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Association of leaf chlorophyll content with the stay-green trait and grain yield in wheat grown under heat stress conditions

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Abstract: Heat stress is a major production constraint of wheat in South Asia, particularly in the Gangetic plains of India and Bangladesh. The leaf chlorophyll status is a key determinant for a high rate of photosynthesis under stress. The present experiments included 238 genotypes in 2016–2017 and 321 genotypes in 2017–2018 under optimum and under heat stress conditions. Subsequently, a set of 100 genotypes selected on basis of the heat susceptibility index was evaluated in 2018–2019 under heat stress conditions to study the relationship between important physiological traits and yield under stress. A significant correlation of soil plant analysis development (SPAD) value of the two upper leaves with stay-green trait and grain yield indicates the importance of chlorophyll content, both in flag and penultimate leaf, in maintaining leaf areas under greenness (LAUG) and grain yield under heat stress. The SPAD in the flag and penultimate leaf was responsible for 8.8% and 10.9%, respectively, of the variation in grain yield. For the stay-green trait, 8.4% and 7.2 % of the variation was governed by the SPAD value in the flag and penultimate leaf, respectively. These results suggest that, in addition to the flag leaf, the chlorophyll status of the penultimate leaf can be an important criterion for the selection of superior wheat genotypes under heat stress. The genotypes SW-139; SW 108; DWR-F8-35-9-1; NHP-F8-130; DWR-F8-3-1 that maintained a high chlorophyll content in the flag and penultimate leaf can be used further in breeding programmes addressing heat resistance in wheat.

Keywords: abiotic stress; chlorophyll stability; leaf greenness; SPAD value

Extreme temperature fluctuations during critical growth stages (like anthesis and the grain filling stage) causes serious yield losses in most of the wheat producing areas (Balla et al. 2019). Wheat growing areas in the eastern Gangetic plains of India and Bangladesh are affected by heat stress and, as a result, the average wheat productivity of the north eastern plain zone of India is much lower than the productivity of the north western plain zone. Terminal heat stress under late sown conditions has been reported to cause up to a ~45% yield reduction in wheat (Joshi et al. 2007b). High-temperature stress causes several morphological and physiological changes in the plant. The heat stress directly affects the photosystem II and enzymatic activity of Rubisco that reduces the photosynthetic activity in the leaves.

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An increased photosynthetic activity is associated with an increased biomass production and grain yield (Brestic et al. 2018). The negative impact of terminal heat stress due to delayed sowing was observed on the grain yield, biological yield, and thousand kernel weights (Moshatati et al. 2017). Heat stress accelerates the rate of leaf senescence, shortens the grain filling duration, leads to a reduction in the biomass, seed size and grain yield (Reynolds et al. 2000; Kumari et al. 2013). A strong correlation of the leaf chlorophyll content and grain yield under heat stress was recorded, suggesting the use of leaf chlorophyll content for screening genotypes under heat stress (Reynolds et al. 2000; Rosyara et al. 2010; Lopes et al. 2012).

Stay-green (SG) has been reported to be an important yield determining parameter under abiotic and biotic stress in wheat (Joshi et al 2007a; Vijayalakshmi et al. 2010; Kumari et al. 2013). The leaf area under greenness (LAUG) is used as a measure of the SG trait in wheat, based on the proportion of green areas in the flag leaf and spike (Joshi et al. 2007a). High heritability was recorded for the SG, indicating the chances of selection for suitable genotypes and further improvement (Joshi et al. 2007a; Kumari et al. 2013). SG cultivars reveal high photosynthetic activity and provide higher longevity during grain filling (Chen et al. 2010). SG lines contribute more photosynthates towards grain development than the non-stay-green lines (Reynolds 2002). Therefore, under late sown conditions, SG lines become capable of maintaining a higher LAUG that increases the grain filling duration, thousand kernel weights and yield (Kumari et al. 2013).

