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▶ **To cite this version:**

Yang Liu, Xiaoyu Liu, Jing Cao, Chunlin Shi, Shouli Xuan. Effects of Waterlogging and Shading at Jointing Stage on Dry Matter Distribution and Yield of Winter Wheat. 10th International Conference on Computer and Computing Technologies in Agriculture (CCTA), Oct 2016, Dongying, China. pp.80-92, 10.1007/978-3-030-06155-5_8 . hal-02179968

**HAL Id: hal-02179968**

**https://inria.hal.science/hal-02179968**

Submitted on 12 Jul 2019

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# Effects of waterlogging and shading at jointing stage on dry matter distribution and yield of winter wheat

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**Abstract.** Continuous rain is the main meteorological constraint for winter wheat production in Jiangsu Province, accompanied by stresses of both waterlogging and shading. To evaluate the independent and combined effects on winter wheat at jointing stage, pot experiments were conducted using two cultivars, Ningmai 13 and Yangmai 13. Four treatments, CK (non-stressed), WA (waterlogging alone), SA (shading alone) and WS (both waterlogging and shading) were established with different durations. In the non-stressed environment, Yangmai 13 had higher production than Ningmai 13. However, Ningmai 13 had better production under stresses, indicating a better tolerance to waterlogging and shading. Comparing dry matter distribution and grain production showed that the negative effects of the stresses were in the order WA > WS > SA, demonstrating that shading had compensative effects on waterlogging at jointing stage. Results indicate that production loss of winter wheat due to continuous rain at jointing stage might be overestimated.

**Keywords:** waterlogging; shading; jointing stage; grain yield; winter wheat

## 1 Introduction

The winter wheat production area in Jiangsu Province is 2.13 M ha, representing about 9% of the overall winter wheat area of China in 2012 [1]. However, most winter wheat in Jiangsu is planted in paddy fields in a rice–wheat rotation [2] and results in poor drainage conditions. Furthermore, there is frequent continuous rain during the growth season of winter wheat (from jointing stage to maturity) due to the subtropical monsoon climate [3, 4]. The total average rainfall is about 500–800 mm during the growth season [5], which far exceeds requirements for winter wheat. In addition, frequency of extreme weather events is increasing globally and regionally [3, 6]. Therefore, soils are easily waterlogged in Jiangsu and waterlogging has become a major constraint for wheat production.

Experimental results on plants have shown that waterlogging stress retards root growth, reduces root hydraulic conductance [7], induces leaf senescence [8], shortens the duration of grain filling in wheat [9] and reduces dry matter accumulation and final grain yield [10, 11]. However, damage from waterlogging depends on the growth stage of plants, duration of waterlogging and the cultivar [12, 13]. There is scant knowledge of the physiology of recovery after varying durations of waterlogging [10].

Shading always accompanies waterlogging during continuous rain events. Similarly to waterlogging, shading can reduce crop dry matter accumulation and grain yield [14, 15] by reducing radiation, impairing net photosynthesis in leaves [16] and reducing the LAI (leaf area index) [17]. However, diffuse light increase under shading can compensate for the reduced radiation [18]. The reduction in LAI is partially compensated by increases in the fraction of the top and bottom leaf area to total leaf area, while the decrease in photosynthetic rate (Pn) of flag leaf is partially compensated by the increase in Pn of the third leaf from the top [17]. In addition, shading increases the redistribution of dry matter from vegetative organs to grain [2]. Thus, the shading effect on grain yield depends on the level of shading applied and on the cultivar [2].

In addition to independent studies on waterlogging and shading, some researchers have recently investigated combined stresses – most of this research has shown that combined waterlogging and

shading stress (WS) significantly decreased dry matter weight and final grain yield. In a pot experiment, the grain filling rate was decreased, although the apparent remobilization of carbohydrate reserves from stem to grain was stimulated under WS [19]. In an outdoor experiment, the growth and morphological responses of four wetland species to combined and independent effects of waterlogging and shading differed [20]. There was an amplified effect of WS stress for the least tolerant species but a reduced effect for the other species.

