EVALUATION OF HEAT TOLERANCE INDEX, SUSCEPTIBILITY INDEX OF CANOPY TEMPERATURE AND LEAF CHLOROPHYLL CONTENT OF WHEAT (*TRITICUM AESTIVUM* L.)

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Abstract. Heat negatively affects wheat production in the world. At present, heat tolerance remains one least understood field in wheat genetics and breeding, because there are not enough effective methods for identification of heat stress and tolerance. This study used sixteen cultivars of winter wheat to evaluate a new method with stress intensity (δ) and heat tolerance index (HTI), and assess the susceptibility index of canopy temperature (SCT) and leaf chlorophyll content (SLCC) at different days after the anthesis to identify heat tolerance. The result showed HTI had significant positive correlations with yield under heat stress, and significant negative correlations with yield reduction rate, indicating that HTI is a good indicator of both yield potential and stability under heat stress which can be used to identify heat tolerance in later generations. SCT at 28 DAA had significant positive correlation with yield and significant negative correlation with yield reduction rate under heat stress. SLCC at 29 DAA had positive correlation with yield and significant negative correlation with yield reduction rate, and especially kernel weight reduction under heat stress. The two indicators may be used to screen for heat tolerant germplasm in early generations, or to identify a large number of wheat cultivars concurrently.
Keywords: *wheat, heat stress, stress intensity, yield, yield reduction*

Introduction

Wheat (*Triticum aestivum* L.) is one of the most important foods in the world, and it has become a basic food staple in Asia, Europe, and North Africa. Therefore, global wheat yield needs to be increased to meet the rising demand for this grain, and high grain value must be maintained under climate change to ensure ongoing human nutrition, end-use functional properties, as well as commodity value (Nuttall et al., 2017). As climates warm, heat stress during the post-anthesis period (terminal heat) negatively affects wheat production. This increased temperature not only hastens the phenological stages of wheat development but also reduces the duration of the grain filling stages, thereby lowering grain yield and quality (Faroop et al., 2011; Figueiredo et al., 2015; Gooding et al., 2003; Hoffmann et al., 2006). Increases in mean daily air temperatures of 1 °C during wheat development are projected to shorten the grain filling period by 3.1 d and decrease the weight per grain by as much as 2.8 mg (Wiegand and Cuellar, 1981). These wheat yield losses severely threaten global food security.

Most heat stress studies were aimed at predicting and maximizing yield (Amani et al., 1996; Blum et al., 2001; Rane and Nagarajan, 2004; Reynolds et al., 1998; Tewolde

et al., 2006; Xu et al., 2000). Some other studies focused on stability under heat stress. For example, some wheat genotypes were demonstrated to be tolerant to the effects of heat stress on grain quality at 29 days after anthesis (days after anthesis, DAA) (Blumenthal et al., 1995). In a recent paper, 54 genotypes were classified in two overall groups: stable and unstable, based on their rank shifts in different environments (Hernández-Espinosa et al., 2018). However, these numerical values cannot completely reflect the heat resistance of different wheat cultivars. The ultimate indicator of cultivar-specific heat resistance is manifested in the absolute and relative yield, that is, both yield potential and stability. Ideal heat resistance would be displayed as a durable, consistent yield with minimal yield reduction under heat stress conditions.

Some characteristics like leaf chlorophyll content and canopy temperature depression may be correlated with field performance, especially under heat stress (Jin et al., 2012; Wu et al., 2015). Ayeneh et al. (2002) compared the leaf, spike, peduncle, and canopy temperature depression in wheat under heat stress. Bahar et al. (2008) studied the effect of canopy temperature depression on grain yield and yield components in bread and durum wheat. Webber et al. (2015) used a multi-model approach including canopy temperature to simulate heat stress in irrigated wheat in a semi-arid environment. Gautam et al. (2015) found canopy temperature may be used as a selection parameter for grain yield and its components in Durum Wheat under terminal heat stress in late sown conditions. The term of susceptibility index ($1-Y/Y_p$) was used to reflect response of plant yield to heat stress, where Y is yield under stress, Yp is yield without stress, which was commonly used for estimating stress resistance (Li et al., 2018; Mason et al., 2010).