The soil plant analysis development (SPAD) value is often used for the indirect estimation of the leaf chlorophyll content. A strong positive correlation of the SPAD value and leaf chlorophyll content has been obtained in wheat (Reeves et al. 1993), rice (Turner & Jund 1991) and maize (Zotarelli et al. 2003). Leaf SPAD units have shown a linear correlation with the leaf chlorophyll content and photosynthetic rate (Netto et al. 2005). A positive correlation of the SPAD value with the grain yield under optimum and heat stress conditions was observed (Narendra et al. 2021). In most of the cases, the flag leaf has been used for determining the SPAD reading, photosynthetic activity and stomatal conductance (Reynolds et al. 1994; Paliwal et al. 2012; Kumari et al. 2013; Islam et al. 2014). It has been reported that the flag leaf contributes ~30–50% of the grain assimilates in wheat (Sylvester-Bradley et al. 1990). However, the role of the penultimate leaf (i.e., the leaf next to the flag leaf from the top) during the grain formation under heat stress has not been studied in detail. We hypothesised that the penultimate leaf also contribute significantly to increasing the grain yield and longevity of the plant under stress. Therefore, in the present study, the effect of heat stress on the important yield attributing parameters has been analysed. Furthermore, the relationship of the leaf chlorophyll content in the flag and penultimate leaves with the grain yield and SG under heat stress conditions was studied.

MATERIAL AND METHODS

Plant material. A total of 513 wheat genotypes (including checks) comprised of advanced breeding lines, genotypes selected from different national and international nurseries (SSN – segregating stock nurseries; NHP – National Hybridization Programme; SAWYT – Semi-arid Bread Wheat Yield Trial; W × S – Winter × Spring hybridisation nurseries and HPYT – Harvest Plus Yield Trial) and released varieties (Table S1 in the electronic supplementary material (ESM)) were evaluated under field conditions in the research farm of Bihar Agricultural University, Sabour, India.

Experimental layout. The experiments were carried out with 238 genotypes in 2016–2017 and 321 genotypes in 2017–2018 under optimum and heat stress (late sown) conditions. Sowing was performed using a seed drill on November 25, 2016 and November 24, 2017 for the optimum; January 02, 2017 and January 01, 2018 for the heat stress condition with 6 rows of 4 m length per plot having row to row distance of 20 cm. Forty-six tolerant genotypes identified in the first year were repeated for the evaluation in the second year. Five flood irrigations were scheduled at the crown root initiation (CRI) stage (after 21 days of sowing), maximum tillering stage [Zadok's growth stage (GS) 32], booting stage (GS 45), milk development stage (GS 73), and dough development stage (GS 85). There were 5 rainy days in 2016–2017 (0.6 to 12.4 mm) and 2 rainy days in 2017–2018 (6.6 to 24.2 mm) (Table S2 in the ESM). The soil moisture percentage was recorded from the weights of the fresh and oven-dried soil samples taken from a 5 cm soil depth of each block during the CRI, anthesis and physiological maturity stages of the crop growth (Table S3 in the ESM). Standard agronomic practices were carried out time to time to raise a healthy crop. Fertilisers @ 150 : 60 : 40 kg/ha, $N:P_2O_5:K_2O$ in the optimum and @ 120 : 60 : 40 kg/ha, $N:P_2O_5:K_2O$ in the heat stress conditions were applied. The genotypes were evaluated for the grain yield (GY), number of tillers (NT), biomass (dry weight of the plant) at maturity, panicle length (PL) of the main tiller and thousand kernel weight (TKW). The heat susceptibility index (HSI) was estimated following Fischer and Maurer (1978) using the formula:

$$HSI = (1 - Y_s/Y_p)/(1 - \bar{Y}_s/\bar{Y}_p)$$

where:

Y_s, Y_p – the yield of genotypes evaluated under the stress and optimum conditions, respectively;

$\bar{Y}_s$, $\bar{Y}_p$ – the mean yield of the overall genotypes evaluated under the stress and optimum conditions, respectively.

