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Article

Effects of Multi-Stage Continuous Drought on Photosynthetic Characteristics, Yield and Water Use Efficiency of Winter Wheat

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Abstract: A drought event can cause entire crops to fail or yield loss. In order to study the effects of continuous drought on photosynthetic characteristics, yield, and water use efficiency (WUE) of winter wheat (*Triticum aestivum* L.), the winter wheat variety "Aikang 58" was selected as test material with controlling the water of the pot-planted winter wheat under a mobile rainout shelter. Based on foot planting and safe wintering, winter wheat was evaluated under different drought conditions, including light, moderate and severe drought at the jointing (B), heading (C), and filling (G) stages. The soil water content was controlled in a range of 60% to 70%, 50% to 60%, and 40% to 50% of the field capacity, respectively. In the experiment, there were 9 single-stage droughts, 3 three-stage droughts, and 1 test control (totaling 13 trials). The results are as follows: Under a single-stage drought, the change of net photosynthetic rate (Pn) and stomatal conductance (Gs) have similar trends, and they both decrease significantly with the severity of the drought. Under three-stage continuous droughts, the change curve of Gs shows a constant downward trend; the change curve of Pn showed a "valley shape," and the minimum value of Pn appeared at the heading stage. All droughts will reduce the yield of winter wheat. Under the three-stage continuous drought conditions, except for light drought, moderate drought and severe drought will cause significant yield reduction, mainly due to lack of water at the jointing and heading stages. Continuous drought will reduce the WUE, and the difference will reach a significant level under moderate and severe drought. The present results suggested that when water resources are scarce, it is a better irrigation model to save water and achieve high grain yield by applying appropriate water stress (60%–70% FC) during the critical growth period of winter wheat.

Keywords: Continuous drought; net photosynthetic rate; stomatal conductance; *Triticum aestivum* L; water use efficiency; grain yield

Abbreviations

B	jointing stage
C	heading stage
EEN	effective ear number

FC	field capacity
G	filling stage
GPS	grains per spikes
Gs	stomatal conductance
IA	irrigation amount
NI	normal irrigation
Pn	net photosynthetic rate
TGW	1000-grain weight
WUE	water use efficiency
1	light drought
2	moderate drought
3	severe drought

1 Introduction

Wheat is one of the most important cereal crops in the world, and it is China's third-largest food crop. The annual output is about 110 million tons, accounting for about 40% of total food production [1,2]. The winter wheat planting area accounts for more than 80% of the national wheat, and the total yield reached 90% [3]. Water supply and utilization efficiency are critical to the growth and development of winter wheat [4]. Affected by food habits, natural environment, and the level of economic development, winter wheat is widely planted in the arid and semiarid area in northern China. However, in these areas, precipitation during the growth stage of winter wheat is often inadequate, and the spatial and temporal distribution of precipitation is hugely uneven. Also, the precipitation process is not synchronized with the critical water demand period of winter wheat [5]. These complex factors eventually lead to different degrees of drought stress during the growth stage of winter wheat. Frequent drought events are the main reasons for the low and unstable winter wheat yield [6]. Drought directly limits crop growth and grain yields in a manner that significantly affects economic development and food security [7,8]. Therefore, it is of far-reaching significance to continuously study the effects of drought on winter wheat drought in-depth for agricultural disaster prevention and to maintain social and economic prosperity and stability.

Drought is a recurring extreme climate event over land characterized by below-normal precipitation over months to years [9]. Arid areas are usually suffering from continuous drought with prolonged duration and different intensities during the winter wheat seasons. In recent years, a large number of experts and scholars have studied the effects of drought on photosynthetic characteristics and grain yield, and have achieved results that have both theoretical significance and practical value. Most of these results were obtained based on the artificially controlled drought test methods. At a particular growth stage of winter wheat, the soil moisture was maintained at a specific level, to observe changes in winter wheat physiological functions, growth and development, and yield components; Or after artificially controlling and maintaining short-term drought at a particular growth stage of winter wheat, rewatering with different irrigation amounts is performed to analyze the recovery status of the rehydrated crops after drought [10–12]. For example, Pei et al. [13] studied the effects of water deficit at different growth stages on the growth and physiological characteristics of winter wheat. They found that soil water regulation had effects on winter wheat plant height, leaf area, photosynthesis rate, transpiration, and water use efficiency (WUE). Drought stress can inhibit photosynthesis in leaves, and the supply and demand balance between CO_2 produced by plant and CO_2 required for photosynthesis will be disrupted, resulting in a decrease in photosynthetic rate and stomatal conductance [14], which in turn affects growth conditions and yield [15,16]. Other studies showed that drought at different growth stages has different effects on crop phenophase and yield. The final yield loss is not only related to the intensity of drought stress but also the growth stage of the crop [17,18]. Drought during the jointing stage to the booting stage can cause a large number of inflorescences of florets and reduce the number of grains per spike, while lack of water during the filling stage will make the wheat grains slim and shrivelled [19,20]. Besides, the predecessors studied the water sensitivity index of winter wheat based on the Jensen model. They used this model to better express the relationship between crop water demand and winter wheat yield [21,22].

In summary, previous studies have focused on the effects of short-term drought stress on the growth and yield of winter wheat in a single growth stage, and there are few studies on the effects of continuous drought in multiple growth stages on winter wheat. However, most of the winter wheat is planted in the monsoon region and is extremely prone to seasonal drought. Drought in the early stage of winter wheat will have a significant effect on growth and development in the later stage. Researchers found that with the prolongation of drought time and the intensification of stress, the inhibition of winter wheat growth and development was gradually increased [2]. According to the actual situation, it is necessary to carry out further research on the effects of continuous drought on the growth and development of winter wheat.

