Available online at www.sciencedirect.com

ScienceDirect

The spike weight contribution of the photosynthetic area above the upper internode in a winter wheat under different nitrogen and mulching regimes

Wei Chen[a,b], *Jingjuan Zhang*[c,*], *Xiping Deng*[b,*]

[a] *Key Laboratory of Disaster Monitoring and Mechanism Simulating of Shaanxi Province, Baoji University of Arts and Sciences, Baoji 721013, Shaanxi, China*

[b] *State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Institute of Soil and Water Conservation, Northwest A&F University and Chinese Academy of Sciences, Yangling 712100, Shaanxi, China*

[c] *School of Veterinary and Life Sciences, Murdoch University, Murdoch, WA 6150, Australia*

ARTICLE INFO

Article history:
Received 12 July 2018
Received in revised
form 8 October 2018
Accepted 14 November 2018
Available online 2 December 2018

Keywords:
Grain-filling ratio
Grain-filling duration
Loess Plateau
Photosynthetic organs
Single spike weight
Winter wheat

ABSTRACT

Besides leaves, non-foliar green organs such as stem and spike are also considered photosynthetic organs. To assess the photosynthetic contributions of these organs, the correlations between these photosynthetic areas and single-spike weight were investigated in a winter wheat (*Triticum aestivum* L.) under four nitrogen and mulching treatments: N120, N150, N195, and N195 + M. Two-year repeated field experiments were conducted on the Loess Plateau of China. Non-foliar photosynthetic area, grain-filling ratio and duration, grain yield, and in particular, single-spike weight, were measured, recorded and analyzed. Under the N195 + M treatment, plants showed the largest area of photosynthetic organs (flag leaf and non-foliar organs) and the highest grain yield and single spike weight. Single-spike weight was positively correlated with the areas of all examined non-foliar photosynthetic organs, in particular with the area above the flag leaf node ($R^2 = 0.761^*$) and the area above the exposed part of the peduncle (EXP) ($R^2 = 0.800^{**}$). In addition, single-spike weight was highly correlated with average grain-filling ratio ($R^2 = 0.993^{**}$), whereas it was less highly correlated with grain-filling duration ($R^2 = 0.533$). The morphological traits of non-foliar photosynthetic organs were also more highly correlated with average grain-filling ratio than with average grain-filling duration. The significant correlation between each of the morphological traits (area, length and width) of EXP and single-spike weight indicates that morphological traits of EXP are important in determining spike weight in the Loess Plateau environment.

Abbreviations: DI, drought index; ENP, enclosed (by flag leaf sheath) part of peduncle; ERI, lower internode; ERL, lower internode leaf; ESTI, lowest internode; EXP, exposed part of peduncle; FL, flag leaf; FLS, flag leaf sheath; HI, harvest index; N, nitrogen; PI, penultimate internode; PL, penultimate leaf; WSC, water-soluble carbohydrates

[*] *Corresponding authors.*
E-mail addresses: j.zhang@murdoch.edu.au, (J. Zhang), dengxp@ms.iswc.ac.cn. (X. Deng).

Peer review under responsibility of Crop Science Society of China and Institute of Crop Science, CAAS.

https://doi.org/10.1016/j.cj.2018.10.004

1. Introduction

Wheat yield can be increased by large biomass and improved HI. However, the HI of wheat is now approaching a plateau and further increases in yield will require an increase in productive biomass [1]. Large biomass accumulation leads to high photosynthetic area, in turn increasing grain yield potential [2].

Leaf tissue has been historically considered to be the main photosynthetic organ [3]. Flag leaves in wheat are the leaves most important in photosynthesis, given that lower leaves are shaded by upper ones [4]. Leaf area is one of the most important morphological traits determining winter wheat yield under a wide range of environmental conditions [5,6]. Increasing leaf area can increase yield by increasing the production of photosynthetic assimilates [7]. The length, width, and area of a leaf, which are the three traits determining its shape and size, are [8].

Besides leaves, non-foliar green organs such as stem and spike can also be considered photosynthetic organs contributing to yield [9]. In several studies [10–13], photosynthesis by non-foliar organs contributed significant proportions of grain yield. Depending on wheat variety, environment, and experimental procedure, the ear contributed 10%–59% of total grain weight [14,15], while the exposed peduncle and flag leaf sheath contributed about 9%–12% [16]. Under drought conditions, non-foliar organs may become the main photosynthetic contributors to grain filling [11,17]. The peduncle accounts for much of the total stem length in wheat and is an important organ for carbohydrate storage [18], which plays an important role in stabilizing grain yield under drought conditions [19]. Ear photosynthesis has a direct effect on grain yield [20,21] and ear morphological traits are selection criteria in wheat breeding [22].