Another experiment in the year 2018–2019 was carried out under heat stress conditions using 100 genotypes (including 6 checks) selected using the HSI from the previous two years of experiments. Sowing was undertaken on December 30, 2018 in a randomised complete block design with 3 replications, 2 m row lengths and 3 rows per plot with 20 cm row to row spacing. The irrigation schedule and fertiliser doses were kept the same as the previous years' experiments under the stress conditions. There were seven rainy days in 2018–2019 with the rainfall of 0.6 to 34 mm (Table S2 in the ESM). In this experiment, observations were recorded for the grain yield, plant height, days to heading, panicle length, canopy temperature and SPAD value as a determinant of the chlorophyll content in the flag (F) and penultimate (F-1) leaf, biomass and LAUG as a measure of the stay-green trait.

The canopy temperature was measured for two consecutive days using a handheld infrared thermometer (FLUKE 62 Mini IR Thermometer, FLUKE, China) at the end of the anthesis on fully, sunny days from 12.00 to 14.00 h. Readings were avoided during foggy weather and 2–3 days after irrigation or rain.

The SPAD value was measured using a SPAD-502 (Minolta, Japan) in the flag (F) leaf and penultimate (F-1) leaf at the top, middle and bottom and the average of these readings was considered for each leaf. Based on the SPAD value, the genotypes were classified into low (< 40); intermediate (40–50) and high (> 50) categories.

The stay-green trait was measured as the LAUG in the flag leaf and spike after the late dough stage (GS 77) at 4 day intervals using the procedure described by Joshi et al. (2007a).

Environmental parameters. The weekly maximum and minimum temperatures during the crop growing period were recorded at the university weather station (Figure 1). The average maximum temperature during the growing period of the crop under the optimum conditions was 26.9 °C and was 27.7 °C for the heat stress conditions while the average minimum temperature for the optimum conditions was 12.1 °C and was 13.03 °C for the heat stress conditions.

Statistical analyses. The combined analyses of variances (ANOVAs) were calculated for the year 2016–2017 and 2017–2018 for each trait to determine the genetic variances using the statistical software OPSTAT (http://14.139.232.166/opstat/). The histogram analysis for the heat susceptibility index was

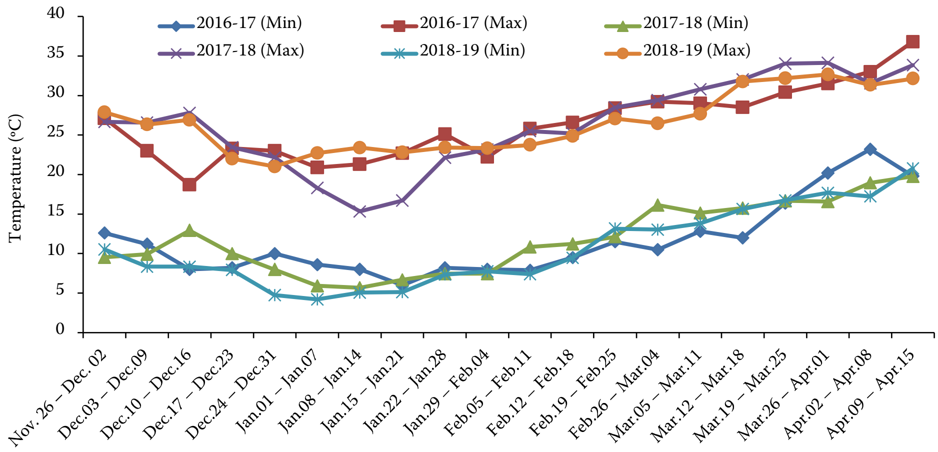

Figure 1. Weekly maximum and minimum temperatures throughout the growing period in 2016–2017, 2017–2018 and 2018–2019