Although the physiology of wheat under waterlogging and shading stress has been studied independently, study of the WS stress is still scant, especially including recovery after different stress durations. Therefore, the objective of this study was to investigate the effects of waterlogging, shading and both together at jointing stage on the dry matter distribution and photosynthesis of winter wheat. The results will advance our understanding of wheat physiology under continuous rain events and could be used to improve wheat growth models by calibrating the effect of combined waterlogging and shading stress.

## 2 Experiments and Methods

### 2.1 Experimental design

The experiment was conducted in the winter wheat growing season of 2013–2014 at the Experimental Station of Jiangsu Academy of Agricultural Sciences, Nanjing (32°2′N,118°52′28″E), Jiangsu Province, China. Two representative local winter wheat (*Triticum aestivum* L.) cultivars, Ningmai 13 and Yangmai 13, were grown in plastic pots (20 cm in height and 25 cm in diameter). Each pot was filled with 12 kg of air-dried clay soil, and seven small holes (1 cm in diameter) were drilled to drain excess water. The soil contained 13.7 g/kg organic carbon, 54.95 mg/kg available nitrogen, 24.25 mg/kg Olsen-phosphorus and 105.03 mg/kg available potassium. Soils of each pot was pre-mixed with 0.7 g of N, 0.3 g of $P_2O_5$ and 0.7 g of $K_2O$, and another 0.4 g of N per pot was applied at jointing stage. Twelve seeds were sown per pot on 5 November 2013, and then thinned to four plants at the three-leaf stage.

Four treatments were established: CK (control, non-stressed plants), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading). The WA treatment was achieved by keeping a 2-cm water layer above the soil surface (pots were placed in an artificial pool and the depth of water layer adjusted manually). For the SA treatment, a black polyethylene screen was fitted about 180 cm above the ground to block about 80% of the total radiation. The WS treatment was achieved by combining both waterlogging and shading treatments. All treatments started when jointing was reached (5 March 2014), and there were three durations (5, 10 and 15 d respectively) for each treatment. At the end of each treatment, excess water was drained and the black polyethylene screen removed.

### 2.2 Sampling

Eight plants from two pots of each treatment were sampled from jointing stage at 5-d intervals until all treatments were completed (15 d after jointing) and on 25 d after jointing for observation of recovery status. Height and tiller number per plant of each plant were recorded. Samples were immediately hand separated as stem, green leaf (green area occupies $\geqslant$ 50% in a leaf) and yellow leaf (yellow area occupies $\geqslant$ 50% in a leaf) after sampling; they were initially heated for 30 min at 105°C and dried to constant weight at 80°C for measurements of dry matter.

Two pots of each treatment were retained until maturity (20 May 2014) for measurement of yield and its components. Generally, kernel weight per plant (KW), spike number per plant (SN), kernel number per spike (KN) and thousand-kernel weight (TKW) were recorded.

### 2.3 Calculation of partitioning indexes of dry matter

Partitioning indexes were calculated to quantify the effects of different treatments on dry matter distribution to avoid the differences between plants and cultivars, according to the equations below [21].

PIS (partitioning index of stem, %)= dry matter weight of stem/dry matter weight of aboveground biomass × 100 (1)

PIGL (partitioning index of green leaf, %)= dry matter weight of green leaf/dry matter weight of aboveground biomass × 100 (2)

PIYL (partitioning index of yellow leaf, %)= dry matter weight of yellow leaf/dry matter weight of aboveground biomass × 100 (3)

### 2.4 Statistical analysis

One-way analysis of variance was conducted to determine significant differences between the treatments. The least significant difference (LSD) between means was estimated using $P<0.05$ as the standard for significance. Statistical analysis was performed using the SPSS 19.0 for Windows software package.

## 3 Results

### 3.1 Partitioning index of dry matter

SA treatment of different durations had a limited effect on height of wheat compared with CK ($P>0.05$). Waterlogging treatment had an obvious effect on height up to the maximum duration. At 5 or 10 d of WA and WS there was a moderate effect on height, but a significant decrease after 15 d of WA and WS for both Ningmai 13 and Yangmai 13 ($P<0.05$, Fig. 1). Moreover, after the removal of waterlogging stress, height for WA did not increase after 10 d of recovery, but significantly increased for WS ($P<0.05$). Thus, WA had a greater impact on height than WS.