This study aimed at the current winter wheat drought experiment and the actual situation and the effects of continuous drought on winter wheat were not systematically studied, and cut from the jointing stage to the filling stage with the highest drought frequency during the growing period of winter wheat. The pot-planted test method was used to study winter wheat in North China Plain (NCP), and the effects of continuous drought at different stages and different degrees on photosynthetic characteristics, yield, and WUE of winter wheat were studied. The purpose is to further study the response mechanism of winter wheat to continuous drought, and to provide theoretical references for optimizing irrigation systems, improving WUE, ensuring food security, and predicting the effects of continuous drought stress on the development trend of wheat and other dry crops in actual future production.

2 Materials and Methods

2.1 Experimental Design and Field Practices

A pot experiment was conducted from October 2016 to June 2017 at the Chinese Academy of Agricultural Sciences, Qiliying experiment base, Xinxiang, Henan Province of China (35°18’N, 113°54’E, Altitude 81 m). The study area has a warm temperate continental monsoon climate zone, with mean annual precipitation, potential evapotranspiration, air temperature, accumulated temperature sunshine duration and a frost-free period of 580 mm, 2000 mm, 14°C, 5070°C, 2399 h and 210 days, respectively.

The winter wheat variety Aikang 58 was sown on October 15, 2016.

Experiment consisted of different drought levels at the jointing (jointing–heading stage), heading (heading–filling stage) and filling stage (filling–mature stage), namely normal, light drought, moderate drought and severe drought, corresponding to 70%–80%, 60%–70%, 50%–60% and 40%–50% of the field capacity, respectively. Due to limited funding and space, the full permutation and combination tests were not adopted. After the screening, there were 9 single-stage droughts, 1 normal irrigation, and 3 three-stage continuous droughts. There was a total of 13 experimental treatments (Tab. 2). Each treatment was replicated four times, one of which was sampled as a destructive test for a total of 52 pots. The pots used in the experiment was an iron made with a diameter of 40 cm and a height of 60 cm. The tested soil was loam. The soil bulk density of each pot was controlled at 1.36 g/cm^3 (Tabs. 1-1 and 1-2). The measuring pot was placed in a 60 cm underground pit and weighed by a hanging scale to monitor the water consumption of the crops. The effective plants were selected at the three leaves stage and 60 plants per pot.

All agronomic measures were consistent except for the difference in moisture in all test treatments. Before the sowing, 10 g compound fertilizer (N, P, K = 15:15:15), and 3.5 g urea (pure nitrogen levels ≥ 46%) was applied as basal does through topdressing in the early stage of jointing stage.

Table 1-1: Basic properties of the tested soil

pH	Organic matter/ ($mg \cdot g^{-1}$)	Total N/ ($mg \cdot g^{-1}$)	Available N/ ($mg \cdot g^{-1}$)	Available P/ ($mg \cdot g^{-1}$)	Available K/ ($mg \cdot g^{-1}$)	Field Capacity (%)
8.72	17.8	0.89	35.5	137.19	50.119	21.0

Table 1-2: Soil parameters of the experimental station

Soil Depth (cm)	Granulometric Composition (%)			Soil Texture
	Clay (<0.002 mm)	Silt (0.002~0.02 mm)	Sand (0.02~2 mm)	
0~20	6.75	69.72	23.53	Loam
20~40	6.41	66.91	26.69	Silty Loam
40~60	10.19	69.96	19.85	Silty Loam
60~80	10.16	73.44	16.41	Silty Loam
80~100	8.22	75.74	16.05	Sandy Soil

Table 2: The pot-planted experimental design

Number	Treatment	Experimental treatment levels					Remark
		Green Stage (172 d)	Jointing Stage (15 d)	Heading Stage (15 d)	Filling Stage (12 d)	Mature Stage (21 d)	
1	NI	Normal	Normal	Normal	Normal	Normal	No Drought
2	B[a]1[b]	Normal	**Light**[c]	Normal	Normal	Normal	Single-stage Drought
3	B2	Normal	**Moderate**	Normal	Normal	Normal	
4	B3	Normal	**Severe**	Normal	Normal	Normal	
5	C1	Normal	Normal	**Light**	Normal	Normal	
6	C2	Normal	Normal	**Moderate**	Normal	Normal	
7	C3	Normal	Normal	**Severe**	Normal	Normal	
8	G1	Normal	Normal	Normal	**Light**	Normal	
9	G2	Normal	Normal	Normal	**Moderate**	Normal	
10	G3	Normal	Normal	Normal	**Severe**	Normal	
11	B1C1G1	Normal	**Light**	**Light**	**Light**	Normal	Continuous Drought
12	B2C2G2	Normal	**Moderate**	**Moderate**	**Moderate**	Normal	
13	B3C3G3	Normal	**Severe**	**Severe**	**Severe**	Normal	

[a]B,C and G represent jointing stage, heading stage and filling stage, respectively.
[b]1,2 and 3 represent light drought, moderate drought and severe drought, respectively.
[c]Normal, Light, Moderate, Severe indicate that soil water content of 70%–80%, 60%–70%, 50%–60% and 40%–50% of the field capacity, respectively.

2.2 Measurement

2.2.1 Soil Water Content and Irrigation Amount

In order to guide the irrigation scheduling, the soil samples were taken from the depth of 10–30 cm and 30–50 cm, once every growth period starting from the tillering stage. The soil water content was determined by the oven drying method. The average water content of the two soil layers was measured as the initial soil water content at the growth stage. After entering the jointing stage, the test winter wheat was controlled to three levels of light drought, moderate drought and severe drought through natural evaporation and quantitative irrigation.