The concept of the photosynthetic area above the flag leaf node in wheat as an important determinant of grain yield has existed for many years [23,24]; however, the role of non-foliar photosynthesis has been omitted from analyses of the areas of specific photosynthetic modules with respect to their influence on grain yield. The correlations between single-spike weight and morphological traits of photosynthetic organs, particularly non-foliar organs have considered [10]. Measuring the dynamics of water-soluble carbohydrates (WSC) during grain filling is a common method for predicting yield [25,26]; however, it is time-consuming and destructive [27]. Crop morphological traits can be quickly, easily, cheaply, and nondestructively observed or measured in the field to give quantitative trait data related to yield [3]. In our study, the associations among single-spike weight, grain-filling parameters including grain-filling duration, average grain-filling ratio, and morphological traits of photosynthetic organs (length, width or diameter, and area) were investigated under different nitrogen and mulching regimes. The purpose of this study was to identify a possible link between photosynthetic morphological traits and single-spike weight. The linked phenotype could be used for yield improvement by increasing single-spike weight in winter wheat in the Loess Plateau environment.

2. Materials and methods

2.1. Site description and crop husbandry

The study was conducted during the 2011/2012 and 2012/2013 winter wheat growing seasons at Changwu Agricultural Research Station of the Chinese Academy of Sciences (35°14′ N, 107°41′ E and 1206 m elevation) located on the Loess Plateau of China. Because the water table is 60 m or deeper below the surface, groundwater is unavailable for crops. The soil was a silty loam according to the USDA classification system. The chemical properties of the soil at 20 cm depth on the experimental site before 2011 were as follows: pH 8.5, bulk density 1.2 g cm^{-3}, organic matter 14.4 g kg^{-1}, total N 0.95 g kg^{-1}, available phosphorus 19 mg kg^{-1}, and available potassium 157 mg kg^{-1}.

Seeds of a winter wheat cultivar (Changhan 58) were manually sown on Sep. 26, 2011 and harvested on July 2, 2012 in the first winter wheat growing season, and sown on Sep. 19, 2012, and harvested on June 26, 2013 in the second winter wheat growing season. In both seasons, the sowing rate was 150 kg ha^{-1}. The germination rate was about 90%. The plot size was 10 m × 10 m and the row space was 20 cm. Fungicides and pesticides were applied at jointing and booting stages and 10 days after anthesis. No diseases or pests were found during the growing period. Rainfall was not supplemented with irrigation in either season.

The patterns of rainfall and temperature in the station during the two growing seasons are summarized in Fig. 1. In the first growing season (2011/2012), the total precipitation was 667 mm including 477 mm and 190 mm in fallow and cropping periods, respectively. In the second growing season (2012/2013), the total precipitation was 422 mm, of which 234 mm was in the fallow and 188 mm in the cropping period (Fig. 1). The rainfall on the crop accounted for 29% and 45% of the total annual precipitation in the respective seasons. The DI for annual precipitation was calculated using Eq. (1) to assess variation in and status of precipitation among different years [28].

$$DI = (P–M)/\sigma \quad (1)$$

where P is annual precipitation, M is average precipitation, and σ is the standard error of precipitation. DI is used to distinguish among wet ($DI > 0.35$), normal ($-0.35 \leq DI \leq 0.35$), and dry ($DI < -0.35$) growing seasons for annual precipitation [29]. In the present study, the DIs were 0.43 and − 0.45 in the first and second winter wheat growing seasons, respectively. Thus, the first season was classified as wet and the second as dry.

2.2. Treatments and experimental design

Four N treatments were applied. P_2O_5 at a level of 120 kg ha^{-1} was applied in all treatments as basal fertilizer. The N treatments were.

1) N120: N (120 kg ha^{-1}) as basal fertilizer, with no mulch applied.
2) N150: N (150 kg ha^{-1}) as basal fertilizer, with plastic film mulching applied during the summer fallow but removed at sowing.