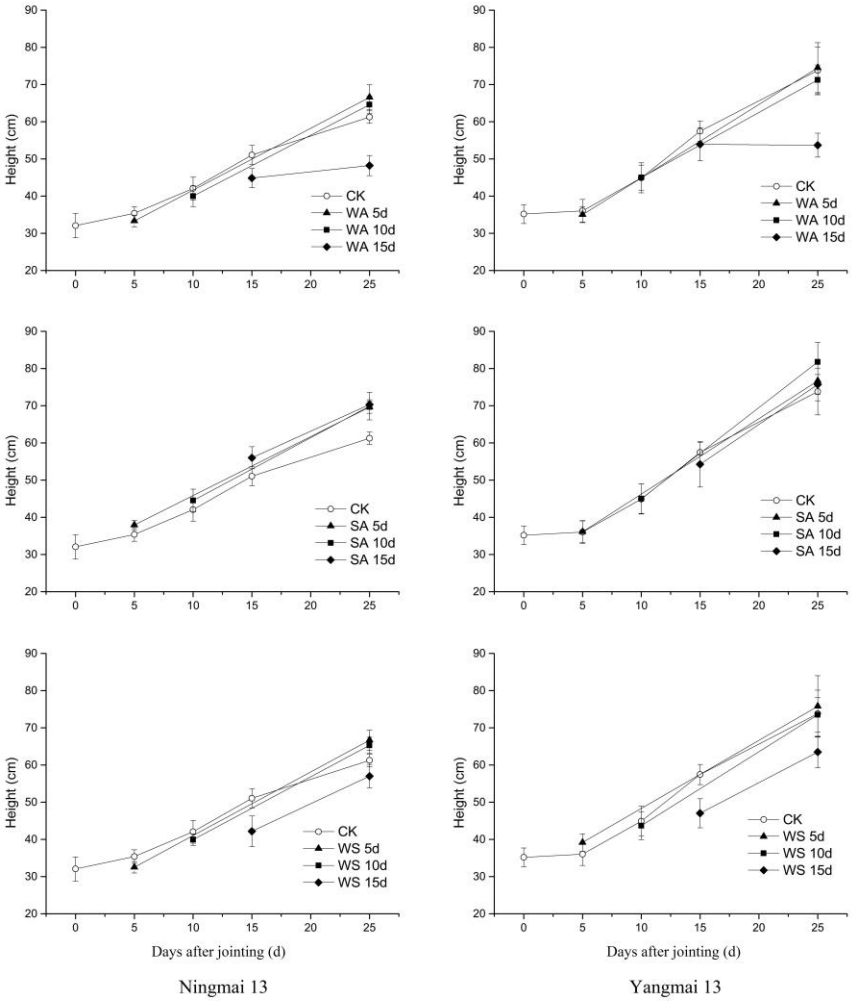



**Fig. 1.** Wheat height under treatments of CK (control), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading) at jointing stage.

Tiller number per plant decreased under most treatments compared with CK (Fig. 2). However, the decrease was not significant, indicating that continuous rain had a limited effect on tiller number of winter wheat at jointing stage.