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computed using software R (Ver. 4.0.0). The analysis of the mean, standard error, pair wise correlation and regression was determined for each trait in the year 2018–2019 for the efficient selection of superior genotypes. The sample means were tested using the function two sample *t*-test in the statistical software WASP (Web Agri Stat Package) Ver. 2.0 (https://ccari.res.in/wasp2.0/index.php). The heritability was estimated following the formula:

$$H^2 = \sigma_g^2/(\sigma_g^2 + \sigma_{g\times y}^2/y + \sigma_e^2/ry)$$

where:
σ_g^2 – the genetic variance;
$\sigma_{g\times y}^2$ – the genotype-by-year interaction;
σ_e^2 – the error variance;
y – the number of years;
r – the number of replications (Narendra et al. 2021).

RESULTS

Effect of heat stress on yield and contributing traits. The mean temperature differences between the months of February and March indicate a hike in the maximum and minimum temperature at the time of flowering and grain filling stages. The relative increase in the minimum night temperature (ranging from 5.3 °C in 2017–2018 to 5.9 °C in 2016–2017) was higher than the maximum day temperature (3.4 °C in 2016–2017 to 6.3 °C in 2017–2018). A significant variation in all the traits evaluated under optimum and heat stress conditions indicates the presence of genetic differences among the genotypes. The genotype × environmental interaction for the grain yield and biomass was significant. All the traits were affected by the heat stress; but the maximum reduction was recorded for the biomass and grain yield (in the year 2016–2017) and the number of tillers, grain yield and biomass (in the year 2017–2018) (Table 1). The panicle length was the least affected trait under the heat stress.

A sufficiently large variation for the HSI was recorded among the entries evaluated in 2016–2017 and 2017–2018. The frequency distribution of the test entries based on the HSI was found to be skewed to some extent, but more numbers of the genotypes (179) as being tolerant to moderately tolerant were recorded in the year 2017–2018 (Figure S1 in the ESM). In the year 2016–2017, forty-six entries were identified as tolerant to moderately tolerant. When these entries were re-evaluated in the year 2017–2018, twenty-five genotypes out of the forty-six entries were found to be tolerant.

The variations in all the traits were found to be highly significant when grown under the heat stress conditions in the year 2018–2019 (Table S4 in the ESM). A high heritability was recorded for the biomass, canopy temperature and grain yield (Table 2). Moderate to high heritability for the SPAD value in the F and F-1 leaf, stay-green trait and number of tillers, which advocates the scope for the selection of suitable genotypes. The genotypes SSN-F8-1433-1, DWR-F9-98-2, SW-108 and DWR-F8-19-7 for the grain yield; SW-139, SW 108 and DWR-F8-35-9-1 for the high chlorophyll content in the flag leaf; NHP-F8-130, SW-139, DWR-F8-3-1 and SW-138 for the high chlorophyll content in the penultimate leaf; SW-152; SW-508; W×S-F8-10-1 and HPYT-430 for the low canopy temperature were identified.

Effect of heat stress on leaf chlorophyll content and its relationship with other physiological pa-

Table 1. Effect of heat stress on the traits evaluated under optimum and heat stress conditions

Year	Growing environment	Tillers per plant	Biomass (g/plant)	Panicle length (cm)	Thousand kernel weight (g)	Grain yield (q/ha)
2016–2017	optimum	6.32**	22.61**	9.65*	35.55**	42.9**
	heat stress	3.9**	9.12**	8.95*	24.72**	17.63**
	percent of reduction	38.29	59.66	7.25	30.46	58.90
2017–2018	optimum	6.9**	20.8**	9.9	33.68**	33.15**
	heat stress	4.2**	14.5*	8.6*	26.19**	22.33**
	percent of reduction	39.13	30.29	13.13	22.24	32.64
	genotypes	1.156*	34.91**	1.38*	10.29*	51.74**
	environments	73.57**	4 279.38**	25.37**	1 652.74**	6 952.12**
	genotypes × environment	1.68	20.81**	0.38	10.63	18.2*

*, **Significant at $P < 0.05$ and 0.01, respectively, in the two sample *t*-test