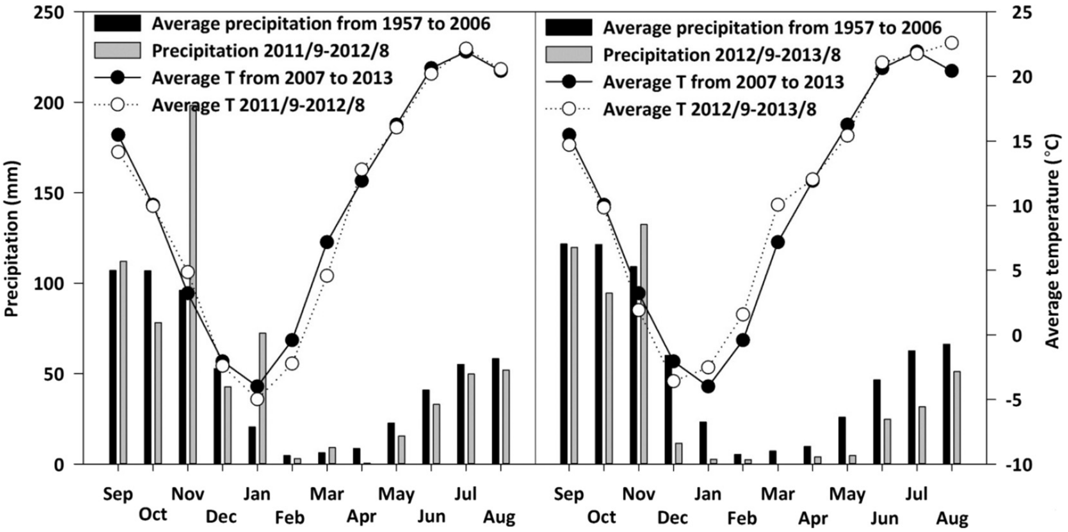

Fig. 1 – Precipitation and mean temperature in 2011/2012 (left) and 2012/2013 (right) along with the long-term means at the experimental site (the average precipitation was from 1957 to 2006 and the average temperature (T) was from 2007 to 2013).

3) N195: N (120 kg ha^{-1}) as basal fertilizer with additional N (75 kg ha^{-1}) top dressed at jointing and no mulch applied.
4) N195 + M: N (120 kg ha^{-1}), ox manure at 4.5 t ha^{-1} as basal fertilizer with additional N (75 kg ha^{-1}) top dressed at jointing, and wheat straw mulch applied during the summer fallow but removed at sowing.

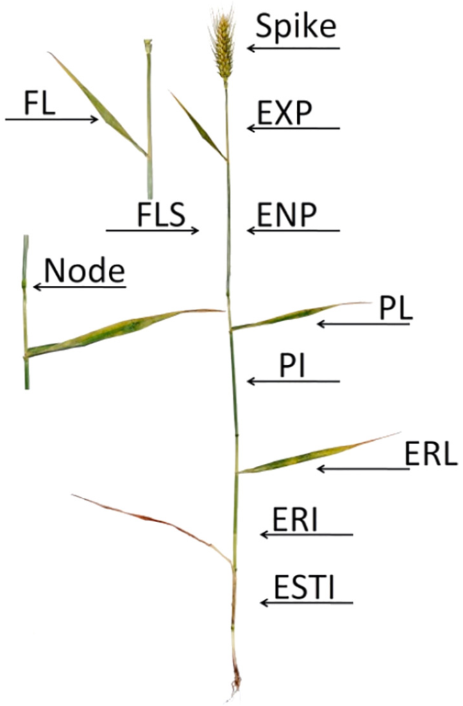

Fig. 2 – Diagram of segments of the main stem. EXP, exposed part of peduncle; ENP, enclosed part (by flag leaf sheath) of peduncle; PI, penultimate internode; ERI, lower internode; ESTI, lowest internode; FLS, flag leaf sheath; PL, penultimate leaf; ERL, lower internode leaf.

Nitrogen application at 120 kg ha^{-1} as basal fertilizer (treatment 1) is a common local practice in winter wheat cultivation, so that N120 represents the current farmers' practice in this region. N150 is derived from integrated soil–crop system management (ISSM), combined with plastic film mulching for maintaining soil water content (treatment 2). Topdressing with N at the jointing stage is used in the region for increasing yield, so that N195 represents the practice of compensating for the N shortage during late grain filling (treatment 3). The application of ox manure mulch during summer fallow periods was intended to increase soil water content and soil organic matter. Thus, N195 + M represents high-yielding practice (treatment 4).

There were four replications for each treatment, resulting in 16 plots. Plots were randomized. Nitrogen and P were in the form of urea and superphosphate. The ox manure contained 362.1 g kg^{-1} of C, 20.3 g kg^{-1} of total N, 8.5 g kg^{-1} of total K, and 18.2 g kg^{-1} of total P.

2.3. Sampling

Twenty uniform plants in each plot were sampled at anthesis. The stems were divided into five segments as shown in Fig. 2.

Table 1 – The main parameters of grain filling in two growth seasons.

Treatment	A		B		W (g)	
	2012	2013	2012	2013	2012	2013
N120	43.69	43.18	0.183	0.199	1.536	1.283
N150	23.71	38.89	0.167	0.183	1.693	1.630
N195	23.12	31.61	0.163	0.171	1.656	1.591
N195 + M	21.77	29.79	0.167	0.184	1.905	1.706

A and B, coefficients determined by regression; W, final spike weight.