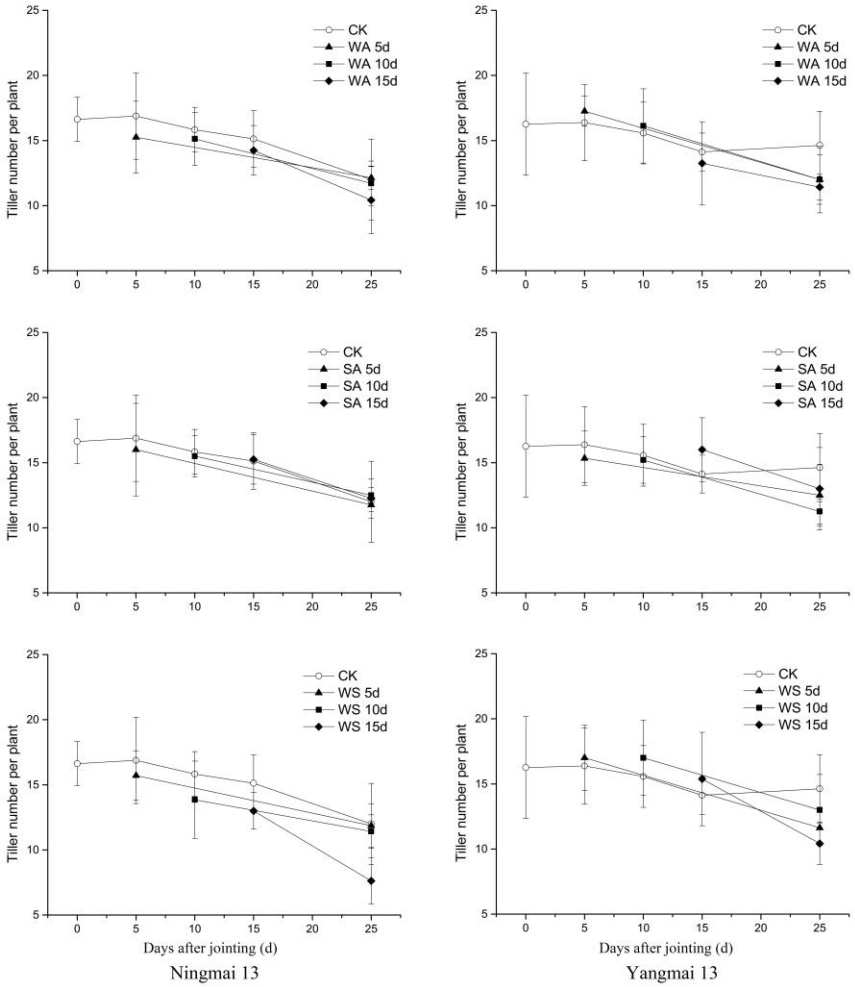



**Fig. 2.** Tiller number under treatments of CK (control), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading) at jointing stage.

WA had no significant effect on PIS, while PIS significantly decreased under both SA and WS compared with CK, especially for duration over 5 d. Although PIS had a significantly lower level under stress of SA and WS (Fig. 3), there was a quick recovery after removal of stress. There were no obvious differences in PIS between treatments and CK after a 10-d recovery.