Irrigation was applied when the soil water content of a specific growth stage was reached to the set limit of the experimental treatment, and each irrigation amount was calculated according to Eq. (1). The measuring cup was used for quantification of the amount of water. After each irrigation, the pots were weighed every 1~2 days, the mass change of the measuring pot represents the water-consumption of crops, and the next irrigation time was predicted.

$$W = \gamma HA(\theta_1 - \theta_2) \tag{1}$$

where W is the irrigation amount (g); γ is the soil dry bulk density (g/cm^3); H is the scheming wetted soil layer depth (cm); A is the cross-sectional area of the pot (cm^2); θ_1 is the upper limit water content of the design irrigation; θ_2 is the soil water content before irrigation.

2.2.2 Water Consumption and Water Use Efficiency

The soil water content was monitored during the whole growth period, and the soil water content of the planned moisture layer was measured by the drying method before and after each growth stage. The water consumption was calculated by daily weighing. The evapotranspiration of winter wheat was calculated using the soil water balance equation [28].

$$ET_0 = I + P - R - D - SW \tag{2}$$

$$ET = ET_0 \times A \tag{3}$$

where ET_0 is the total evapotranspiration of the winter wheat growing season (mm); I is the irrigation water (mm); P is the effective precipitation (mm); R is the surface runoff (mm); D is the deep percolation (mm); SW is the change in soil water content in the wet layer from planting to harvest (mm); ET is the total evapotranspiration per pot in the winter wheat growing season (m^3).

The experiment was performed in the bottomed pots under a rain shelter; thus P, R, and D can be ignored. Calculate water use efficiency based on the yield and water consumption of each treatment.

$$WUE = Y/ET \tag{4}$$

where WUE is the water use efficiency (kg/m^3); Y is the yield (kg).

2.2.3 Physiological Characteristics

The photosynthetic parameters of flag leaves were measured every 7 days from the beginning of the winter wheat treatment to the corresponding growth stage. By selecting fine weather, the photosynthetic rate (Pn) and stomatal conductance (Gs) were measured from the flag leaves between 09:00 and 11:00 using the LI-6400XT portable photosynthesis system (LI-COR, USA), following the user manual instructions. Test data were recorded as the average from three different pots in each treatment. During the measurement, the CO_2 concentration in the leaf chamber was set to 400 μmol/mol, the flow rate was set to 500 μmol/(m^2s^2), and an artificial light source LED provided the light intensity in the system, and set to 1200 μmol/(m^2s). In the face of cloudy weather, before actually measuring each sample, induce with LI-6400XT at a light intensity of 1200 μmol/(m^2s) for 30 min.

2.2.4 Yield and Yield Components

After the winter wheat was artificially harvested on June 1, it was dried, threshed, tested for seed production, and counted for production. Its effective panicle number, grains per spike, 1000-grain weight, and economic yield were determined.

2.3 Statistical Analysis

The data were processed and graphed in Microsoft Excel 2016. Analysis of variance (two-way ANOVA) was performed using SPSS software version 25.0 (SPSS Inc., USA). Statistical differences in photosynthetic parameters, yield, WUE were examined using LSD multiple comparison test and declared at the 0.05 probability level. Pearson correlation coefficients were conducted to represent the relationships between drought and winter wheat indicators.

3 Results and Discussion

3.1 Net Photosynthetic Rate and Stomatal Conductance

Stomatal conductance is an important parameter reflecting leaf exchange capacity and is closely related to leaf photosynthetic rate.

The changes in net photosynthetic rate and stomatal conductance of the pot-planted winter wheat during the test growth stages are shown in Figs. 1 and 3. The changing trend of Pn is similar to that of Gs. Compared with NI, the net photosynthetic rates of B1, B2, and B3 measured at the jointing stage on April 13 decreased by 1.8%, 24.2%, and 38.1%, respectively; the stomatal conductance decreased by 16.5%, 29.6%, and 73.4%, respectively. The net photosynthetic rates of C1, C2, and C3 measured at each heading stage on April 23 decreased by 4.7%, 27.5%, and 58.2%, respectively; the stomatal conductance decreased by 16.2%, 30.4%, and 65.2%, respectively. The net photosynthetic rates of G1, G2, and G3 measured at the filling stage on May 5 decreased by 5.1%, 20.6%, and 46.5%, respectively; the stomatal conductance decreased by 3.8%, 17.5%, and 56.6%, respectively. Pearson correlation analysis showed that the correlation coefficients between drought stress and the net photosynthetic rate at the jointing, heading and filling stages were –0.96 ($p < 0.05$); the correlation coefficients of stomatal conductance were –0.96, –0.97, –0.97 ($p < 0.05$). The regression curve was established with the degree of drought stress as the independent variable x, and Pn and Gs at the jointing, heading and filling stage as the dependent variables y (Figs. 2 and 4). The Pn regression equations are $y = –2.004x + 16.24$, $y = –3.648x + 19.38$, and $y = –2.154x + 16.775$, all of which are linear subtraction functions. The Gs regression equations are $y = –51.07x + 285.8$, $y = –31.79x + 208.6$, and $y = –26.81x + 184.6$, all of which are linear subtraction functions. These show that drought stress can significantly reduce Pn and Gs at jointing, heading and

