

## Effect of heat stress at anthesis on yield formation in winter wheat

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### ABSTRACT

Hlaváčová M., Klem K., Smutná P., Škarpa P., Hlavinka P., Novotná K., Rapantová B., Trnka M. (2017): Effect of heat stress at anthesis on yield formation in winter wheat. *Plant Soil Environ.*, 63: 139–144.

Heat stress around anthesis is considered to have an increasing impact on wheat yield under the ongoing climate change. However, the effect of high temperatures and their duration on formation of individual yield parameters is still little understood. Within this study, the effect of high temperatures applied during anthesis for 3 and 7 days on yield formation parameters was analysed. The study was conducted in growth chambers under four temperature regimes (daily temperature maxima 26, 32, 35 and 38°C). In the periods preceding and following heat stress regimes the plants were cultivated under ambient weather conditions. The number of grains per spike was reduced under temperatures  $\geq 35^\circ\text{C}$  in cv. Bohemia and  $\geq 38^\circ\text{C}$  in cv. Tobak. This resulted in a similar response of spike productivity. Thousand grain weight showed no response to temperature regime in cv. Tobak, whereas in cv. Bohemia, a peak response to temperature with maximum at  $35^\circ\text{C}$  was observed. The duration of heat stress had only little effect on most yield formation parameters.

**Keywords:** high temperature stress; grains number per spike; spike productivity; phytotron; *Triticum aestivum* L.

The wheat is known to be very sensitive to heat stress during reproductive stages, especially at anthesis (Saini et al. 1983, Alghabari et al. 2014). Furthermore, the critical heat stress during wheat

anthesis is expected to be more frequent due to climate change (Stratonovitch and Semenov 2015). An increase in mean annual temperature approximately by  $1^\circ\text{C}$  for the period of 2021–2040 by

Supported by the individual IGA project IGA AF MENDEL, Project No. IP 11/2016 with the support of the Specific Research Grants, provided by the Ministry of Education, Youth and Sports of the Czech Republic.

doi: 10.17221/73/2017-PSE

ensemble of models is predicted for the Czech Republic ([www.czechadapt.cz](http://www.czechadapt.cz)) as well as days with temperature above damaging threshold (estimated to be about 32°C for wheat) are supposed to be more frequent (Trnka et al. 2014, Pradhan and Prasad 2015). The temperatures above 30°C at anthesis can result in reduction of grain size or complete grain sterility (Saini and Aspinall 1982, Fischer 1985, Pradhan and Prasad 2015). While previous studies focused on the effect of heat stress on wheat yield after several days of exposure, the effect of heat stress duration and detailed temperature response of individual yield formation parameters are still not well understood. The objectives of the present study were: (1) to investigate the effect of the high temperature (32, 35 and 38°C compared to 26°C as a control) on the selected yield formation parameters – grain number per spike (GN), spike productivity (SP) and thousand grain weight (TGW); (2) to evaluate the effect of heat stress duration on the selected yield formation parameters, and (3) to analyse interactions between temperature and heat stress duration.

## MATERIAL AND METHODS

**Experimental design.** Two winter wheat cultivars, Bohemia and Tobak, were sown on the 12<sup>th</sup> October 2015 in the number of 2 seeds per 1 pot. The dimensions of pots (10.5 × 10.5 × 21.5 cm) enabled normal growth and development of main stems (but the number of tillers was zero) until full ripening stage. The pots were placed in the vegetation hall of the Mendel University in Brno (235 m a.s.l.; 16°36'48.64716"N, 49°12'36.62892"E). A long-term (1981–2010) average annual precipitation and annual temperature measured at the near meteorological station (distance 650 m), is 516.6 mm and 9.8°C, respectively. The pots were filled prior to the sowing with the soil from Polkovice experi-

mental station (199 m a.s.l.). The soil was a luvic chernozem with a silt-clay texture (26% clay, 68% loam, 6% sand); pH<sub>CaCl<sub>2</sub></sub> 7.4; total carbon 2.66%; total nitrogen 0.25%; contents of P, Ca, Mg and K (Mehlich III) were 143, 6045, 252 and 567 ppm, respectively. The plants were placed onto the concrete floor in the open-air (covered by protecting net) vegetation hall and exposed to ambient weather conditions. The plants were regularly irrigated to provide water supply for optimum growth and development (total irrigation during placement in the vegetation hall was 80 mm). The amount of nutrients was based on the analysis of available nutrients at the time of sowing and was kept at levels to avoid limitation by these factors while pesticide screen against most common pests and diseases was used to ensure healthy plants during the whole experiment (Table 1).