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Table 2. Identification of the trait specific genotypes suitable for heat stress

Traits	Mean ± SE	Range	Genotypes identified	Heritability
Days to heading (days after sowing)	71.2 ± 1.13*	63–81	W X S-F8-8-2, SW-502, SW-115, SW-310, SW-457, SAWYT-29, SSN-F7-61-5, SW-164 (< 70 days after sowing)	50.261
Biomass (g/plant)	12.02 ± 1.13**	4.66–27.75	SW-160, SW-362, SW-268, HPYT-446, SW-404 (> 20 g/plant)	67.47
Canopy temperature (°C)	25.72 ± 0.46**	23.2–29.5	SW-152; SW-508; WxS-F8-10-1, HPYT-430; DWR-F8-35-12; SW-515 (< 24 °C)	66.89
SPAD value (flag leaf)	46.6 ± 1.15**	35.9–57.2	SW-139; SW 108; DWR-F8-35-9-1; NHP-F8-130; DWR-F8-3-1 (> 54.0)	54.44
SPAD value (penultimate leaf)	42.8 ± 1.09**	30–53.5	NHP-F8-130, SW-139, DWR-F8-3-1, SW-138, (> 50.0)	51.42
Stay green (LAUG)	17.43 ± 1.87**	–12 to 59	NHP-F8-85-1, SW-138, DWR-F8-19-7, SAWYT-4, (> 50)	56.02
Grain yield (q/ha)	26.33 ± 0.27**	16.25–37.42	DWR-F9-98-2, W X S-F8-27-2, SW-108, HPYT-446, DWR-F8-19-7, SSN-F8-1433-1, DWR-F8-35-13 and W X S F8-11-4 (> 33.0 q/ha)	61.6

*, **Significant at $P < 0.05$ and 0.01, respectively, in the ANOVA; SPAD – soil plant analysis development; LAUG – leaf areas under greenness

rameters. A significant genetic variation for the SPAD value in the F and F-1 leaf was recorded. The range of the SPAD value at anthesis under heat stress in the F leaf was 35.9 to 57.2 and was from 30.0 to 53.5 in the F-1 leaf (Table 2). A higher SPAD reading in the F leaf was observed than the F-1 leaf; the genotypes with the higher chlorophyll content in the flag leaf also maintained a high chlorophyll content in the penultimate leaf. Classification of the genotypes using the SPAD reading under the heat stress revealed that 21% and 4% of the studied genotypes carried a higher chlorophyll content in the flag leaf and penultimate leaf, respectively (Table 3). Most of the genotypes contained an intermediate level of chlorophyll in the flag and penultimate leaves. A significant difference in the stay-green score was observed for all the three classes while, for the grain yield, the low and intermediate class also differed significantly.

A highly significant association of the SPAD in the flag leaf was recorded with the days to heading, canopy temperature, stay-green (as measured through the LAUG parameter), and grain yield. Similarly, the association of the SPAD in the penultimate leaf was also significant with the days to heading, plant height, canopy temperature, panicle length, stay-green and grain yield (Table 4). The R^2 value indicates that the SPAD in the penultimate leaf explains 10.9% of the grain yield variation while, in the flag leaf, it explained 8.8% of the total genetic variation (Figure 2). For the stay-green, 8.4% of the variation was explained by the SPAD in the flag leaf and 7.2% of the variation

Table 3. Classification of 100 genotypes using the soil plant analysis development (SPAD) in the flag leaf and penultimate leaf

Classes	No. of genotypes	SPAD in flag leaf	Mean stay-green score	Mean yield (q/ha)
Low (< 40)	3	37.83**	5.33	22.611
Intermediate (40–50)	76	45.47**	16.11**	26.09**
High (> 50)	21	51.96**	23.95**	27.75**
	No. of genotypes	SPAD in penultimate leaf	mean stay-green score	mean yield (q/ha)
Low (< 40)	28	36.86**	10.86	23.79**
Intermediate (40–50)	68	44.23**	19.46*	27.27**
High (> 50)	4	52.33**	29.00*	28.15**