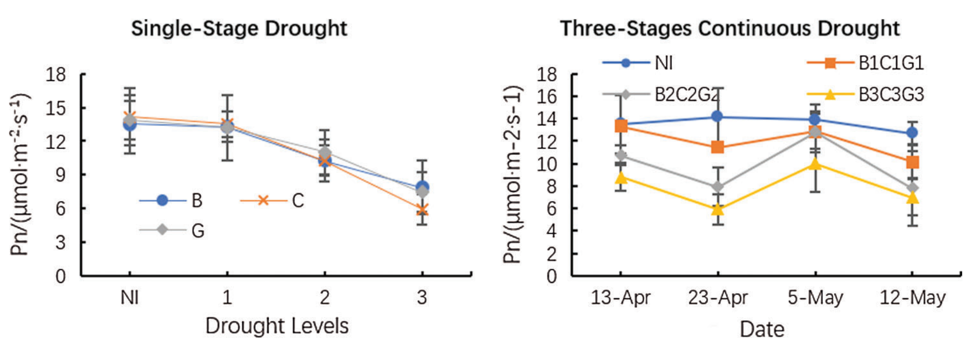

Figure 1: Effects of different degrees of drought on net photosynthetic rate of winter wheat. B, C and G represent the jointing stage, heading stage and filling stage, respectively; 1, 2 and 3 represent light drought, moderate drought and severe drought, respectively

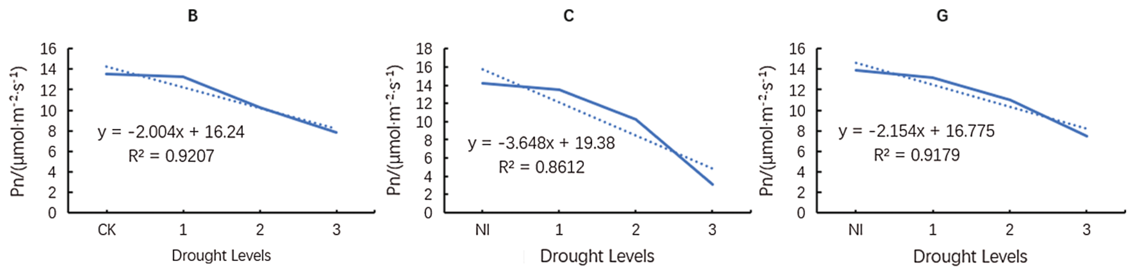

Figure 2: Simple regression model of drought stress and net photosynthetic rate of winter wheat

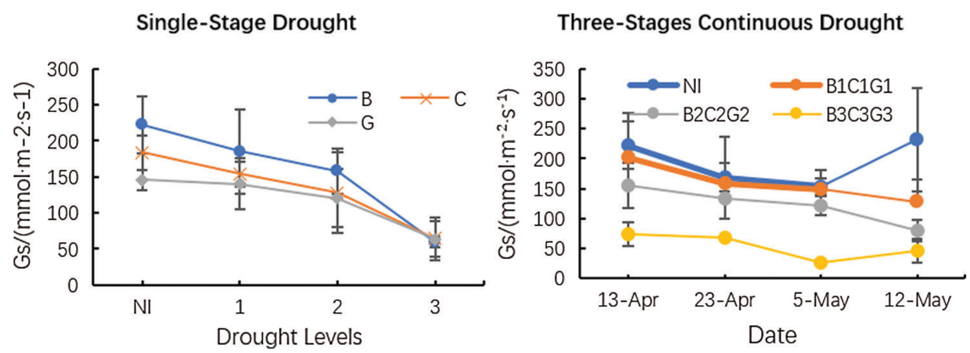

Figure 3: Effects of different degrees of drought on stomatal conductance of winter wheat

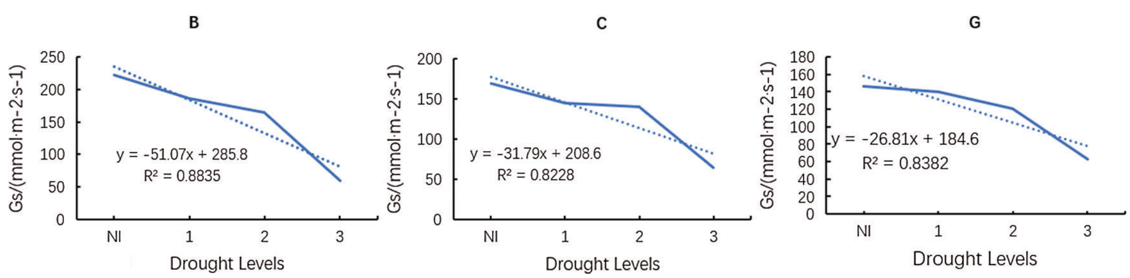

Figure 4: Simple regression model of drought stress and stomatal conductance of winter wheat

filling stage and that Pn at the heading stage is most sensitive to drought stress, and Gs at the jointing stage is most sensitive to drought stress.

In three-stage continuous drought treatment, the Pn curve of winter wheat showed a "valley shape" under different levels of drought stress, and the lowest value of Pn appeared at the heading stage. However, the changing trend of the change curve of Gs is consistent, and all of them gradually decreases. Compared with NI, the Pn measured at the jointing, heading and filling stage in the B1C1G1 treatment decreased by 1.6%, 19.1%, and 7.8%, respectively; Gs decreased by 9.0%, 5.9%, and 3.2%, respectively. In the B2C2G2 treatment, Pn decreased by 20.8%, 44.1%, and 7.8%; Gs decreased by 30.1%, 21.6%, and 21.2%, respectively. In the B3C3G3 treatment, Pn decreased by 34.9%, 58.2%, and 28.0%, respectively; Gs decreased by 66.8%, 59.8%, and 82.6%, respectively. Compared with NI, there were no significant differences in Pn and Gs in all treatments under light drought stress. Under moderate drought stress, there was no significant difference between single-stage drought and three-stages continuous drought. Still, under severe drought, Pn and Gs under three-stages continuous drought stress showed significant differences compared with single-stage drought treatment.