**Heat stress regimes.** The pots were transported to the Global Change Research Institute CAS, v.v.i. (Brno, Czech Republic) at the end of heading (59 BBCH) where the pots were put into 4 growth chambers (FytoScope FS-SI 3400 model; Photon Systems Instruments, spol. s.r.o., Brno, Czech Republic) for 7 days of acclimation to the control chamber protocols (Table 2). The pots were watered every second day. The temperature regimes (Table 2) were initiated at the growth stage beginning of anthesis (61 BBCH). 7 pots of each cultivar were exposed to each temperature/duration treatment. The plants were kept in each temperature regime for 3 (indicated as an upper index of D3; until 65 BBCH) and 7 days (indicated as an upper index of D7; until 69 BBCH). When the particular stress regime was finished, the plants were moved to another growth chamber with the control regime (26°C). After finishing all temperature/duration treatments the pots were transported back to the vegetation hall where the plants were exposed to the ambient weather conditions until full ripeness when manual harvest was carried out (middle of July 2016).

Table 1. Overview of all treatments applied

Application date (2016)	Application	Concentration (%)
4 <sup>th</sup> April	fertilizer Kristalon <sup>TM</sup> Start	0.1
6 <sup>th</sup> April	fungicide Fandango <sup>®</sup> 200 EC	0.3
22 <sup>nd</sup> June	fungicide Prosaro <sup>®</sup> 250 EC*	0.2
11 <sup>st</sup> April	insecticide Karate with Zeon technology <sup>®</sup> 5 CS	0.1
22 <sup>nd</sup> June	insecticide Karate with Zeon technology <sup>®</sup> 5 CS *	0.1

\*combined chemical treatment

Table 2. Protocols established within the growth chambers during the pot experiment

Time	PAR ( $\mu\text{mol}/\text{m}^2/\text{s}$ )*	RH (%)*	Temperature ( $^{\circ}\text{C}$ )**
0:00–4:00	0	85–90	20–18
4:00–6:00	0	90	18
6:00–12:00	0–1500	90–45	18– $t_{\text{max}}$
12:00–14:00	1500	45	$t_{\text{max}}$
14:00–20:00	1500–0	45–75	$t_{\text{max}}$ –22
20:00–24:00	0	75–85	22–20

\*all chambers; the environmental factors changed continuously; \*\* $t_{\text{max}}$  = 26°C (control chamber), 32, 35 or 38°C; PAR – photosynthetically active radiation; RH – relative humidity

**Yield formation parameters assessment.** The grain number (GN) and spike productivity (SP) at 14% grain moisture per main spike were analysed for particular temperature/duration treatment.

**Statistical analysis.** The data sets were checked for outlier values prior to the data analysis by the 3 SD method, i.e. if the observation was outside the intervals  $x > \pm 3 \text{ SD}$  (where  $x$  – sample mean; SD – sample standard deviation) than the suspicious value was removed from another processing. The data were analysed using the Statistica 12.0 software (StatSoft, Inc., Tulsa, USA). The two-way ANOVA and Tukey's *HSD* (honest significant difference) tests were run at the level  $P = 0.05$  to determine the statistical significant differences among treatments of the temperature stress and the duration of the exposition to this stress factor for each winter wheat cultivar separately.

## RESULTS AND DISCUSSION

**Grain number per spike.** The results show that the plants of cv. Tobak exposed to the heat stress for the period of 3 days showed higher GN values than after 7 days (Figure 1, Table 3). The treatment 38<sup>D7</sup> exhibited statistically significantly lower GN values compared to other treatments except the treatment 38<sup>D3</sup> and 32<sup>D7</sup> where the differences are considerable, nevertheless, statistically not significant. ANOVA results within cv. Tobak also showed that the effect of temperature was statistically significant at  $P < 0.001$  whereas the effect of duration of exposure to temperature regime at  $P = 0.042$ . The highest GN in cv. Tobak was observed under treatment 35<sup>D3</sup> (GN = 32.7), contrary to the lowest mean value that was reached within treatment 38<sup>D7</sup> (GN = 20.9).