*, **Significant at $P < 0.05$ and 0.01, respectively, in the two sample t-test

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Table 4. Pearson correlation coefficient among the traits under the heat stress conditions

	Plant height	Days to heading	Panicle length	Canopy temperature	SPAD (flag leaf)	SPAD (F-1 leaf)	Biomass	Stay-green	Grain yield
Plant height	1	0.156[NS]	0.480**	0.242*	0.183[NS]	0.281**	0.250*	0.099[NS]	0.333**
Days to heading		1	0.194[NS]	–0.227*	0.236*	0.225*	0.283**	0.280**	0.025[NS]
Panicle length			1	0.192[NS]	0.163[NS]	0.201*	0.324**	–0.024[NS]	0.127[NS]
Canopy temperature				1	0.271**	0.306**	0.070[NS]	–0.029[NS]	0.221*
SPAD (flag leaf)					1	0.760**	0.186[NS]	0.291**	0.297**
SPAD (F-1 leaf)						1	0.143[NS]	0.270**	0.331**
Biomass							1	0.094[NS]	0.287**
Stay-green								1	0.191[NS]
Grain yield									1

*, **Significant at $P < 0.05$ and 0.01, respectively; SPAD – soil plant analysis development; F-1 leaf – penultimate leaf; NS – not significant

was explained in the penultimate leaf. A highly significant linear correlation between the SPAD in the flag leaf and in the penultimate leaf was observed. The R^2 value indicates 57.7% of the variation of the SPAD in the flag leaf, which is explained by the same in penultimate leaf (Figure 3). Nearly, 58% and 68% of the genotypes with a high SPAD value in the flag and penultimate leaf, respectively, also showed a higher grain yield than the average.

DISCUSSION

In this study, the post-anthesis heat stress severely affected the grain yield, biomass, TKW and tillers, which is in agreement with previous studies (Joshi et al. 2007b; Rosyara et al. 2010). Heat stress reduces the dry matter accumulation in the vegetative plant parts and, subsequently, into the kernels. To cope with the loss under heat stress, plants increase the