The comprehensive comparison found that in 9 single-stage drought treatments and 3 three-stages continuous drought treatments, the effects of drought stress on Pn and Gs in different treatments were: Severe drought > Moderate drought > Light drought. The effects of drought stress on Pn at different growth stages are as follows: three-stages continuous drought > heading stage > filling stage > jointing stage; the effects of Gs in different growth stages were: three-stage continuous drought > jointing stage > heading stage > filling stage. Rehydration was performed after the end of the filling stage. May 12 was the second day after rehydration; thus, the data measured on that day will not be discussed here.

3.2 Yield and WUE

Wheat yield is determined by three factors: effective ear number, grains per spike, and 1000-grain weight [23]. Water stress affects the three factors, which in turn leads to a decline in yield [24]. In addition, the factors and extent of yield reduction are also related to the growth stages under drought stress.

Data shows that drought stress caused simultaneous reductions in effective ear number, grain per panicle, and 1000-grain weight at different growth stages, and the three factors showed a continuous downward trend with the increase in drought severity and increase drought stress time. Therefore, the yield of winter wheat also shows a similar pattern. The more severe the drought, the higher the yield reduction. However, there are significant differences in the sensitivity to drought at different growth stages. From light to severe drought, yield reduction at the jointing stage increased from 4.1% to 36.7%; yield reduction at the heading stage increased from 0.6% to 17.36%; yield reduction at the filling stage increased from 6.3% to 9.9%, and did not cause a significant reduction; yield reduction in three-stage continuous drought treatment increased from 8.0% to 54.0%. All treatments under light drought stress were not significantly different from NI control; Under moderate drought treatment, the yields of B2 and B2C2G2 decreased significantly, and the yield reductions were 21.7% and 25.0%, respectively, but there was no significant reduction in yield during the heading and filling stages; under severe drought treatment, the yield reductions of B3 and B3C3G3 have increased to 36.7% and 54.0%, and the C3 production has also been significantly reduced by 17.4%. Still, the yield during the filling stage was not decreased significantly. Therefore, the sensitivity of different growth stages to the same level of drought stress is: three-stage continuous drought treatment > jointing stage > heading stage > filling stage.

WUE is the economic yield per unit of water produced during the whole growth stage of the crop, taking into account the two processes of yield formation and water consumption.

The test results showed that the water consumption of winter wheat showed a downward trend with the increase of drought and the increase of drought time. Under the same drought degree, the water consumption in different growth stages was as follows: grouting stage > heading stage > jointing stage > three-stage continuous drought. The WUE of all treatments except for the treatment at the filling stage increased with the aggravation of drought, while that of all treatments decreased. Compared with NI, only B2, B3, B2C2G2, and B3C3G3 had a significant decrease in WUE, and the other treatments had no significant difference. Even the WUE of B1, C1, and C2 increased by 0.6%, 2.5%, and 1.3%. For continuous drought treatment, B1C1G1 decreased by 2.5% and 4.3% compared to B1 and C1 but increased by 4.0% compared to G1. This may be due to the pre-anthesis light drought hardened the biological body, which strengthened its physiological functions and stress resistance.

Comprehensive analysis shows that proper water stress during jointing and heading stage can save agricultural water resources to a large extent and improve the water use efficiency of winter wheat. However, in view of production and WUE, the three-stage continuous drought should be avoided in the production (Tab. 3).

3.3 Pearson Correlation Network Diagram

In the Pearson correlation diagram, there is no negative correlation between all parameters, and although there is no direct correlation between individual parameters, there is an indirect correlation. In the

Table 3: Yield and its components of winter wheat under different treatments

Number	Yield ($g \cdot pot^{-1}$)	Effective Ear Number (number)	Grains Per Spike (number)	1000-Grain Weight (g)	Water Consumption (mm)	WUE ($kg \cdot m^{-3}$)
NI	106.33a	70.3a	51.2a	52.6a	268.22	1.58a
B1	101.99ab	67b	44.6b	48.6ab	255.84	1.59a
B2	83.25cd	54c	41.3c	46.5b	252.30	1.31d
B3	67.27e	50.6d	38.4d	44.3c	241.68	1.11bcd
C1	105.72a	69.3ab	47.6ab	47.8b	259.79	1.62a
C2	103.56ab	68b	44.5b	45.4bc	257.38	1.60a
C3	87.87bcd	64.5bc	40.9c	42.1c	250.18	1.40abc
G1	99.66abc	68.5ab	48.5ab	47.4b	265.39	1.49abc
G2	98.52ab	66.3b	44.7b	43.8c	262.56	1.49abc
G3	95.80abcd	62.4bc	42.1c	40.6d	252.65	1.51abc
B1C1G1	97.78abc	61.7bc	46.1ab	46.3b	244.02	1.55ab
B2C2G2	79.79de	54.7c	40.5c	39.6d	220..90	1.28cd
B3C3G3	48.92f	42d	36.7d	35.8e	211.41	0.82e

[a]B, C and G represent jointing stage, heading stage and filling stage, respectively.
[b]1, 2 and 3 represent light drought, moderate drought and severe drought, respectively.
[c]Means in the same column with the different letters indicate significant difference according to Duncan's multiple range test ($p < 0.05$).

photosynthetic parameter group, Gs is directly positively correlated to IA and Pn only; Pn is directly positively correlated to Gs, Yield, and EEN. There is no direct correlation between Pn, Gs, and remaining other parameters of, but there is an indirect positive correlation. IA is directly positively correlated with Gs, TGW, GPS and EEN, and indirectly positively correlated with Yield, Pn and Gs. For the yield traits group, the three factors were not directly related to Gs, and GPS, TGW and Pn were not directly related, but there were also indirect positive correlations. The above results show that under continuous drought, the relationship between photosynthetic physiology and yield traits of winter wheat is more complicated. There are both direct and indirect effects between them, and they are both positively correlated (Fig. 5).