The purpose of this study was to evaluate a new method for identifying the heat tolerance of wheat cultivars based on yield and yield reduction under heat stress, and the related physiological indicators of canopy temperature and leaf chlorophyll content at different times after heat stress treatment. We designed a movable greenhouse for temperature control and created a heat tolerance index (heat tolerance index, HTI) for testing different wheat cultivars. This identifying method combining yield potential and stability could be a useful selection method for characterizing cultivar performance under heat stress. The susceptibility index of canopy temperature (susceptibility index of canopy temperature, S_{CT}) and leaf chlorophyll content (susceptibility index of leaf chlorophyll content, S_{LCC}) at different days after the anthesis have potential application for breeding in indirect selection to identify physiologically superior genotypes under heat stressed environment.

Materials and methods

Cultivars

The 16 winter wheat (*Triticum aestivum* L.) cultivars most commonly grown in the North China Plain (*Table 1*) were evaluated for heat tolerance. Cultivars were mechanically sown by Wintersteiger plotseed TC at the research station in Hebei Hengshui (37°44′N, 115°42′E; elev. 20 m). Meteorological data for this area is shown in *Figure 1*.

The experiment was conducted on sandy loam soil in field conditions. A split block design with 3×2 treatments and three replicates was employed during the 2016-2017 and 2017-2018 crop seasons. Each cultivar was grown in an 11.16-m² plot (nine rows of 8 m length with 15.5 cm space between rows). After adjusting for seed size, the seed

density was maintained at a uniform population of 300 plants per m^2 (3 million plants per hectare), according to the 1000 grain weight and germination percentage. Standard agronomic practices recommended for normal fertility (340 kg/ha N: 172.5 kg/ha P$_2$O$_5$: 40 kg/ha K$_2$O) were followed. All K$_2$O and P$_2$O$_5$ were applied at the time of sowing. Nitrogen was supplied in split applications: 170 kg/ha N at sowing and 170 kg/ha N at the first irrigation. Care was taken to avoid moisture and biotic stress by ensuring timely irrigation and pesticide control. Cultivars were harvested by Wintersteiger and weighted by electronic scale.
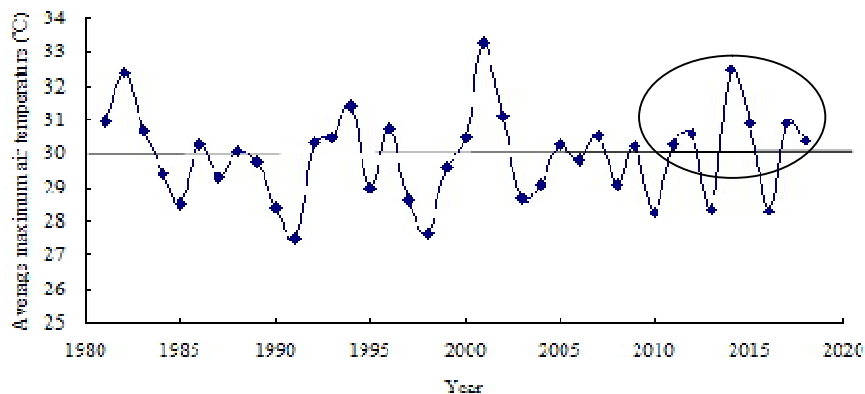

Figure 1. *Average daily maximum air temperature (14 May - 14 June) per year from 1981 to 2018 in the wheat growing region from this study. Temperature varied from 27.5 °C in 1991 to 33.3 °C in 2001. The increased frequency with higher daily maximum temperature than average value in a 10-year cycle indicates an increased risk in environmental heat pressure during the wheat grain filling stage in this region*

Table 1. *The 16 winter wheat cultivars evaluated in this study*

No.	Cultivar	No.	Cultivar
1	Heng11-6021	9	HengH14-4019
2	Heng9966	10	HengH14-5051
3	HengH14Guan14	11	HengH15-4489
4	HengH13Guan26	12	HengH13-5062
5	Heng5835	13	Heng12-6098
6	HengS13-5022	14	Heng14-K2-3
7	Heng5109	15	Jimai22
8	HengH15-4585	16	Heng4399