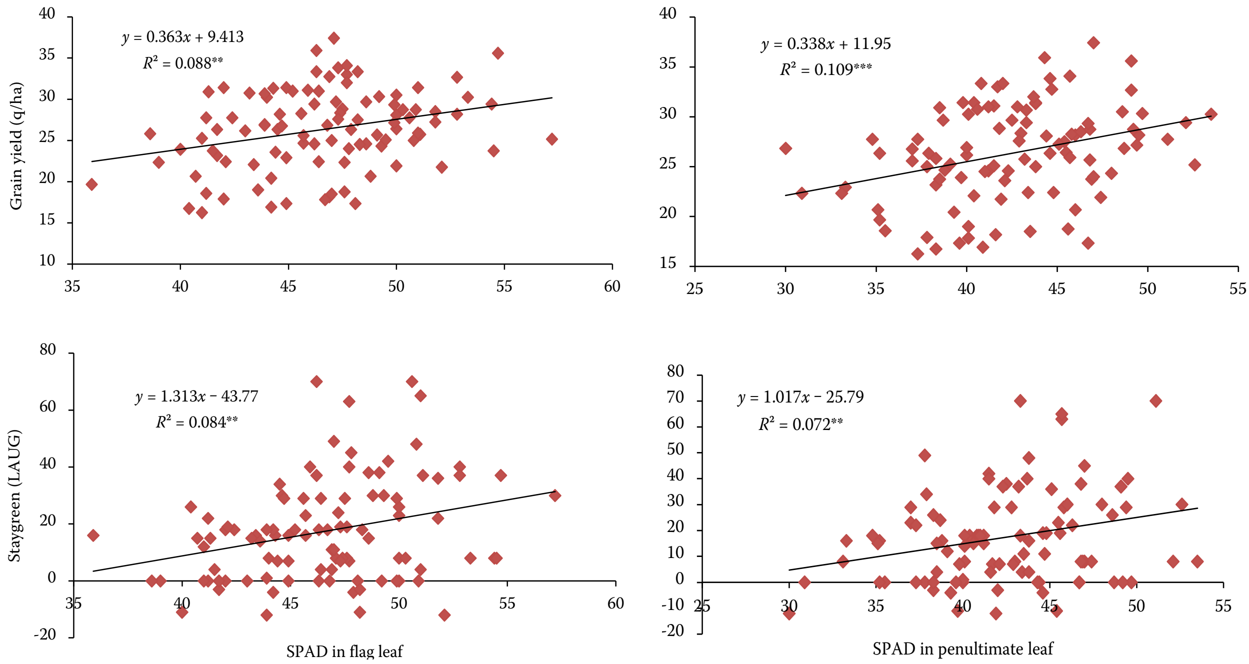

Figure 2. Relationship of the soil plant analysis development (SPAD) in the flag and penultimate leaf with the grain yield and stay green trait

**, ***Significant at 0.01 and 0.001, respectively

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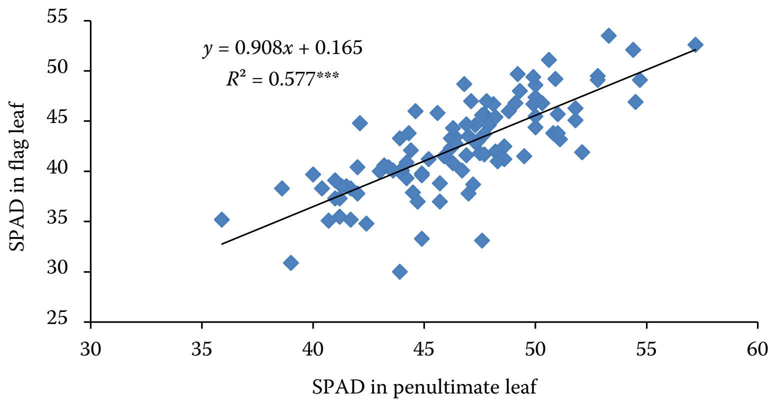

Figure 3. Relationship of the soil plant analysis development (SPAD) in the flag leaf with the penultimate leaf
***Significant at 0.001

rate of the translocation of the photosynthates into kernels from the flag leaves and reserve carbohydrates in the stem (Plaut et al. 2004). Wardlaw et al. (1989) proposed that a per unit increase in the temperature from the optimum at the time of grain filling stage causes a 3–4% yield reduction. In our case, there was a drastic increase in the maximum and minimum temperature during the grain filling stage. Almost every year, the maximum temperature at anthesis and the grain filling stages both in normal and heat stress conditions exceed the optimum temperature limit, i.e., 23 °C and 21.3 ± 2.17 °C as documented earlier for the respective growth stages (Farooq et al. 2011). The relative increase in the minimum night temperature than the maximum day temperature was high. An increase in either the day or night temperature at the time of anthesis or grain filling may cause a yield loss. In previous reports, high night temperatures, high day temperatures and high night and day temperatures at post-anthesis decreased the grain yield, seed setting, leaf photosynthesis, antioxidants, and photochemical activities in wheat (Prasad et al. 2008; Narayanan et al. 2014). The increasing trend of the average night temperatures during March in South Asia was earlier reported, reducing the thousand kernel weight and enhancing the spot blotch disease incidence (Sharma et al. 2007). Undoubtedly, high night temperatures would affect the physiological activity in plants, enhancing the dark respiration causing early senescence. Therefore, an increase in the thermo-tolerance can provide a higher chlorophyll stability. A variation among the wheat genotypes for photosynthetic thermostability has been observed; also, modern wheat cultivars were found to be more tolerant and photosynthetically more active under high temperature stress (Brestic et al. 2012, 2018).