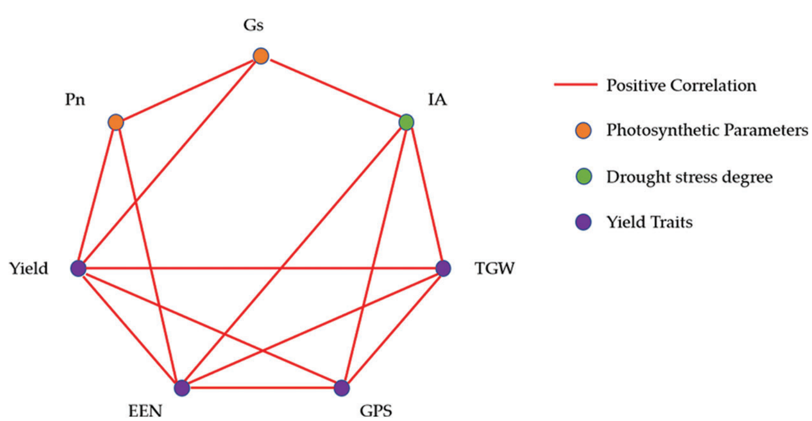

Figure 5: Pearson correlation network diagram on drought stress, photosynthetic parameters, and yield traits. Pn, Gs, IA, EEN, GPS and TGW represent net photosynthesis rate, stomatal conductance, irrigation amount, effective ear number, grains per spikes and 1000-grain weight, respectively.

4 Discussion

Tan et al. [25] believe that the jointing stage to the filling stage is a crucial period affecting wheat photosynthesis. Drought stress will reduce the photosynthesis of winter wheat, which will inevitably limit the productivity and yield. Previous studies have shown the effects of drought at different growth stages with drought levels, duration of drought stress on the net photosynthetic rate (Pn), stomatal conductance (Gs), and intercellular CO_2 concentration (Ci), are quite different [13].

The results of this study indicate that the change curves of Gs and Pn under the single-stage drought condition are consistent, and both continue to decline as the drought stress intensifies. Under the three-stage continuous drought conditions, Gs and Pn showed different trends. Gs showed a constant decline with the increase of the degree of drought and the duration of drought, while Pn showed a "valley shape" change curve, and the minimum value of Pn appears at the heading stage. It is worth noting that from the heading stage to the filling stage, Pn showed an upward trend, but the Pn value was always smaller than the control. According to the analysis of related literature reviewed, the flowering stage and filling stage is the key periods of wheat reproductive growth and grain formation [26]. Water stress will reduce the net photosynthetic rate of crops, inhibit wheat grain filling, and reduce the transport and accumulation of dry matter to grain [27], reduce yield and change grain quality characteristics [28]. In this experiment, the Pn measured at the filling stage and the final yield of each treatment under the three-stage continuous drought conditions was reduced compared with NI, which is consistent with the previous research results. However, the measured Pn at the filling stage showed an upward trend compared with that during the heading stage. This is because after the winter wheat enters the flowering stage, it self-adjusts to the drought stress to ensure the wheat filling process to the greatest extent. This is consistent with Yang et al. [29] found that from the booting stage to the mature stage of winter wheat, the flag leaf Pn showed a trend of rising first and then falling, which was consistent with the conclusion reached the maximum at the flowering stage.

The growth and development of wheat and the formation of final yield are the processes of energy exchange between the crop and the external environment, which is reflected in the process of dry matter accumulation and photosynthesis [30]. Water stress at different growth stages has a high impact on winter wheat yield and its constituent factors [31]. The test results show that all droughts during the growth stages of winter wheat caused a decline in yield and its constituent factors. Under light drought conditions, multiple comparative analysis found that there was no significant difference in yield between B1C1G1, B1, C1, G1, and NI. When agricultural water resources are scarce, in order to save water and ensure food production, appropriate water stress (60% to 70% FC) can be allowed during the critical growth stages of winter wheat. This means that the moderate or severe drought at the jointing stage will significantly reduce the yield, and it is mainly because the water stress at the jointing stage causes the yield of continuous drought treatment to decrease significantly. This is consistent with the results of Sheng et al. [32] that the negative impact of water stress at the jointing stage on photosynthesis during grain filling is even higher than the effect of water stress during grain filling. In the same way, the three-stage severe continuous drought treatment is mainly due to the water stress at the jointing and heading stages reducing the filling rate and shortening the filling time of winter wheat, which ultimately leads to a reduction in yield [33,34]. Therefore, the effect of continuous drought on the yield is mainly concentrated in the jointing and heading stages. The effect of drought on growth in the growth stages is as follows: three-stage continuous drought > jointing stage > heading stage > filling stage.