The morphological traits of photosynthetic organs were measured as follows:

Flag leaf area was calculated using Eq. (2) as follows [30]:

$$\text{Flag leaf area} = \text{leaf length} \times \text{leaf width} \times 0.83 \tag{2}$$

Whole ear area was determined by Eq. (3) following previous studies [31,32].

$$\begin{aligned}\text{Whole ear area} = {} & \text{ear length} \times \text{ear width} \\ & \times 3.8\ (\text{glume surface area}) \\ & + \text{total awn length of the top third spikelet} \\ & \times \text{seed spikelet number} \\ & \times 0.1\ (\text{awn surface area})\end{aligned} \tag{3}$$

Surface area of specific internode segment as a cylinder was calculated by Eq. (4) according to a previous study [33].

$$\begin{aligned}\text{Internode area} = {} & \text{internode length} \times \pi \\ & \times \text{internode diameter}\end{aligned} \tag{4}$$

The diameter was measured with an electronic caliper. Because the diameter of the same internode segment varies from bottom to top, the diameter value used was the average of the top, middle, and bottom sections in one internode segment.

2.4. Determination of grain filling

Four hundred spikes that headed on the same day were chosen and tagged in each plot. Twenty tagged spikes from each plot were sampled at intervals of five days from anthesis to maturity. All kernels from each spike were removed by hand, dried at 70 °C to constant weight, and weighed. The grain-filling process was fitted into the growth equation [34] as described previously [35]:

$$W = W_0/\left(1 + Ae^{-Bt}\right) \tag{5}$$

where W_0 is kernel weight (g), t is time after anthesis, and A and B are coefficients determined by regression.

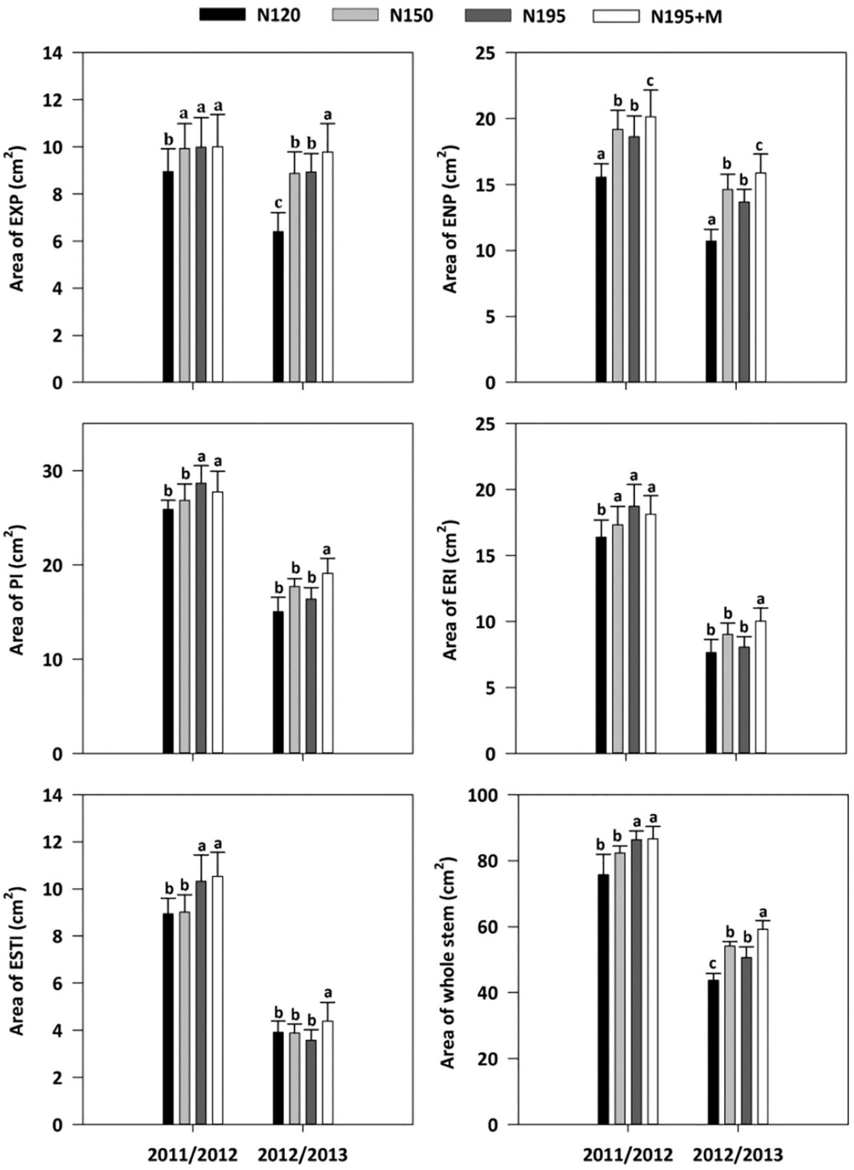

Fig. 3 – Effect of different treatments on areas of internode segments at anthesis in two growing seasons. Different letters above bars denote significant differences under different treatments ($P < 0.05$).