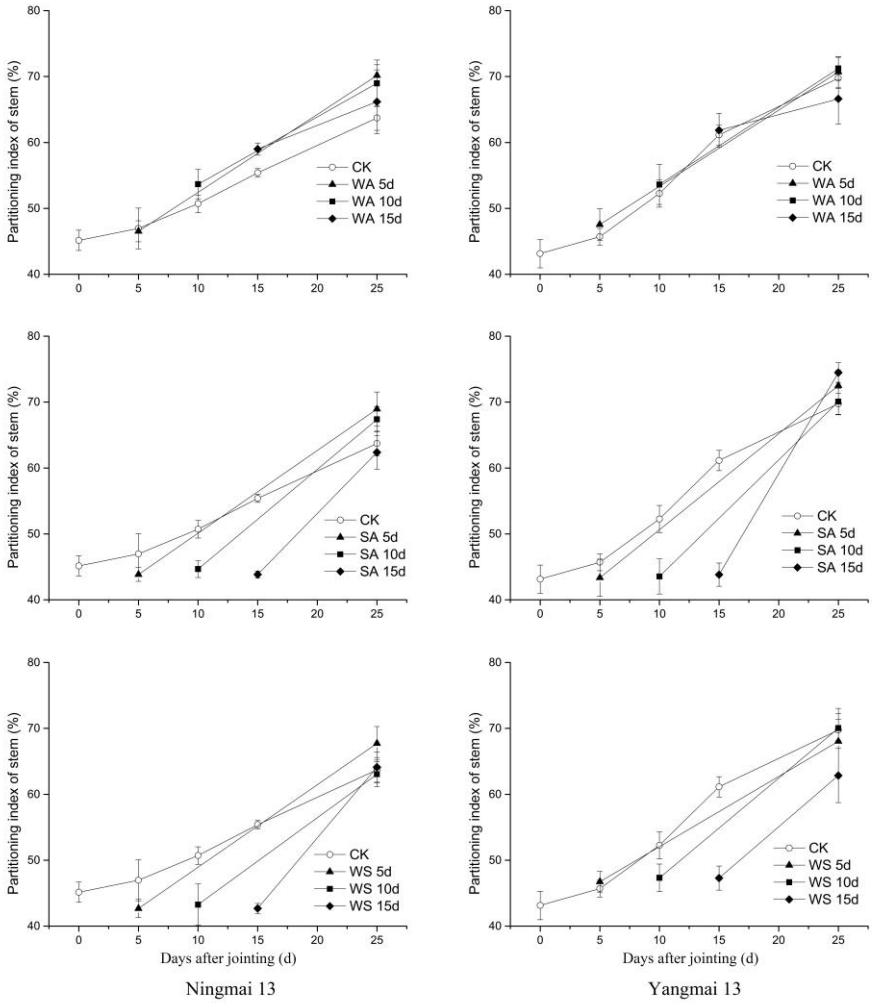



**Fig. 3.** Partitioning index of stem (PIS) under treatments of CK (control), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading) at jointing stage.

PIGL showed opposite variations under WA and SA compared with CK: a decrease for WA and an increase for SA. WS induced an increase similar to SA; however, with a lower increment. After removal of stresses, the gaps in PIGL between treatments and CK were narrowed (Fig. 4).

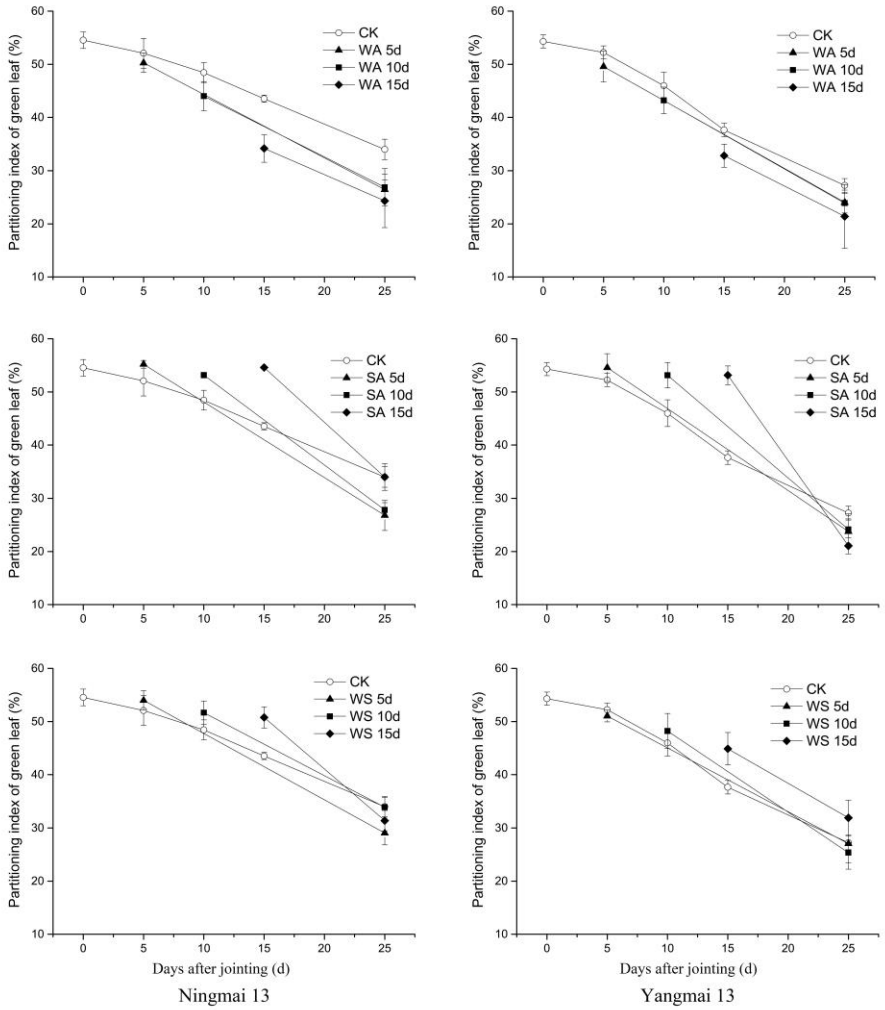



**Fig. 4.** Partitioning index of green leaf (PIGL) under treatments of CK (control), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading) at jointing stage.