Zhang et al. [35] pointed out that under the condition of water deficiency, crop yield increases linearly with water consumption, while WUE decreases with increasing water consumption. At the same time, WUE is also different due to different water supply methods and periods. This experimental study shows that in a single-stage drought, the higher the degree of drought, the lower the water use efficiency of wheat. This is because, although the yield and water consumption of wheat decrease with the increase of drought, the yield is more affected by drought. Compared with NI, except for B1, B3, B2C2G2, and B3C3G3, there was no significant difference in the WUE of the other treatments, and even B1, C1, and C2 were higher than the control. This is because proper drought stress at certain stages of crop growth and development can regulate the growth process of crops and distribute assimilating substances to different tissues and organs, thereby achieving the purpose of improving water use efficiency. However, in the fully irrigated control group, part of the water was lost in the form of soil evaporation, which has no practical significance for crop production, and part of it participated in the production of the crop but did not increase the crop yield, only increased the biomass of the crop [36].

Due to the limitation of the test site, and in order to expediently control the soil, fertilizer, water, and other conditions, an open sky pot-planted experiment was adopted. Still, there was some difference between artificial conditions and the natural environment conditions, so the experimental results should be further studied in combination with the actual farmland experiment. Since this experiment only sets a single level of drought for continuous drought treatment, to simulate more realistic conditions, continuous drought treatments with different levels of drought should be set for research in the future. In addition, further research should be done on rehydration after multiple stages of drought or discontinuous drought. Although there are still many shortcomings in this paper, it is still rare to see the impact of winter wheat multi-stage drought on yield. The above conclusions are of great significance for growing economically dry crops in arid regions, improving water productivity, optimizing crop irrigation systems, and ensuring regional food security.

5 Conclusion

The winter wheat crop exhibited diverse responses to light, moderate, and severe drought at the jointing (B), heading (C), and filling (G) stages are as follows:

1. Both the three-stage continuous drought and the single-stage drought will lead to a continuous decrease of Gs. However, Pn at the three-stage continuous drought and single-stage drought have different trends. The three-stage continuous drought showed a "valley" curve that first fell and then rose, and the minimum value of Pn appeared at the heading stage.
2. All droughts will reduce the yield of winter wheat. The main reason for the three-stage drought reduction is water stress during the jointing and heading stages. Under the three-stage continuous drought conditions, all but the light drought will cause a significant reduction in yield.
3. Compared with NI, continuous drought will reduce WUE of winter wheat, and the difference will reach a significant level under moderate and severe drought. Combining yield factors, when water resources are scarce, the three-stage light continuous drought (60% to 70% FC) is a suitable choice for water-saving and ensuring grain production.

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References

1. Xu, B., Miao, W., Dong, Y., Li, B., Xu, S. (2012). Resource distribution and processing quality of commercial wheat germ in China. *Transactions of the Chinese Society of Agricultural Engineering, 28,* 244–249.

2. Lüttger, A. B., Feike, T. (2018). Development of heat and drought related extreme weather events and their effect on winter wheat yields in Germany. *Theoretical and Applied Climatology, 132(1–2),* 15–29. DOI 10.1007/s00704-017-2076-y.
3. Liu, R., Cheng, L., Hu, C. (2019). Response of root-canopy index of winter wheat on persist drought. *Chinese Journal of Agrometeorology, 40,* 702–711.
4. Rampino, P., Pataleo, S., Gerardi, C., Mita, G., Perrotta, C. (2006). Drought stress response in wheat: physiological and molecular analysis of resistant and sensitive genotypes. *Plant, Cell & Environment, 29(12),* 2143–2152. DOI 10.1111/j.1365-3040.2006.01588.x.
5. Le, Z., Liao, R., Liu, J., Lu, J., Bai, Y. et al. (2014). Effects of water stress on stem, plant and yield of winter wheat over North China plain. *Journal of Meteorology & Environment, 30,* 120–124.
6. Kang, S., Zhang, L., Liang, Y., Hu, X., Cai, H. et al. (2002). Effects of limited irrigation on yield and water use efficiency of winter wheat in the Loess Plateau of China. *Agricultural Water Management, 55(3),* 203–216. DOI 10.1016/S0378-3774(01)00180-9.
7. Mupangwa, W., Twomlow, S., Walker, S. (2008). The influence of conservation tillage methods on soil water regimes in semi-arid southern Zimbabwe. *Physics and Chemistry of the Earth, Parts A/B/C, 33(8–13),* 762–767. DOI 10.1016/j.pce.2008.06.049.
8. Turner, N. C. (2004). Sustainable production of crops and pastures under drought in a Mediterranean environment. *Annals of Applied Biology, 144(2),* 139–147. DOI 10.1111/j.1744-7348.2004.tb00327.x.
9. Dai, A. (2011). Drought under global warming: a review. *Wiley Interdisciplinary Reviews: Climate Change, 2(1),* 45–65. DOI 10.1002/wcc.81.
10. Yan, T., Zhu, G., Ge, F., Jiang, Y., Li, W. et al. (2019). Effects of staged drought stress on photosynthetic pyhsiological characteristics and yield structure of wheat. *Meterology and Disaster Reduction Research, 42,* 46–53.
11. Yang, W., Wang, S., Li, Y., Sun, H., Yin, X. et al. (2018). Effects of supplementary irrigation on root system and water consumption of winter wheat in Semiarid Region. *Arid Zone Research, 35,* 920–928.
12. Yuan, J., Ma, C., Feng, Y., Zhang, J., Yang, F. et al. (2018). Response of chlorophyll fluorescence transient in leaves of wheats with different drought resistances to drought stresses and rehydration. *Plant Physiology Journal, 54,* 1119–1129.
13. Pei, D., Sun, Z., Chen, S., Zhang, X., Chen, S. et al. (2006). Effects of water stress on physiological and agronomic features of winter wheat. *Transactions of the Chinese Society of Agricultural Engineering, 20(08),* 68–72.
14. Flexas, J., Bota, J., Galmes, J., Medrano, H., Ribas-Carbó, M. (2006). Keeping a positive carbon balance under adverse conditions: responses of photosynthesis and respiration to water stress. *Physiologia Plantarum, 127(3),* 343–352. DOI 10.1111/j.1399-3054.2006.00621.x.
15. Hu, M., Zhang, Z., Xu, P., Dong, B., Li, W. et al. (2007). Relationship of water use efficiency with photoassimilate accumulation and transport in wheat under deficit irrigation. *Acta Agronomica Sinica, 33(11),* 1884–1891.
16. Shen, X., Sun, J., Liu, Z., Zhang, J., Liu, X. (2010). Effects of low irrigation limits on yield and grain quality of winter wheat. *Transactions of the Chinese Society of Agricultural Engineering, 26,* 58–65.
17. Cakir, R. (2004). Effect of water stress at different development stages on vegetative and reproductive growth of corn. *Field Crops Research, 89(1),* 1–16. DOI 10.1016/j.fcr.2004.01.005.
18. Wang, C., Guo, T., Peng, Y., Zhu, Y., Ma, D. et al. (2004). Effects of post-anthesis irrigation on grain quality indices and yield in winter wheat (*Triticum aestivum* L.). *Acta Agronomica Sinica, 30(10),* 1031–1035.
19. Innes, P., Blackwell, R. (1981). The effect of drought on the water use and yield of two spring wheat genotypes. *Journal of Agricultural Science, 96(3),* 603–610. DOI 10.1017/S0021859600034584.
20. Budak, H., Kantar, M., Yucebilgili Kurtoglu, K. (2013). Drought tolerance in modern and wild wheat. *Scientific World Journal, 2013(10),* 1–16. DOI 10.1155/2013/548246.
21. Ali, M. (2009). Irrigation-yield response factor of winter wheat for different growth phases. *Journal of Agrometeorology, 11,* 9–14.
22. Wang, K., Fu, Q., Ji, F., Xu, S. (2007). Study on comparison of different methods to calculating sensitivity index of Jensen model. *Journal of Northeast Agricultural University (English Edition), 14,* 278–282.