The software CURXPT was used to determine the duration of grain-filling and average grain-filling ratio. Briefly, the parameters A and B in Eq. (5) were determined by regression of the grain weight (W_0) on time after anthesis (t), and duration of grain filling (T) and average grain-filling ratio (V_a) were then calculated as follows.

$$t_1 = \left[\ln A - \ln\left(2 + 3^{1/2}\right)\right]/B \tag{6}$$

$$t_2 = \left[\ln A + \ln\left(2 + 3^{1/2}\right)\right]/B \tag{7}$$

$$t_3 = [\ln A + 4.59512]/B \tag{8}$$

$$T = T_1 + T_2 + T_3 \tag{9}$$

where T_1 is the duration of the lag stage, T_2 is the duration of the linear increase stage, and T_3 is the duration of physiological maturation.

The average grain-filling ratio (V_a) was calculated as.

$$V_a = W_0/T \tag{10}$$

where T is grain-filling duration (days) and W_0 is grain weight (g).

The grain-filling parameters are shown in Table 1.

2.5. Statistical analysis

The data were analyzed with SPSS software (SPSS 20.0; SPSS, Inc., Chicago, IL., USA) using one-way analysis of variance (ANOVA). Duncan's multiple-range test was used to compare treatment means. Linear regression was used to explore the

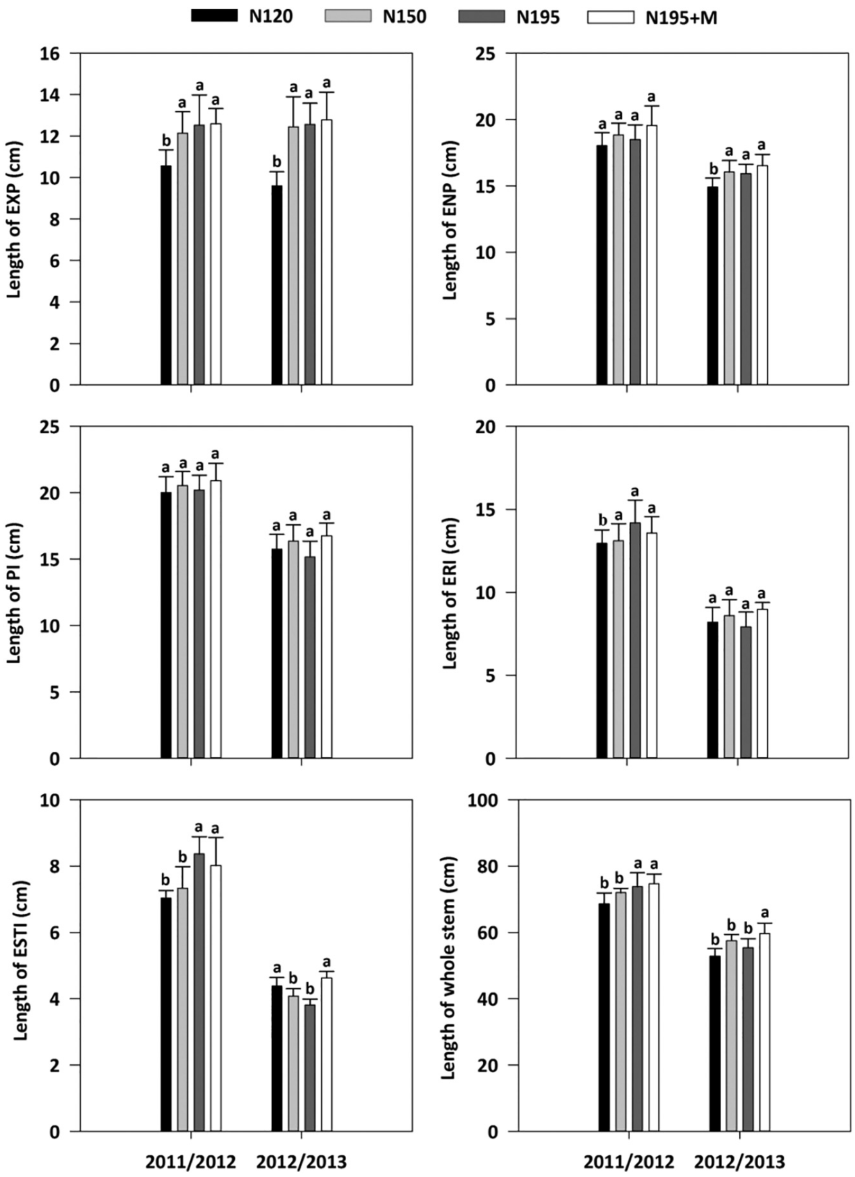

Fig. 4 – Effect of different treatments on lengths of different internode segments at anthesis in two growing seasons. Different letters above bars denote significant differences under different treatments (P < 0.05).

