one final reading per plot. All NDVI and CT measurements were taken at midday (12 pm–2 pm) on clear, sunny days with minimal wind when the plant is most water stressed.

Grain yield (gm/plot) was determined on whole plots by combine harvesting when grains were dry at about 4%–5% moisture, and weighing the grains.

Result and Discussion

Effect of soil moisture on canopy temperature

Canopy temperature for all the forty-four genotypes was recorded throughout the growing period in delayed sown well-watered (heat stress) as well as delayed sown water stress (combined heat and drought stress) conditions. Area under curve plotted with minimum temperature observed in the canopy vs days after anthesis (DAA) indicated that canopy temperature did not vary across the genotypes. However, soil moisture deficit did not significantly affect the canopy temperature of the genotypes Duram and GW 322. In Westonia and HD 2864 genotypes canopy temperature was higher under combined stress plots.

The mean soil moisture in the heat and combined stress plots were 16.87 and 7.78 (%) respectively. The range of soil moisture in heat stress plots were 14.84 to 19.12 (%) during maturity and anthesis stage respectively. The range of soil moisture in combined stress plots were 05.08 to 10.49 (%) during maturity and anthesis stage respectively (Fig. 1).

The mean NDVI in heat stress and combined stress genotypes were 0.57 and 0.50 respectively. The NDVI in the heat stress genotypes was higher compare to the combined stress. The 13 % higher NDVI was observed in the heat stress genotypes (Fig. 4). The mean CT in heat stress and combined stress genotypes were 24.11⁰C and 29.47⁰C respectively. The CT in the heat stress genotypes was lower compare to the combined stress. The 18 % lower CT was observed in

the heat stress genotypes (Fig. 5). The mean grain yield per plot in heat stress and combined stress genotypes were 106.7 and 123.4 gm/plot respectively. The grain yield in the heat stress genotypes was lower compare to the combined stress. The 13.5 % lower grain yield was observed in the heat stress genotypes (Fig. 6). The difference in the NDVI readings taken at the anthesis and the maturity stage are negatively correlated with the final grain yield of heat stress genotypes (Fig. 7 and 8).

The normalized differential vegetation index

The difference in the NDVI readings under heat stress at anthesis and maturity stage genotypes Viz., Duram, GCP 36, DBW 43, HD 2985 and HD 2932 were lower with 0.12, 0.13, 0.16, 0.17 and 0.18 respectively. The difference in the NDVI readings under heat stress at anthesis and maturity stage genotypes Viz., NP 4, GCP 16, HD 2329, Hindi 62 and GCP 1 were higher with 0.30, 0.30, 0.30, 0.34 and 0.35 respectively. The difference in the NDVI readings under combined stress at anthesis and maturity stage genotypes Viz., C 306, Chiriya 7, Kalyan sona, Raj 3765 and DBW 43 were lower with 0.20, 0.23, 0.23, 0.23 and 0.24 respectively. The difference in the NDVI readings under combined stress at anthesis and maturity stage genotypes Viz., PBW 550, WH 730, HD 2329, Westonia and HD 2851 NDVI were lower with 0.37, 0.38, 0.39, 0.39 and 0.39 respectively. The NDVI was higher under the heat stress than the combined stress (Fig. 4).

The canopy temperature

The genotypes were showing lower CT value under heat stress Viz., GCP 16, NP 818, GCP 2, GW 322 and HD 3086 with 22.3, 22.4, 22.4, 22.5 and 22.6⁰C respectively. The genotypes were showing lower CT value under combined stress Viz., Duram, GW 322,

NP 818, C 306 and HD 2932 with 27.1, 27.2, 27.5, 27.9 and 27.9⁰C respectively. The genotypes were showing higher CT value under heat stress Viz., WR 544, Westonia, Babax, HD 2733 and Chiriya 7 with 26.1, 26.1, 26.2, 26.3 and 27.0⁰C respectively. The genotypes were showing higher CT value under combined stress Viz., Kundan, GCP 30, Babax, HD 2864 and Westonia with 30.7, 31.3, 31.3, 31.8 and 31.8⁰C respectively. The Canopy temperature was higher under the combined stress than the heat stress (Fig. 5).

The grain yield

The genotypes were showing lower grain yield under heat stress Viz., GCP 30, GCP 16, GCP 23, Westonia and GCP 2 with 35.2, 36.5, 37.2, 38.7 and 45.5 gm/ plot respectively.

The genotypes were showing lower grain yield value under combined stress Viz., NP 818, GCP 16, GCP 30, GCP 23 and Westonia are 53.5, 67.6, 71.6, 72.1 and 88.7 gm/ plot respectively.