Table 3. The results of descriptive statistics of spike productivities at 14% moisture (SP) for each treatment and cultivar (in grams per treatment)

Temperature <sup>Days</sup>	Bohemia					Tobak				
	$\bar{x}$	me	$\sigma$	min	max	$\bar{x}$	me	$\sigma$	min	max
26	1.31	1.51	0.46	0.71	1.94	1.60	1.62	0.37	0.82	2.23
32 <sup>D3</sup>	1.42	1.39	0.42	0.64	2.22	1.70	1.72	0.39	1.07	2.29
32 <sup>D7</sup>	1.36	1.37	0.51	0.41	2.01	1.34	1.38	0.58	0.33	2.38
35 <sup>D3</sup>	1.08	0.99	0.24	0.78	1.49	1.72	1.71	0.19	1.41	2.10
35 <sup>D7</sup>	1.31	1.25	0.26	0.83	1.65	1.66	1.60	0.30	1.13	2.18
38 <sup>D3</sup>	0.94	0.98	0.32	0.50	1.55	1.36	1.39	0.51	0.57	2.13
38 <sup>D7</sup>	1.18	1.06	0.50	0.50	2.03	1.08	1.26	0.50	0.31	1.83

The values are arithmetic means ( $\bar{x}$ ); medians (me); standard deviations ( $\sigma$ ); minimum (min) and maximum (max) values reached within particular treatments. The heat stress duration was not distinguished within the control treatment 26 (the phytotron with  $t_{\text{max}} = 26^{\circ}\text{C}$ ), therefore, the columns are the same for 26<sup>D3</sup> and 26<sup>D7</sup>