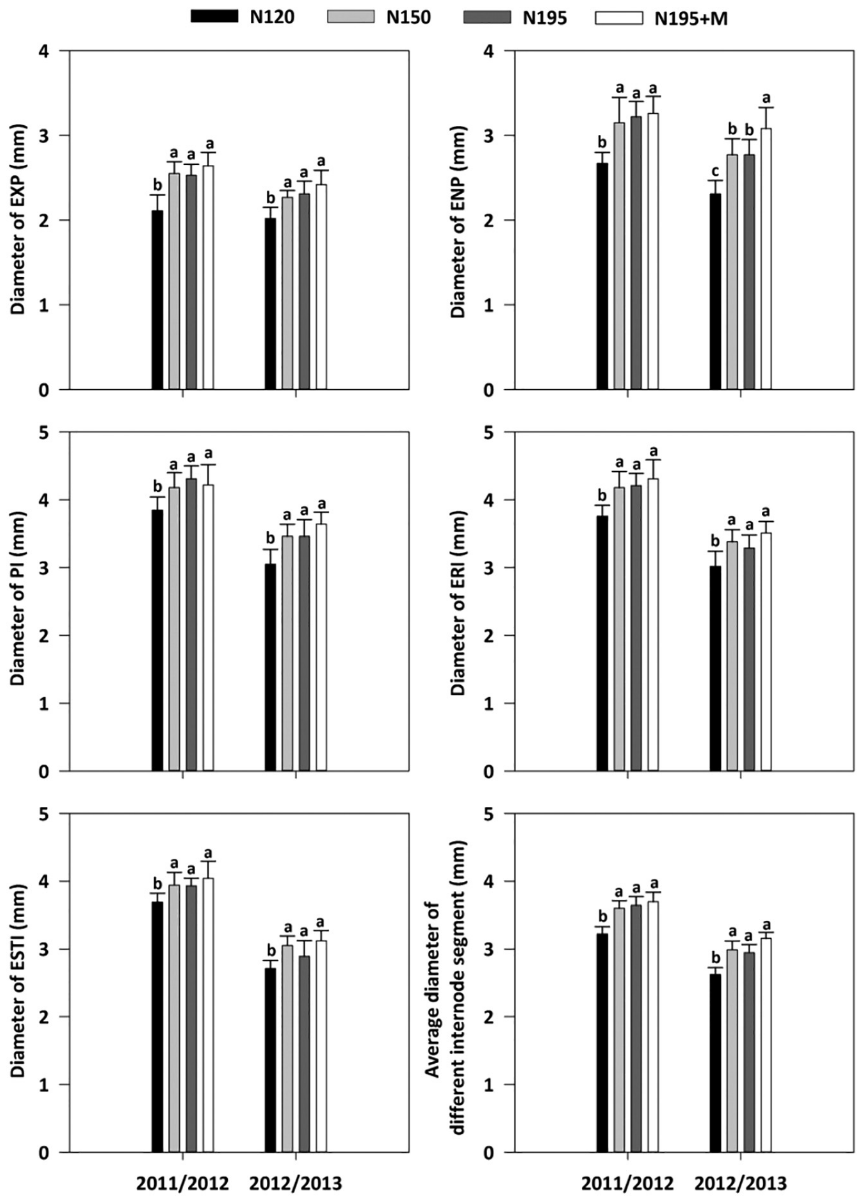

Fig. 5 – Effect of different treatments on diameters of internode segments at anthesis in two growth seasons. Different letters above bars denote significant differences under different treatments ($P < 0.05$).

correlations between single-spike weight and morphological traits of specific photosynthetic organs.

3. Results

3.1. Photosynthetic organs and grain yield under different treatments

Across treatments, the largest photosynthetic area was observed in N195 + M and the smallest in N120 in both seasons (Figs. 3–5). Among the photosynthetic organs, on average, the whole stem contributed the largest area (67.32 cm^2), followed by the spike (37.77 cm^2) and the flag leaf (15.53 cm^2) (Table 2).

Plants produced the largest stem area in treatment N195 + M, followed by N195 in 2011/2102 and N150 in 2012/2013. The smallest stem area was found in N120 (Fig. 3). Within different internode segments, the PI presented the longest length and the largest area, followed by ENP, ERI, EXP, and ESTI (Fig. 4). The trend in diameter differed from the trend in area and length. The descending order of diameters was PI, ERI, ESTI, ENP, and EXP (Fig. 5). On average, the area and length of the top three internode segments (EXP, ENP, and PI) were significantly larger than those of the two basal internodes (ERI and ESTI), whereas the diameter showed the opposite trend.