PIYL was a sensitive indicator of waterlogging and shading stresses. PIYL under WA increased with duration and was significantly higher than for CK (Fig. 5). Furthermore, PIYL under WA for 15 d increased even after the removal of stress. PIYL increased for SA compared with CK but was moderate relative to that for WA. PIYL increased with duration of WS and was significantly higher compared with CK, similar to that for WA. However, PIYL under WS was close to CK values during the recovery period (including WS for 15 d), which meant that WS caused temporary damage while WA caused permanent damage to leaves of wheat under long-term stress. The negative effects of the treatments on PIYL were in the order of WA > WS > SA.

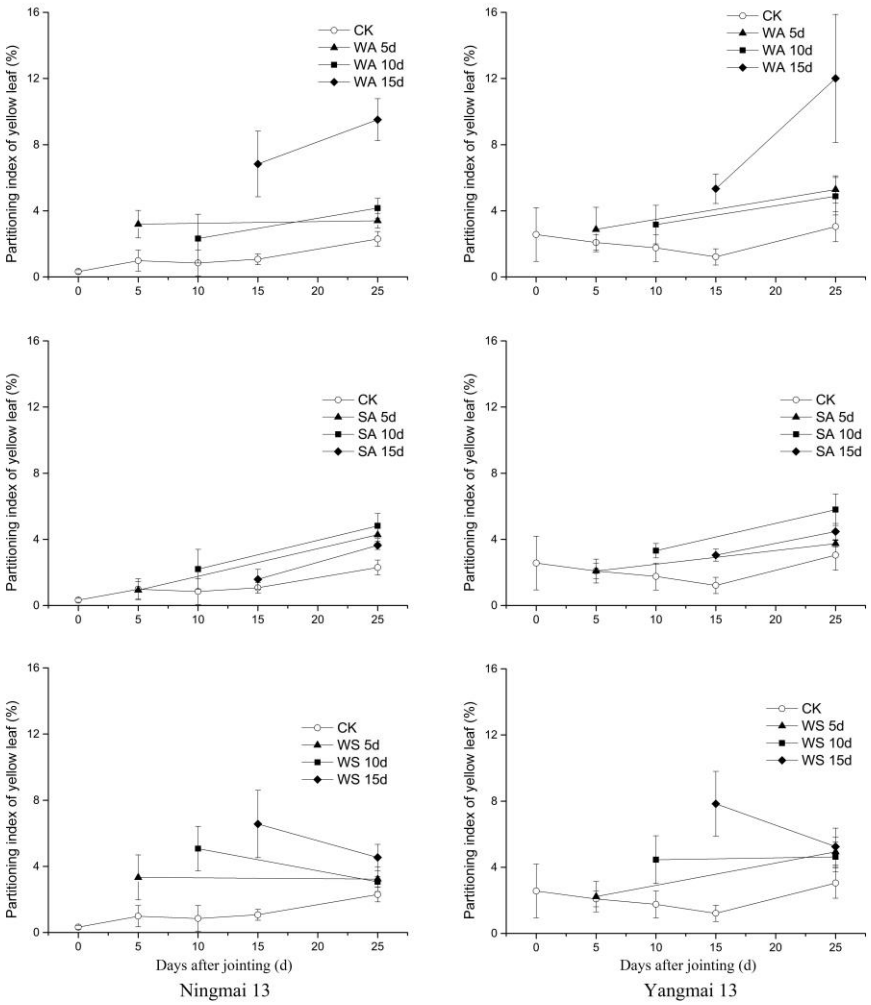



**Fig. 5.** Partitioning index of yellow leaf (PIYL) under treatments of CK (control), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading) at jointing stage.

### 3.2 Yield and yield components

With increased duration of WA treatment, KW of both cultivars decreased. After 15 d of waterlogging, KW significantly decreased (14.25 to 9.57 g and 15.97 to 9.40 g for Ningmai 13 and Yangmai 13, respectively; $P<0.05$). The effects of SA and WS on KW depended on the cultivar. KW of Ningmai 13 under SA and WS decreased non-significantly compared with CK. However, there were greater decreases in KW for Yangmai 13 under SA and WS and were significant when duration reached 10 d. Under all stresses, KW was significantly reduced for Yangmai 13 and was lower than for Ningmai 13 (Table 1). Thus, Ningmai 13 had better tolerance to stresses than Yangmai 13.