The genotypes were showing higher grain yield value under heat stress Viz., Chiriya 3, GCP 6, HI 1544, GW 322 and HD 2733 with 171.2, 172.2, 180.1, 182.5 and 195.7 gm/ plot respectively.

The genotypes were showing higher grain yield value under combined stress Viz., HD 2987, GCP 6, Raj 3765, HD 3086 and Chiriya 7 with 168.0, 172.2, 172.5, 173.5 and 175.5 gm/ plot respectively. The grain yield was higher under the combined stress than the heat stress (Fig. 6).

Statistical analysis

All statistical analyses were done using procedures in SAS v9.3 (SAS Institute Inc., Cary, NC, USA) with each treatment (heat and combined stress) analysed separately (Table 1).

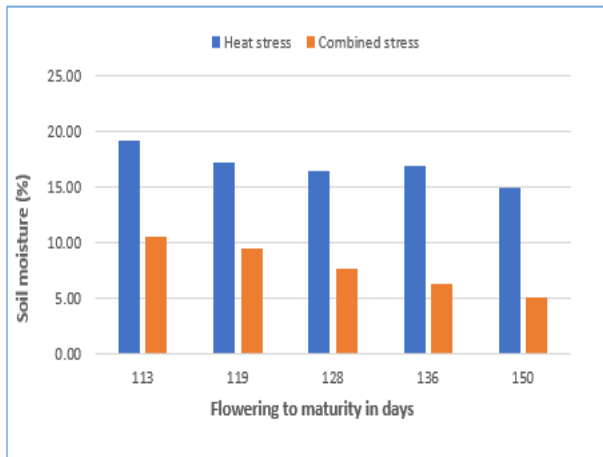


Fig. 1 The soil moisture measured in the wheat season flowering to maturity (days)

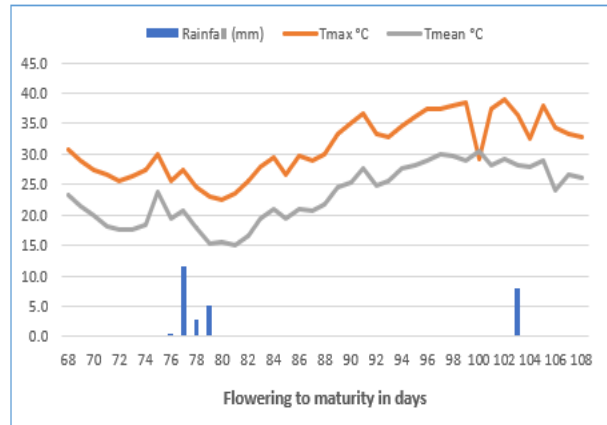


Fig. 2 Daily mean (light line) and maximum temperature (dark line) and precipitation (bars) measured throughout the wheat flowering to maturity (days)

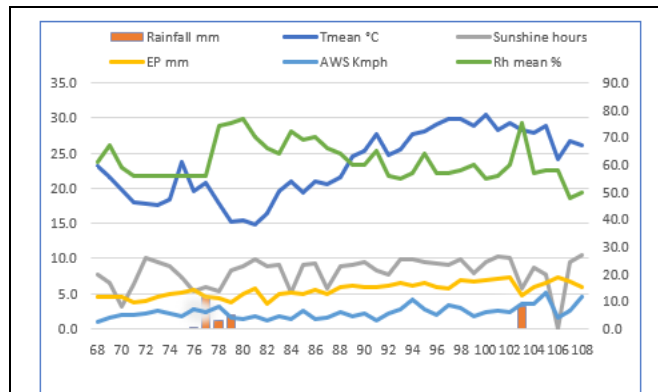


Fig. 3 Different weather parameters during wheat crop growing season for the period 2016-2017

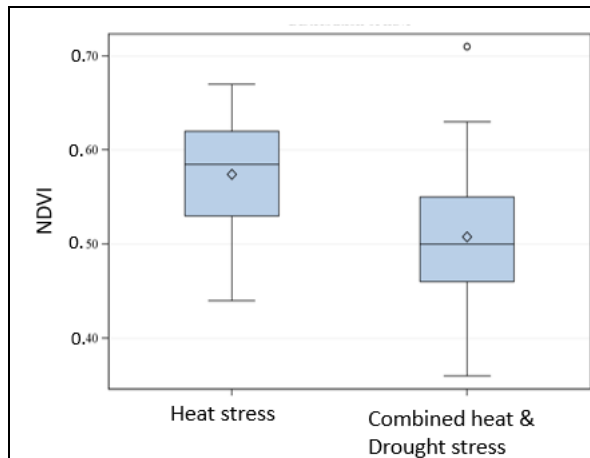


Fig. 4 NDVI under both heat and combined stress.