Heat treatments

On 14 May 2016, the mobile temperature-controlled greenhouse was used to cover the 16 cultivars at 14 DAA (for 80% of individuals). Heat treatments continued for 32 d, increasing the daily maximum temperature from 28.1 °C to 48.0 °C during 2016-2017 and from 22.3 °C to 48.6 °C during 2017-2018 (*Fig. 2*). Temperature was maintained 5 °C (± 1 °C) higher than ambient conditions.
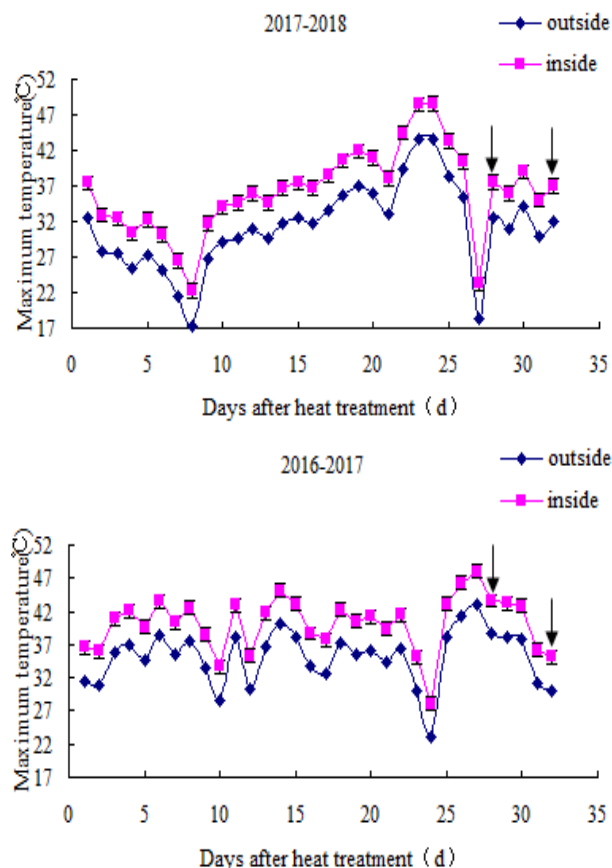
Figure 2. *Daily maximum temperature inside and outside of the greenhouse in the 2016-2017
and 2017-2018 seasons. An approximate 5 °C difference between the inside and outside
temperatures was maintained for 32 d. Bars mean SE. Arrows indicate the time of canopy
temperature measurements. The air temperature had been measured parallel with the canopy
temperature*

X and Xp indicate yield averaged over all cultivars under stress and control treatments, respectively. 1-X/Xp represents 'stress intensity' (δ). These criteria can be used to estimate resistance to stress (Li et al., 2018; Mason et al., 2010).

The heat tolerance index (HTI) includes both yield potential and stability after heat stress. To include the value of yield potential after heat stress, we substituted absolute yield under heat stress (yield, Y) into the formula, resulting in:

$$HTI = \frac{Y_S^{\ 2}}{Y_{S.P}} \times \frac{Y_{CK.P}}{Y_{CK}^{\ 2}} \qquad \text{(Eq.1)}$$

where Y_S: yield of identified cultivars under heat stress; $Y_{S.P}$: yield of identified cultivars under normal treatment; $Y_{CK.P}$: yield of CK cultivar under normal treatment; Y_{CK}: yield of CK cultivar under heat stress. We calculated relative yield (Y/Y_P) to account for variation in yield under the control conditions. Relative yield is commonly used in studies of stress resistance (Blum, 1973; Fischer and Maurer, 1978). We also included control data to account for variability in other environmental factors.

HTI was used to classify each cultivar (*Table 2*) as tolerant (≥ 1.20), moderately tolerant (1.00 to ≤ 1.19), moderately susceptible (0.80 to ≤ 0.99), or susceptible (≤ 0.79).