All three treatments had a limited effect on SN compared with CK. The only significant reduction was for Yangmai 13 with 15 d of WS (Table 1).

KN and TKW showed no significant differences for both cultivars under SA. There was a significant reduction of KN for both cultivars under WA (Table 1). Ningmai 13 showed good tolerance to WA and WS with no significant decreases of TKW. However, TKW of Yangmai 13 significantly decreased under both WA and WS.

**Table 1** Wheat yield and its components under different treatments [CK (control), WA (waterlogging alone), SA (shading alone) and WS (combined waterlogging and shading)] and duration (5, 10 and 15 d).

| KW (g) | Ningmai 13 | Duration | CK | WA | SA | WS | Yangmai 13 | Duration | CK | WA | SA | WS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0 | 14.25 | a | a | a | | 0 | 15.97 | a | a | a |
| | | 5 | | 11.49a | 12.42a | 12.87a | | 5 | | 12.33ab | 11.86ab | 12.67ab |
| | | 10 | | 12.77a | 12.91a | 13.13a | | 10 | | 13.15ab | 11.91ab | 11.76b |
| | | 15 | | 9.57b | 14.98a | 12.59a | | 15 | | 9.4b | 10.34b | 11.52b |
| SN | Ningmai 13 | Duration | CK | WA | SA | WS | Yangmai 13 | Duration | CK | WA | SA | WS |
| | | 0 | 7 | a | a | a | | 0 | 7.88 | a | a | a |
| | | 5 | | 6.75a | 6.75a | 7.5a | | 5 | | 7.25a | 6.75a | 7ab |
| | | 10 | | 7a | 7.5a | 6.75a | | 10 | | 7.25a | 7.5a | 6.63ab |
| | | 15 | | 6.63a | 7.5a | 6.38a | | 15 | | 7.25a | 7a | 6.38b |
| KN | Ningmai 13 | Duration | CK | WA | SA | WS | Yangmai 13 | Duration | CK | WA | SA | WS |
| | | 0 | 47.48 | a | a | a | | 0 | 42.53 | a | a | a |
| | | 5 | | 41.63b | 42.39a | 41.03b | | 5 | | 39.05ab | 43.61a | 43.83a |
| | | 10 | | 44.25ab | 46.18a | 43.17ab | | 10 | | 41.69a | 39.66a | 45.12a |
| | | 15 | | 40.47b | 44.27a | 47.54a | | 15 | | 32.18b | 36a | 42.4a |
| TKW (g) | Ningmai 13 | Duration | CK | WA | SA | WS | Yangmai 13 | Duration | CK | WA | SA | WS |
| | | 0 | 42.51 | a | a | a | | 0 | 46.89 | a | a | a |
| | | 5 | | 40.75a | 42.39a | 41.91a | | 5 | | 43.6ab | 40.56a | 42.85ab |
| | | 10 | | 41.87a | 46.18a | 45.19a | | 10 | | 43.67ab | 40.22a | 39.98b |
| | | 15 | | 36.39a | 44.27a | 41.4a | | 15 | | 40.57b | 40.34a | 42.99ab |

KW, kernel weight per plant; SN, spike number per plant; KN, kernel number per spike; TKW, thousand-kernel weight;

Different letters following values in a column indicate significant differences at $P<0.05$ by LSD.

Wheat yields under different treatments showed an order of CK > SA > WS > WA. Ningmai 13 had a significant yield reduction only under WA, mainly due to the reduction of SN. Yangmai 13 had significant yield reductions for all three treatments for different reasons: WA by decrease of KN and TKW; SA by decrease in KN; and WS by decrease in SN and TKW.