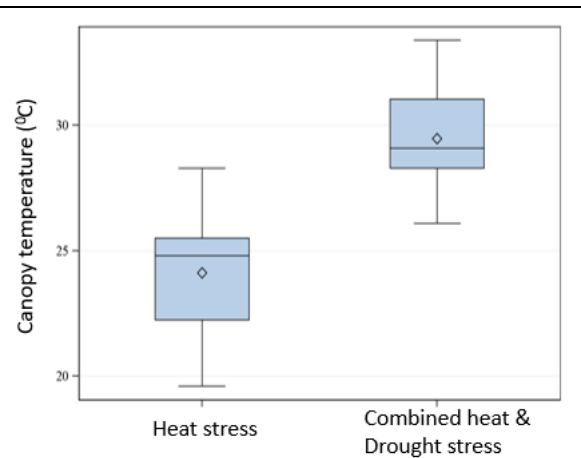


Fig. 5 Canopy temperature under both heat and combined stress.

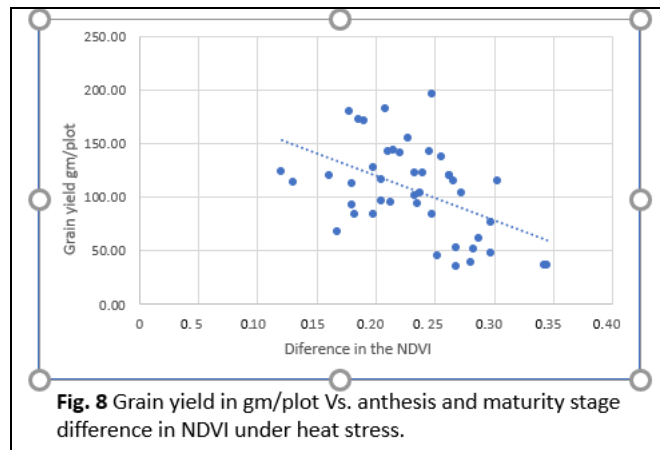
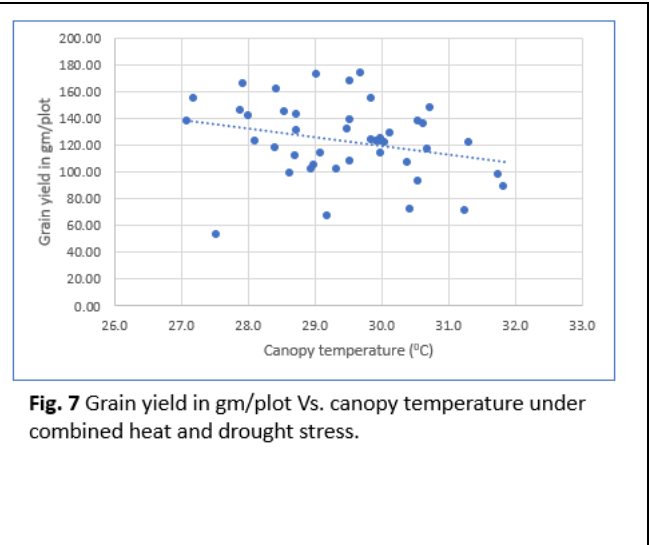
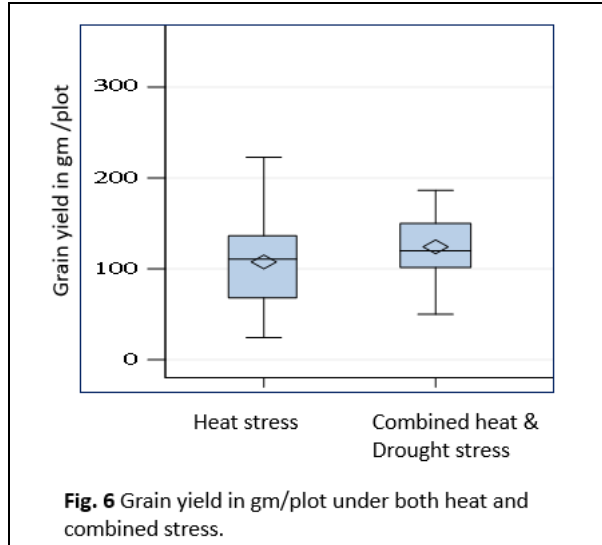


Table.1 The mean NDVI, canopy temperature, grain yield and soil moisture under heat and combined stress

Condition	NDVI	CT (°C)	Yield (gm/plot)	Soil moisture (%)
Heat stress	0.57	24.11	106.71	16.87
Combined heat & drought stress	0.50	29.47	123.43	7.78

Analysis of variance using PROC MIXED was carried on the forty-four genotypes to determine sources of variation within and across trials and determine if genotype by trial interaction was present for the traits measured. All effects in the model (rep, incomplete block, trial, entry x trial) with the exception of entries were treated as random.