In N195 + M, plants also showed the largest spike area (Fig. 6). In the 2011/2012 growing season, the area of the spike in N195 + M (43.50 cm^2), N195 (42.47 cm^2) and N150 (40.54 cm^2) was significantly larger than that in N120 (35.66 cm^2), whereas there were no significant differences between N150, N195, and N195 + M. The same trend in spike length was found in the

Table 2 – Effect of rainfall levels on plant morphological traits.

	Spike					Flag leaf			Whole stem		
	Area (cm^2)	Length (cm)	Width (cm)	Spikelet number	Weight (g)	Area (cm^2)	Length (cm)	Width (cm)	Area (cm^2)	Length (cm)	Average diameter (mm)
2011/2012	40.54	8.39	1.15	19.31	1.60	20.03	16.06	1.50	82.73	72.24	3.54
2012/2013	34.99	7.81	1.02	17.59	1.47	11.03	11.03	1.15	51.91	56.36	2.93
Average	37.77	8.10	1.09	18.45	1.54	15.53	13.55	1.33	67.32	64.32	3.24

Values are means of four treatments.

second growing season, when the spike length was greatest (8.45 cm) in N195 + M and least (6.92 cm) in N120.

The effect of treatment N195 + M on flag leaf area was more pronounced. In the 2011/2012 growing season, the flag leaf area (25.60 cm^2) was significantly greater in N195 + M than in other treatments. Treatment N120 showed the smallest leaf area (13.54 cm^2) (Fig. 7). A similar trend was observed in the second growing season. No significant difference in flag leaf area was detected in N195 and N150. The treatment effects on flag leaf length and width showed trends similar to that on flag leaf area. The large photosynthetic area in the treatment N195 + M led to high single-spike weights. Single-spike weights were 1.77 and 1.64 g in 2011/2012 and 2012/2013, respectively, significantly greater than in other treatments (Fig. 8). In general, grain yield increased with N supply in both growing seasons. Under treatment N195 + M, plants produced the highest grain yield, 9217 and 7052 kg ha^{-1} in the first and second seasons, respectively, values 36% and 67% greater than that under N120. No significant difference in grain yield was observed for treatments N150 and N195 (Fig. 9).

3.2. Sizes of photosynthetic organs and grain yield in relation to rainfall

The levels of rainfall in the two growing seasons strongly affected the sizes of photosynthetic organs and grain yield. On average, the area of photosynthetic organs in the 2011/2012 growing season was much larger than that in the 2012/2013 growing season (Table 2), owing to the higher rainfall in the 2011/2012 growing season. In the first growing season, the average spike area was 40.5 cm^2, 15.9% greater than that in the second growing season. The average spike width, length,