## 4 Discussion

Wheat genotypes vary significantly in their sensitivity to weather stresses, such as heat [22] and waterlogging[11]. Both Ningmai 13 and Yangmai 13 are major winter wheat cultivars in Jiangsu Province. Under non-stressed conditions (CK), Yangmai 13 produced greater yield than Ningmai 13 (15.97 vs 14.25 g, respectively). However, Yangmai 13 suffered larger yield reductions under WA, SA and WS compared with Ningmai 13 (Table 1). When duration of stresses lasted 10 or 15 d, Ningmai 13 had better production, indicating that it had better tolerance to waterlogging and shading at jointing stage. Thus, when choosing a winter wheat cultivar, both potential yield (without stress) and practical yield (with stress) should be considered. Due to global climatic change, frequency of continuous rain is increasing in the lower basin of the Yangtze River, where Jiangsu Province is located [5, 23]. Therefore, future work should focus on screening and breeding wheat cultivars with better tolerance to waterlogging and shading to reduce or avoid the negative effects of continuous rain on wheat production.

The WA, SA and WS treatments were used to simulate the stresses of continuous rain. Comparing the effects of the different treatments on physiology of wheat showed that the negative effects of stresses were in the order WA > WS > SA. SA had the least effect on wheat of the three treatments mainly due to compensation from diffuse light and physiology partially balances shading stress [17, 18]. Moreover, physiology and growth of wheat recovered soon after shading stress was removed. There were 60 d for the recovery of wheat from jointing (20 March) to maturity (20 May) in the present study. Thus, shading at jointing stage had a limited effect on wheat production.

Waterlogging had a major effect on wheat physiology and production, and the effect was amplified with increased duration. Studies have suggested that grain yield is reduced by 20–50% if wheat suffers waterlogging in vegetative stages (Hossain et al., 2011), and results of the present study also showed that 15 d of waterlogging at jointing stage significantly decreased wheat production (by 33 and 41% for Ningmai 13 and Yangmai 13, respectively). The yield reduction under WA was mainly due to severe damage to leaves. The result of the present study showed that PIGL significantly decreased while PIYL increased (Figs. 4 and 5).

Compared to independent waterlogging and shading stresses, the combined stress (WS) is a more realistic situation in a continuous rain event. Comparing the negative effects of all treatments on dry matter distribution, photosynthesis and yield, WS had a less negative effect compared with WA, indicating a compensative rather than an additive effect of shading on waterlogging. Combined stresses often indicate that one stress limits plant growth so strongly that a second stress has little additional impact [20, 24]. Past and present research has shown that waterlogging mainly affects the relative allocation of carbohydrate between leaves and roots, whereas shade operates on allocation between stems and roots [20]. Since leaves determine the photosynthesis of plants and have a more important effect on production than stems, waterlogging has more impact on plant growth than shading [25], and so shading showed no additive effect when accompanying waterlogging.

The compensative effect from shading under WS might be for both morphological and biochemical reasons. Waterlogging may induce leaf senescence and impair leaf area expansion [8, 25]. In contrast, leaves become larger and thinner as a response to shading – enhancing light interception and reducing respiratory costs [26]. Thus PIGL was higher under WS than WA (Fig. 4). In addition, phytotoxins – e.g. reactive oxygen species [27], ethylene [28] and methane [29] – generated during waterlogging due to the anoxic environment cause severe damage to plants. However, plants under WS are likely to suffer less damage compared with WA, since PIYL and height could recover under WS but not under WA after the removal of stress. These results indicated that shading might relieve the effect of phytotoxins either by reducing their levels or enhancing tolerance to them. Further study should investigate the biochemical mechanism of this combined stress on wheat.

## 5 Conclusions

Waterlogging and shading always reduce winter wheat production. Using treatments of independent and combined stress, the present study showed that the negative effects on production and dry matter distribution with an order of WA > WS > SA. This indicated that shading at jointing stage had a compensative effect on waterlogging. In continuous rain events, which have waterlogging and shading, production loss might be less than for waterlogging alone. Thus, the present study advances understanding on the physiology of winter wheat under combined stress and could further improve growth models to avoid overestimating production losses due to continuous rain at jointing stage of winter wheat.

## Acknowledgment

This study was funded by the Fund of Jiangsu Academy of Agricultural Sciences (No. 6111647), the National Natural Science Foundation of China (No. 31800358), the Research Fund of State Key Laboratory of Soil and Sustainable Agriculture (Y412201417), the Special Fund for Agro-scientific Research in the Public Interest (201203032), and the Fund for Independent Innovation of Agricultural Sciences in Jiangsu Province (No. CX(16)1038).

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