23. Jing, B. W., Khan, C., Yuan, S., Yc-Yang, W., Feng, M. (2020). Yield response of winter wheat (*Triticum aestivum* L.) to water stress in Northern China: a meta-analysis. *Applied Ecology and Environmental Research, 18,* 433–446.
24. Fischer, R. (2007). Understanding the physiological basis of yield potential in wheat. *Journal of Agricultural Science, 145(02),* 99. DOI 10.1017/S0021859607006843.
25. Tan, N., Lin, Q., Jiang, W., Liu, Y., Li, L. (2011). Effect of limited irrigation on diurnal variation in flag-leaf photosynthesis and yield of dryland wheat. *Chinese Journal of Eco-Agriculture, 19(4),* 805–811. DOI 10.3724/SP.J.1011.2011.00805.
26. Mou, H., He, J., Xing, J., Yao, S., Tang, Y. et al. (2016). Water changes in wheat spike during grain filling stage investigated by nuclear magnetic resonance. *Transactions of the Chinese Society of Agricultural Engineering, 32,* 98–104.
27. Chao, M., Shi, X., Zhang, J., Zhang, Y., Wang, Z. et al. (2020). Effects of persistant drought at grain filling stage on flag leaf photosynthesis, antioxidant enzyme activity, grain yield and quality of wheat. *Journal of Triticeae Crops, 40(04),* 1–9.
28. Lan, T., Jiang, D., Xie, Z., Dai, T., Jing, Q. et al. (2004). Effects of post-anthesis drought and waterlogging on grain quality traits in different specialty wheat varieties. *Journal of Soil and Water Conservation, 18(01),* 193–196.
29. Yang, L., Jia, Z., Han, Q., Yang, W., Yang, B. et al. (2015). Effects of water-fertilizer coupling on biomass and photosynthesis characteristics of flag leaf of winter wheat. *Journal of Northwest A & F University (Natural Science Edition), 43,* 83–98.
30. Yuan, Y., Deng, X. (2004). Effect of deficit and rewatering on wheat photosynthesis and yield. *Acta Botanica Boreali-Occidentalia Sinica, 24(07),* 1250–1254.
31. Huang, L., Gao, Y., Qiu, X., Li, X., Shen, X. et al. (2013). Effects of irrigation amount and stage on yield and water consumption of different winter wheat cultivars. *Transactions of the Chinese Society of Agricultural Engineering, 29,* 99–108.
32. Sheng, Y., Zhao, C., Jia, H. (2006). Effect of water stress on photosynthetic and biology character of winter wheat. *Journal of Soil and Water Conservation, 20(01),* 193–196.
33. Chu, P., Wang, D., Zhang, Y., Wang, X., Wang, X. et al. (2009). Effects of irrigation stage and amount on water consumption characteristics, grain yield and content of protein components of wheat. *Scientia Agricultura Sinica, 42,* 1306–1315.
34. Wang, S., Kang, S., Li, T. (2015). Suitable water deficit mode for winter wheat basing objective of water saving as well as high yield and quality. *Transactions of the Chinese Society of Agricultural Engineering, 31,* 111–118.
35. Zhang, S., Shan, L. (2002). Research progress on water use efficiency of plant. *Agricultural Research in the Arid Areas, 20(04),* 1–5.
36. Sun, H., Liu, C., Zhang, Y., Zhang, X. (2003). Effects of water stress in different growth stage on water consumption and yield in winter wheat. *Journal of Irrigation and Drainage, 22(02),* 13–16.