Table 2. HTI scale for tolerance of or susceptibility to heat stress for different wheat cultivars

Scale	HRI	Opinion
1	≥ 1.20	Tolerant
2	1.00-1.19	Moderately tolerant
3	0.80-0.99	Moderately susceptible
4	≤ 0.79	Susceptible

Canopy temperature measurements

Canopy temperature was measured using a hand-held infrared thermometer (IRT) (Model AG-42, Telatemp Crop, Fullerton, CA) at 15 and 20 d after heat treatment (28 May and 2 June). Three measurements were taken per plot with the instrument held at an angle of 30° to the horizontal plane, 1 m away from the edge of the plot and approximately 50 cm above the plants, giving a canopy view of 10 cm × 25 cm. Measurements were taken 0.5 h before and 2 h after solar noon, in full sunshine, 3 or 4 d after irrigation had been applied, as recommended by Amani et al. (1996). The equation of S_{CT} is as follows:

$$S_{CT} = \frac{CT}{CT_P} - 1 \qquad (Eq.2)$$

where CT (canopy temperature, CT) is canopy temperature under stress, CT_p is canopy temperature without stress.

Leaf chlorophyll content measurements

Chlorophyll content was measured in 10 randomly selected flag leaves in each plot by using a portable chlorophyll content meter (CCM-200, Opti-Sciences Inc., NH, USA). Ten measurements were taken for each plot. The measurements were taken at 29DAA and 33DAA. The equation of S_{LCC} is as follows:

$$SLCC = 1 - \frac{LCC}{LCC_P} \qquad (Eq.3)$$

where LCC (leaf chlorophyll content, LCC) is leaf chlorophyll content under stress, LCCp is leaf chlorophyll content without stress.

Statistical analysis

Statistical analysis was done using SAS software (SAS Institute 2003). Statistical analysis using HTI values, SCT, SLCC, yield and yield reduction were performed for sixteen genotypes. The linear correlation coefficient was calculated to determine the

association among of HTI values, SCT and SLCC with yield and yield reduction under heat stress. Differences were considered significant at the P < 0.01 level.

Results

Meteorological data

The average maximum temperature (14th May - 14th June) in Hengshui from 1981 to 2018 was 30 °C. The highest average temperature recorded was 27.5 °C in 1991, and the highest temperature during the grain filling stage was 33.3 °C in 2001. These maximum temperatures have a difference of 5.8 °C. We used this temperature difference in the controlled greenhouse experiments by exposing the treatment plants to a heat stress treatment 5 °C higher than the control treatment. In the first decade (1981-1990), the average maximum temperature was 30 °C, and five of these years were greater than the average value; in the second decade (1991-2000), the average maximum temperature was 29.6 °C, and five years were greater than the average value; in the third decade (2001-2010), the average maximum temperature was 30 °C, with another five years greater than the average value; in 2011-2018, the average maximum temperature was 30.3 °C, which has been higher than the long-term average during six years. The increased frequency with higher daily maximum temperature than average value in a 10-year cycle indicates an increased risk in environmental heat pressure during the wheat grain filling stage in this region.

We obtained the values for 'stress intensity' of $\delta_{2017} = 0.33$ and $\delta_{2018} = 0.11$ in 2016-2017 and 2017-2018 respectively, in accordance with the trend of average maximum air temperature change from 2017 to 2018.

The maximum daily temperature inside and outside of the greenhouse during the 2016-2017 and 2017-2018 crop growing seasons are presented in *Figure 2*. The ranges of the daily maximum temperature inside the greenhouse were from 28.1 °C to 48.0 °C and from 22.3 °C to 48.6 °C during 2016-2017 and 2017-2018, respectively.

Evaluation and application of heat tolerance index (HTI)

Heat stress affected values for mean yield, which dropped from 8864.9 to 5978 kg/ha in 2017 and from 7425.8 to 6588.3 kg/ha in 2018, when comparing the control to the heat stress treatments (*Table 3*). Stress intensity (δ) was 0.33 in 2017 and 0.11 in 2018 (*Fig. 3*). Average yield of the 16 wheat cultivars decreased by 18 kg/ha (2017) and 5.2 kg/ha (2018) under heat stress during the grain filling stages.