In the present study, a higher SPAD reading was observed in the flag leaf than the penultimate leaf, indicating the enhanced proportion of chlorophyll in the flag leaf. However, the coefficient of variations for the SPAD value in the flag leaf and penultimate leaf were in the same range. A significant correlation of the SPAD reading with the grain yield is in unison with the previous findings (Rosyara 2010; Balla et al. 2019; Narendra et al. 2021). Moderate heritability and large variability of the SPAD in the flag and penultimate leaf provided the chance of selecting suitable genotypes.

It was observed that the penultimate leaf contributes a higher proportion of the genetic variation in the grain yield than the flag leaf. Wazziki et al. (2014) reported under disease-free conditions, defoliation of the penultimate leaf causes a bigger yield reduction than the flag leaf. Seck et al. (1991) reported the flag leaf, penultimate leaf and antepenultimate leaf have contributed 26, 12 and 3%, respectively, to the grain yield per tiller. The combined contribution of the upper three leaves is more important than the flag leaf alone in the yield enhancement under leaf rust infected conditions (Seck et al. 1991) and insect damage (Buntin et al. 2004). In the present study, seven genotypes were identified for a higher SPAD value in the penultimate leaf than the flag leaf. The genotypes SW-139; SW-108; DWR-F8-35-9-1; NHP-F8-130; DWR-F8-3-1 were found to be superior for maintaining a high chlorophyll content in the flag leaf while NHP-F8-130, SW-139, DWR-F8-3-1, SW-138 maintained a high chlorophyll content in the penultimate leaf. These genotypes may be effectively used in breeding genotypes with a high chlorophyll content.

Heat stress inhibits the chlorophyll biosynthesis, breakdown of the thylakoid membrane and triggers leaf senescence (Al-Khatib & Paulsen 1984; Farooq et al. 2011). However, delayed senescence provides higher longevity to the top most leaves to translocate the photosynthates into the grains and SG maintains

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the leaf greenness for longer duration under stress. A positive association of the stay-green trait with the days to heading was recorded in our study. The significant positive correlation of the SPAD in the flag and penultimate leaf with the stay-green trait showed the direct relationship of the leaf chlorophyll content in maintaining the leaf greenness under heat stress. It can also be revealed from the significant differences in the stay-green score with respect to the low, intermediate and high SPAD values in the flag and penultimate leaf. Although, a strong association of the SPAD in the flag and penultimate leaf with the SG were not recorded, this can be ascribed to stability of the leaf chlorophyll playing an important role in maintaining the SG. Previous studies have found that the stay-green trait can provide yield advantages; hence, the usefulness of this trait as criteria for the genotype selection has been suggested (Joshi et al. 2007a; Kumari et al. 2013). Four genotypes in our study, i.e., NHP-F8-85-1, SW-138, DWR-F8-19-7, and SAWYT-4 were identified that exhibited the SG trait, whereas a few genotypes were grouped as moderately SG. Most of these genotypes also maintained moderate to a high level of chlorophyll content in the flag leaf and penultimate leaf. As the leaf senescence starts from the lower leaves of the plants, the longevity of the chlorophyll in the penultimate leaf may also protect the flag leaf from the early senescence.

The results of the present investigation suggest that the SPAD in the penultimate leaf can also be an important determinant for screening genotypes under stress as revealed by a significant correlation of the SPAD in the flag leaf and penultimate leaf with the grain yield and stay-green traits. The high heritability for all the traits under stress indicated the extent of the genetic variation and effectiveness in the selection of the genotypes. Promising genotypes for each trait were identified which can be further used as parents in breeding programmes. SW 108 was identified for a higher yield as well as the SPAD value in the flag leaf; SW 138 was identified for the SPAD in the penultimate leaf and stay-green trait; NHP F8-130 was identified for the SPAD in the flag leaf and penultimate leaf. These genotypes can be used as a donor in breeding programmes.

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