Data from all trials was then normalized relative to the performance of Roelfsf2007 and used for further analysis of all entries across trials, similar to methods used to analyse historical trial data over years with a recurrent check (Graybosch and Peterson, 2010). PROC CORR was used to determine the phenotypic correlation between relative

means of traits. Regression was performed using PROC REG to determine the relationship between CT and yield within each trial individually and across all trials. Initially, regression was used to determine the ability of CT and NDVI to predict yield. The interactions of days to heading and plant height with CT and NDVI were then fit as covariates (PROC ANCOVA) in the regression model to determine their effect on the yield predictability of CT and NDVI. The normalized differential vegetation index (NDVI) and the canopy temperature (CT) are efficient low-cost measurement of crop water status and soil water status that has potential for phenotypic selection of stress tolerant and susceptible lines. Overall, cooler CT was favourable for higher yield although this association was stronger and more consistent under heat stress.

The lower NDVI and higher canopy temperature under the combined heat and drought stress resulted in higher yield because of plants wanting to complete their reproductive cycle very fast during the combined stress. The higher NDVI, lower canopy temperature, more vegetative growth and reproductive cycle coincided with the terminal heat stress resulted in lesser yield under heat stress compared with the combined stress.

Variation in the coefficient of determination and the slope across individual trials indicates that genetic background plays a definite role in the ability of CT and NDVI to predict yield and thus background information on the parents of a breeding line may help in making decisions on interpreting the results of CT measurements. The most useful implementation of CT and NDVI for genetic improvement would be in the early generations of breeding, such as on progeny rows where yield testing is not performed, although considerations would need to be made for an experimental design that utilizes a repeated check cultivar to adjust for spatial

variation. In the future, high throughput tools for measuring CT and NDVI that increase speed and accuracy will also allow breeders to maximize potential genetic gain from CT and NDVI.

An innovative method to evaluate wheat genotypes under high temperatures and combined heat and drought stress using GreenSeeker and Sixth Sense LT300 IRT. This cost-effective technique allows the phenotyping of large populations in a short time period. The use of these instruments reduces the time needed for the measurement of NDVI using a GreenSeeker (Cairns *et al.*, 2012) or visual scores (White *et al.*, 2012) from 30 seconds per plot to the fraction of a second per plot.

Effects of heat under well-watered conditions vs. restricted water access during grain filling on grain yield

Relative to heat stress carried out under delayed sown in order to get terminal heat stress temperatures, grain yield was increased by 13.6% under combined heat and drought stress than the heat stress. These results are in not agree with results obtained by (Lobell *et al.*, 2011), showing that each degree-day accumulated above 30°C reduced the final yield by 1% under optimal rain-fed conditions, and by 40% under drought-stress conditions. Optimal temperature for tropical maize is around 35°C (Cichino, Rattalino Edreira, and Otegui 2010), whereas average temperatures measured in our study reached up to 45°C under heat stress and 54°C under combined heat and drought stress. Under well-watered conditions, increases in temperature above 45°C have been shown to reduce photosynthesis by 50% (Crafts Brandner and Salvucci 2002), potentially explaining the strong reductions in grain yield under high temperatures than the combined stress observed here.

NDVI and canopy temperature allow identification of tolerant genotypes

The NDVI explained differences between and within treatments, whereas canopy temperature explained differences in grain yield among treatments and genotypes within the combined heat and drought stress treatment. Under combined heat and drought stress, our results showed positive correlations between canopy temperature and grain yield, indicating that genotypes that show higher canopy temperature under combined high temperatures and drought stress yielded higher because of the higher stem reserve mobilization. Canopy temperature has been used to identify germplasm with increased tolerance to high temperatures in wheat (Cossani and Reynolds 2012). The method presented here would therefore allow the identification of genotypes tolerant to conditions encountered in the farmers' fields. The correlation measured between CT and grain yield is reflective of the importance of sufficient assimilate availability for yield formation under combined stress ($r = 0.275$). The negative correlation measured between difference in NDVI and grain yield under combined stress ($r = 286$). In our study, beneficial effects of a high NDVI on grain yield may have been boon under combined heat and drought stress by reduced stem reserve mobilization.

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