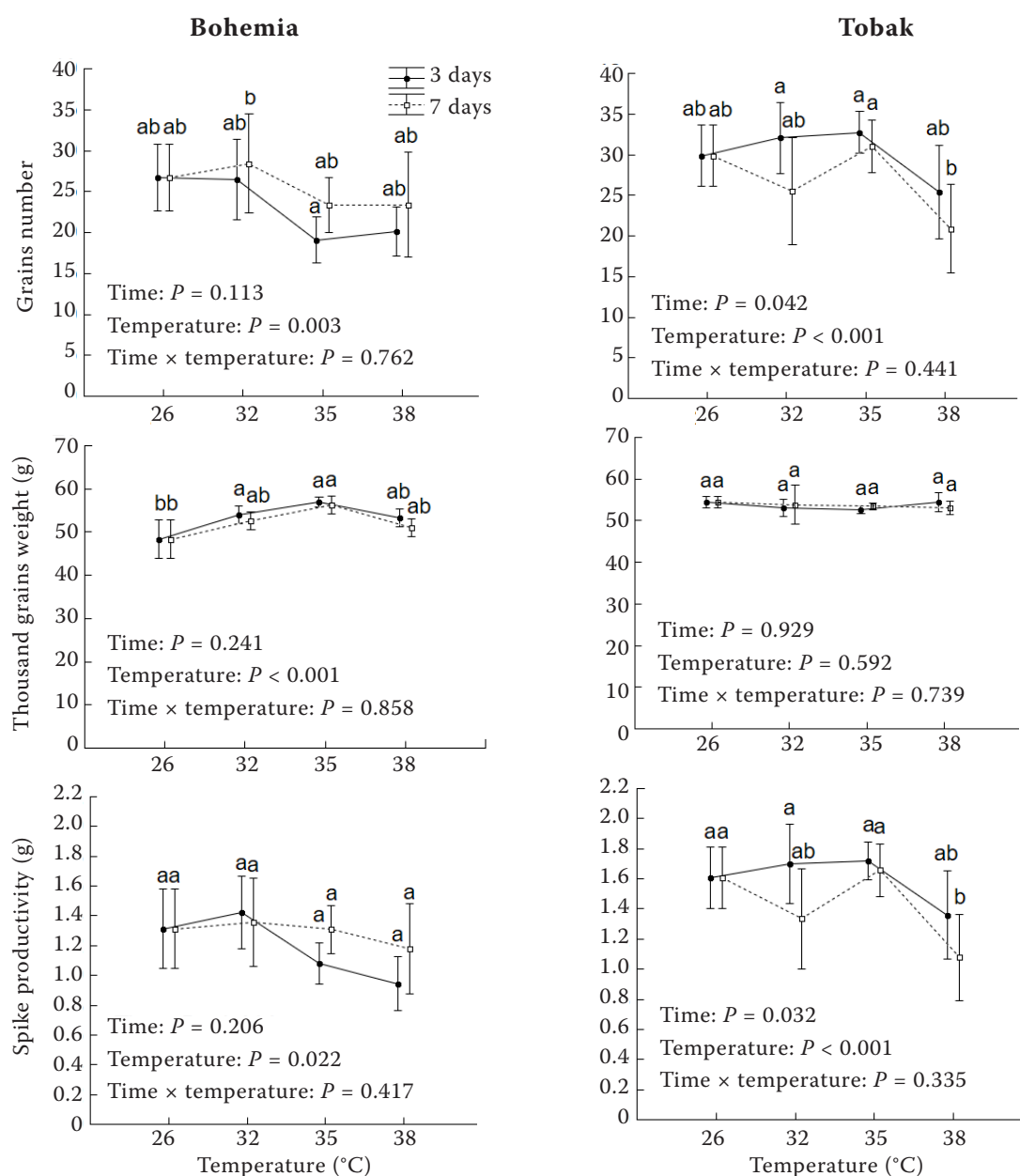


Figure 1. The two-way ANOVA graphical outputs ( $P = 0.05$ ,  $n \geq 10$ ) expressing the mean values and their confidence intervals as error bars for each yield formation parameter for high temperature stress (factor temperature) duration (factor time) for 3 (3 days) and 7 (7 days) days of cv. Bohemia and cv. Tobak. Different letters above columns indicate significant differences among treatments according to the Tukey's *HSD* (honest significant difference) test

The cv. Bohemia showed generally higher GN per spike and also opposite response to the duration of heat stress as compared to cv. Tobak (Figure 1, Table 3). Slightly higher GN values were found in cv. Bohemia for 7 days duration as compared to 3 days. These differences were more pronounced under higher temperatures. The statistically significant difference was found between the treatments 35<sup>D3</sup> and 32<sup>D7</sup>. The ANOVA results revealed that the

effect of temperature was statistically significant while the effect of heat stress duration was statistically insignificant as well as the interaction of these two factors. The highest mean GN value was found for the treatment 32<sup>D7</sup> (GN = 28.4) while the lowest one within the treatment 35<sup>D3</sup> (GN = 19.1). As reviewed by Wardlaw and Wrigley (1994) heat stress during anthesis increases floret abortion, which results in reduced GN per spike. The

optimum temperature for wheat anthesis ranges from 12°C to 22°C (Farooq et al. 2011). Higher temperatures during anthesis may cause pollen sterility, tissue dehydration or lower CO<sub>2</sub> assimilation rate. Temperatures above 30°C during floret formation may cause complete sterility (Saini and Aspinal 1982; pot experiment). Shpiler and Blum (1991) in a field experiment also demonstrated that GN per spike is positively associated with heat tolerance and can be thus used for selection of tolerant genotypes.

**Spike productivity.** In general, the SP results show very similar pattern of response to temperature/duration regimes. The results for cv. Tobak showed that the plants exposed to heat stress for the period of 3 days were less affected in comparison to those exposed to the stress for 7 days. The highest mean SP value was found for treatment 35<sup>D3</sup> (1.7 g) and the lowest one for treatment 38<sup>D7</sup> (1.1 g).

The cv. Bohemia demonstrated different responses to the duration of exposure to heat stress as compared to cv. Tobak. The plants left within stress conditions for 3 days showed lower SP values than those exposed to the stress for 7 days. This response was more evident in temperatures above 32°C. Nevertheless, the effect of time was proved to be statistically insignificant. The values of final SP were statistically significantly affected only by temperature. The statistically significant differences among treatments were, however, not proved. The highest mean SP value was found for the treatment 32<sup>D3</sup> (1.4 g) while the lowest one for treatment 38<sup>D3</sup> (0.9 g).