HTI calculated by *Equation 1* with maximum value in cultivars exhibited lower YR and higher yield under heat stress. HTI reached a maximum of 1.42 for the HengH15-4489 cultivar in 2017 and of 1.16 for the Heng14-K2-3 cultivar in 2018. HTI attained a minimum of 0.74 for the Heng9966 cultivar in 2017 and 0.84 for the HengH14-5051 cultivar in 2018. There was no detectable difference in HTI between the two experimental years. Cultivars, yield, reduction under heat stress, and HTI showed significant correlations (*Table 4*). Positive correlations (R = 0.9775 and R = 0.9415 in 2017 and 2018, respectively) were found between HTI and mean yield under heat stress (*Fig. 3*). Significant negative correlations (R = -0.9419 and R = -0.7458 in 2017 and 2018, respectively) existed between HTI and YR rate (*Fig. 3*).
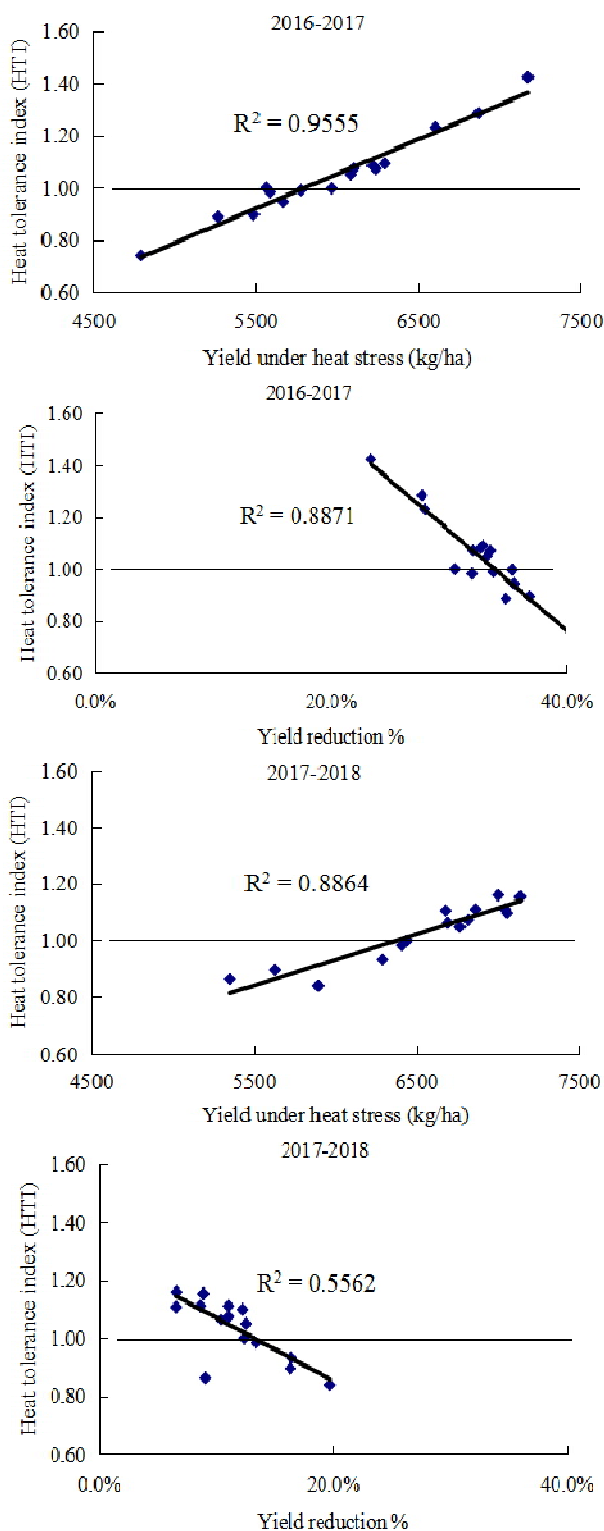
Figure 3. *Correlations based on averaged values for the 16 wheat studied cultivars for yield under heat stress (kg/ha) and yield reduction (YR) with heat tolerance index (HTI) in the 2016-2017 and 2017-2018 growth seasons. Positive correlations (R = 0.9775 in 2016-2017; R = 0.9415 in 2017-2018, P < 0.001) existed between HTI and yield under heat stress. Negative correlations (R = -0.9419 in 2016-2017; R = -0.7458 in 2017-2018, P < 0.001) were observed between HTI and YR. The line at 1 on the y-axis indicates standard heat tolerance. Dots indicate average values for each cultivar*

Across the two years, 5 cultivars consistently showed HTI < 1 (Heng9966, Heng12-6098, Hengs13-5022, Heng5835, and Hengh14-5051) with low yield and high yield reduction under heat stress (*Table 3*). There were 8 cultivars, HTI > 1 (Heng5109, Jimai22, Heng11-6021, Heng14-K2-3, HengH15-4585, HengH13Guan26, HengH14-4019, and HengH15-4489), indicating high yield and low yield reduction under heat stress.

Table 3. *The evaluation on the yield, yield reduction, HTI, S_{CT} at 28DAA and S_{LCC} at 29DAA of 16 wheat cultivars. Y: yield, YR: yield reduction, HTI: heat tolerance index, S_{CT}: susceptibility index of canopy temperature, S_{LCC}: susceptibility index of leaf chlorophyll content*

Cultivar	2016-2017					2017-2018				
	Y	**YR**	**HTI**	**S_{CT}**	**S_{LCC}**	**Y**	**YR**	**HTI**	**S_{CT}**	**S_{LCC}**
HengH15-4489	7172.73	23.37%	1.42	0.50	0.69	6859.94	8.7%	1.11	0.22	0.22
HengH14-4019	6867.01	27.72%	1.28	0.37	0.19	7143.68	8.9%	1.15	0.25	0.25
HengH13Guan26	6600.57	27.95%	1.23	0.26	0.37	7057.68	12.3%	1.10	0.17	0.19
HengH15-4585	6289.61	32.90%	1.09	0.21	0.19	6813.76	11.1%	1.07	0.21	0.18
Jimai22	6215.86	32.66%	1.08	0.24	0.19	7045.74	11.1%	1.11	0.20	0.20
Heng14-K2-3	6102.14	32.06%	1.07	0.32	0.10	7001.97	6.6%	1.16	0.22	0.26
Heng11-6021	6233.34	33.53%	1.07	0.26	0.13	6693.87	10.4%	1.06	0.19	0.16
Heng5109	6082.20	33.26%	1.05	0.23	0.09	7136.10	8.9%	1.15	0.23	0.21
Heng4399	5559.11	30.51%	1.00	0.30	0.25	6439.05	12.4%	1.00	0.14	0.12
Heng5835	5962.75	35.37%	1.00	0.21	0.11	5352.49	9.1%	0.86	0.18	0.18
Heng12-6098	5777.02	33.78%	0.99	0.25	0.19	6292.44	16.4%	0.93	0.19	0.14
HengH14Guan14	5582.43	31.98%	0.98	0.21	0.01	6766.73	12.5%	1.05	0.20	0.09
HengH13-5062	5664.56	35.52%	0.95	0.29	0.12	6680.69	6.6%	1.11	0.28	0.11
HengH14-5051	5484.24	36.84%	0.90	0.19	0.23	5894.21	19.7%	0.84	0.10	0.14
HengS13-5022	5262.46	34.83%	0.89	0.08	0.11	5627.07	16.3%	0.90	0.06	0.07
Heng9966	4792.11	40.40%	0.74	0.19	0.16	6406.01	13.4%	0.98	0.17	0.12

Evaluation of susceptibility index of canopy temperature (S_{CT}) and leaf chlorophyll content (S_{LCC})

In order to evaluate the susceptibility index related physiological indicators, we detected canopy temperature and chlorophyll content. S_{CT} and S_{LCC} were calculated by *Equations 2* and *3* respectively. S_{CT} reached a maximum of average value 0.36 for the HengH15-4489 in 2017 and 2018. Meanwhile S_{CT} attained a minimum of average value 0.07 for the HengS13-5022 in 2017 and 2018. S_{LCC} reached a maximum of average value 0.46 for the HengH15-4489 in 2017 and 2018. Meanwhile S_{LCC} attained a minimum of average value 0.05 for the HengH14Guan14 in 2017 and 2018. The result (*Table 4*) showed that there were significant positive correlations (R = 0.7356 and R = 0.6579 in 2017 and 2018, respectively; *P* < 0.01) between S_{CT} at 28DAA and yield under heat stress, and significant negative correlations (R = -0.7645 and R = -0.8164 in 2017 and 2018, respectively; *P* < 0.001) between S_{CT} and yield reduction under heat treatment. Meanwhile there were significant negative correlations (R = -0.6762 and R = -0.5548 in 2017 and 2018, *P* < 0.01 and *P* < 0.05, respectively) between S_{LCC} at 29 DAA and yield reduction under heat stress, and significant negative correlations (R = -

0.7497 and R = -0.8556 in 2017 and 2018, respectively; $P < 0.001$) between S_{LCC} and kernel weight reduction under heat treatment.