Heat stress is a function of the rate of temperature increase and the duration of exposure to the raised temperature (Wahid et al. 2007). The duration of heat stress seems to be an important factor affecting final yield response to high temperatures. For example, Rawson and Bagga (1979) in a pot experiment showed an inverse relationship between duration of heat stress and yield parameters. However, within our study, the differences between the duration of heat stress for 3 and 7 days were low and cv. Bohemia showed even less impact of the longer exposure to high temperatures on yield parameters. Such response can be attributed to stem reserve mobilization under heat stress (Blum 1998). Stem reserve storage depends on stem length, which is considerably higher in cv. Bohemia.

**Thousand grains weight.** The results for cv. Tobak showed that both duration and temperature as well as their interaction have statistically insignificant effect on TGW. The comparison with cv. Bohemia also shows that cv. Tobak is less sensitive in TGW even under longer exposition to heat stress (7 days). The highest mean TGW value was 54.5 g (26) while the lowest mean TGW value was 52.7 g (35<sup>D3</sup>).

TGW in cv. Bohemia exhibited the increasing trend with higher temperatures up to temperature maxima 35°C in both length of heat stress duration (3 and 7 days). An increase of temperature to 38°C led, however, to reduction of TGW. The effect of temperature on TGW was statistically significant. On the contrary, the effect of heat stress duration was statistically insignificant. The statistically significant differences were proved among the treatments 26, 32<sup>D3</sup>, 35<sup>D3</sup> and 35<sup>D7</sup>. The mean maximum of TGW was reached within the treatment 35<sup>D3</sup> (56.97 g) while the mean minimum value was found within the treatment 38<sup>D7</sup> (51.06 g).

The results from the Central Institute for Supervising and Testing in Agriculture (Brno) over several years (2011–2014) showed that the mean TGW values in field trials were 46 g for cv. Tobak and 52 g for cv. Bohemia (Horáková et al. 2015). The results within this study show above-average values of TGW in comparison to field testing of the same cultivar. Higher TGW within this experiment is probably also response to generally lower GN. Acreche and Slafer (2006) in a field study showed a close inverse correlation between GN and TGW proving the compensation ability of TGW in response to reduced GN. However, the TGW is not able to fully compensate yield loss caused by reduction in GN. As demonstrated by Shpiler and Blum (1991; field study), GN is a more important yield parameter under heat stress than TGW. The reduction in TGW under heat stress is generally associated primarily with shortening of grain filling period rather than the rate of grain filling and thus associated with longer heat stress duration (Wardlaw and Wrigley 1994). On the other hand, Stone and Nicolas (1995) in a pot experiment demonstrated that also short-term exposition to high temperatures at the stage of early grain filling can significantly affect TGW. The sensitivity to such short-term heat stress, however, reduces throughout grain filling period.

It was concluded that the magnitude of the heat stress at anthesis is more important for yield for-



doi: 10.17221/73/2017-PSE

mation than the heat stress duration. An ambient temperature in a growth chamber and not spike temperature is always used within such experiments. Nevertheless, the spike temperature is lower than the ambient air temperature since the plant cooling effect. When cultivars have awned spikes, this cooling effect may be stronger (Ayeneh et al. 2002). Both cultivars showed the highest sensitivity to elevated temperature in the yield parameter GN and these responses are also reflected in SP. Higher sensitivity was revealed in cv. Bohemia, in which the reduction in GN and SP occurs already under lower maximum temperatures. TGW was affected by temperature only little and more in cv. Bohemia, showing the maximum at 35°C. The response of TGW is probably more complex as high air temperature can induce mobilization of stem reserves and TGW can partly compensate reduction in GN. The decrease in GN was not compensated by the increase in harvest indices (HI). Generally, HI values were high, i.e. reached the upper limit of the genotypes (Hlaváčová et al. 2016).

## Acknowledgement

The authors would like to thank: (1) Ing. Vladimíra Horáková (CISTA, Brno) for providing seeds and valuable consultations, (2) Ing. Vítězslav Křetinský and Ing. Milena Žídková (GEOtest, Brno) for soil analyses and (3) Sára Šejnohová and Barbora Vítková for primary analyses of harvest.

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Received on February 6, 2017

Accepted on March 8, 2017

Published online on March 31, 2017