Table 4. *Correlations among yield under heat stress (Y), yield reduction under heat stress (YR%) and kernel weight reduction (KWR%) as canopy temperature (CT), leaf chlorophyll content (LCC), susceptibility index of canopy temperature (S_{CT}), and susceptibility index of leaf chlorophyll content (S_{LCC}) in 2016-2017 and 2017-2018*

Time	Content	2016-2017			Daily maximum temperature	2017-2018			Daily maximum temperature
		Y	YR	KWR		Y	YR	KWR	
28 DAA	CT	-0.6984**	0.6336**	0.3517	38.1 ℃	-0.6763**	0.5888*	0.5872*	32.4 ℃
	S_{CT}	0.7356**	-0.7645***	-0.5975*		0.6579**	-0.8164***	-0.4093	
29 DAA	LCC	-0.1605	0.3492	0.6289**	33.7 ℃	0.0123	-0.2359	0.2013	31.8 ℃
	S_{LCC}	0.5870*	-0.6762**	-0.7497***		0.5614*	-0.5548*	-0.8556***	
32 DAA	CT	0.0611	0.0062	-0.3757	35.5 ℃	0.3611	-0.0752	-0.2019	36.9 ℃
	S_{CT}	-0.1131	-0.2099	0.1720		0.0006	0.3464	-0.3252	
33 DAA	LCC	-0.0316	0.1222	0.1788	36.2 ℃	-0.3246	-0.0845	0.1610	35.9 ℃
	S_{LCC}	-0.4802	0.4129	0.6002*		0.2476	0.0317	0.1477	

*, **, *** significant at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively

Discussion

Meteorological change

According to the meteorological data in 38 years, the frequency of the average maximum temperatures appeared in the winter wheat grain filling stage (14 May-14 June) in Hengshui are increasing, which is similar with findings of the Intergovernmental Panel on Climatic Change (IPCC, 2013). Heat tolerance identification and breeding should be preferred to be researched, even with extremely high temperature.

Evaluation and application of heat tolerance index (HTI)

HTI showed significant correlations with yield and yield reduction under heat stress in 2 crop seasons, which supports the conclusion that HTI can be used to identify heat tolerance by indicating the yield potential and yield stability of wheat cultivars under heat stress.

HTI has several advantages over other methods for identifying heat tolerance. It is easier to quantify using the 'stress intensity' (δ) and HTI. Correlations between HTI, yield, and yield reduction were significant across the two experimental years, indicating reliability across variable growth conditions. HTI can also measure heat tolerance by incorporating both the values of yield stability and potential, linking yield reduction under heat stress to maximum possible yield.

There were 8 wheat cultivars whose HTI reached or exceeded level 2 ('moderately tolerant') across both years indicating adequate heat tolerance. These cultivars should be considered for wheat breeding with high heat tolerance. There were 5 wheat cultivars that reached level 3-4, indicating adequate heat susceptibility, and may be used for genetic analysis. In further study, we can use HengH15-4489 with the highest HTI and Heng9966 with the lowest HTI according to the two-year average, as parents to construct genetic population to analysis heat tolerant mechanism, heat tolerant genes mapping and tagging.

HTI can give a fuller picture of heat tolerance than yield reduction alone. In one cultivar, Heng5835, HTI in 2017-2018 was low as 0.86, but the yield reduction reached only 9.1%. This may be due to its early and low yield in the control treatment. Therefore, the use of only yield reduction may give incorrect information about heat tolerance.

HTI may be more able to identify heat tolerance when stress intensity is higher. In 2017-2018, stress intensity reached $\delta_{2018} = 0.11$ due to the severe heat waves that occurred in late stage of filling, and the yield under the control treatment reduced. Results would otherwise be clustered due to this strong effect, but HTI was reliable.

Evaluation of susceptibility index of canopy temperature (S_{CT}) and leaf chlorophyll content (S_{LCC})

Measuring Canopy temperature and leaf chlorophyll content was inexpensive, fast, easy, and suitable for breeding applications (Brennan et al., 2007; Reynolds et al., 2007). In this study we used susceptibility index of canopy temperature (S_{CT}) and leaf chlorophyll content (S_{LCC}) at different days after the anthesis to identify heat tolerance. The wheat cultivar with the highest S_{CT} and S_{LCC} during 2017-2018 was HengH15-4489, which was in agreement with the result of HTI. This cultivar performed heat tolerance well during 2017-2018. In further research, we can study on heat tolerant mechanism by using this cultivar.

The result showed that S_{CT} at 28 DAA had significant positive correlation with yield and significant negative correlation with yield reduction rate under heat stress, indicating that the wheat lines with higher S_{CT} may have high yield potential and yield stability in early generation under heat stress. It was reported that the canopy temperature and transpiration rate had a very significant negative correlation (Wall et al., 2006). Here, the canopy temperature of the cultivars with good heat tolerance increases greatly. It may be related to the regulation of reducing the transpiration rate with water loss getting slower and plant function keeping longer. The result need to be further verified.

S_{LCC} at 29 DAA had positive correlation with yield and significant negative correlation with yield reduction rate, and especially kernel weight reduction under heat stress, indicating that wheat lines with higher S_{LCC} may have lower yield and kernel weight reduction for stability under heat stress. The results agree with previous studies, showing that plant adapt to stress environment by reducing chlorophyll content in plant leaves to alleviate photo-inhibition and improve photosynthesis (Melis, 2009; Ort et al., 2011, 2015; Polle et al., 2003).

The result also indirectly proved the mechanism of heat tolerance: under heat stress, stomatal closure reduces transpiration, and then canopy temperature rise; meanwhile chlorophyll content in plant leaves reduce to alleviate photo-inhibition then improve photosynthesis, and to reduce light absorption then reduce canopy temperature.

The new methodology better estimated heat tolerance compared to other methods for four reasons. First, the heat treatment was easier to quantify because of using 'stress intensity' (δi). Second, HTI, SCT and SLCC had greater repeatability over different years and different stress intensities. Third, the correlations among HTI, SCT, SLCC, yield and yield reduction under heat stress was always significant in different years. Finally, HTI, SCT and SLCC linked the yield under heat stress to the yield reduction and reflected a heat tolerance with yield potential and stability in heat stress of different wheat cultivars simultaneously.

The different cultivar responses to heat stress indicated that the heat resistance ability of wheat cultivars varied. HTI and SCT was able to represent these varied responses and significantly correlate with the yield and yield reduction of wheat under heat stress. Accordingly, this new methodology can be used to reflect the yield potential and the stability to heat stress of different wheat varieties simultaneously. Cultivars with a high HTI (> 1) or higher SCT can be considered to be heat tolerance, as they exhibited higher yield and smaller yield reductions under heat stress compared with the other cultivars. The new methodology was a reliable evaluation for heat-tolerance of wheat cultivars. HTI is a good indicator of both yield potential and stability under heat stress which can be used to identify heat tolerance in advanced generations. The other two indicators SCT and SLCC may be used to screen for heat tolerance germplasm in early generations, or to identify a large number of wheat cultivars concurrently. HTI combined with SCT and SLCC maybe a good breeding strategy for heat tolerance improving.

Conclusions

In this study of heat tolerance across 16 wheat cultivars, HTI, S_{CT} and S_{LCC} showed significant correlations with yield and yield reduction under heat stress, and may be efficient indicators reflecting heat tolerance, including both yield potential and stability simultaneously. HTI can be used to identify heat tolerance of wheat cultivars in advanced generations, and S_{CT} at 28 DAA and S_{LCC} at 29 DAA may be used to screen for heat tolerant germplasm in early generations, or to identify a large number of wheat cultivars concurrently.

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