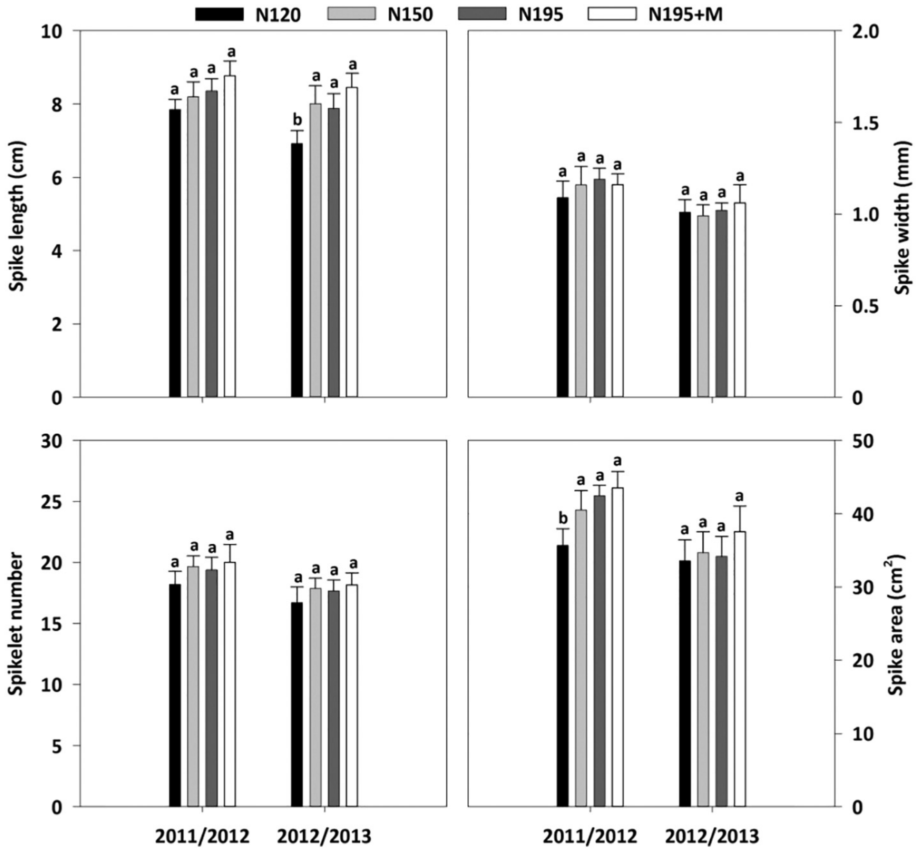

Fig. 6 – Effect of different treatments on spike morphological traits at anthesis in two growing seasons. Different letters above bars denote significant differences under different treatments ($P < 0.05$).

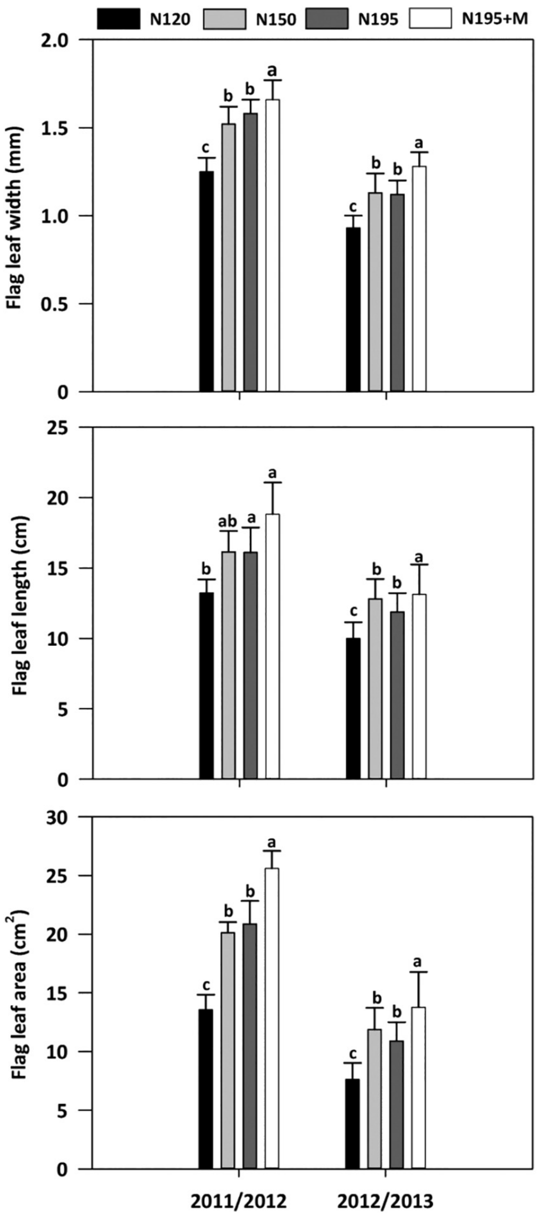

Fig. 7 – Effect of different treatments on morphological traits of flag leaf at anthesis in two growing seasons. Different letters above bars denote significant differences under different treatments ($P < 0.05$).

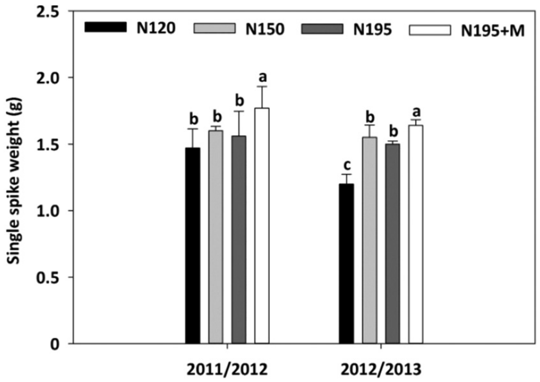

Fig. 8 – Effect of different treatments on single-spike weight at maturity in two growing seasons. Different letters above bars denote significant differences under different treatments ($P < 0.05$).

and spikelet number were respectively 1.1, 8.3, and 19.3 cm in 2011/2012 (the first season), values 11.3%, 5.8%, and 8.9% greater than those in the second growing season. The reduction in spike area under low rainfall was due mainly to reductions in spike width and spikelet number. This finding indicates that spike length has high heritability. The low level of rainfall during 2012/2013 season markedly reduced flag leaf and stem areas. On average, the flag leaf area (11 cm^2) was 82% lower than in the first season. Both flag leaf width and length were decreased by 35%. The length and area of the whole stem were also decreased by 22% and 56%, respectively, in the second growing season (Table 2). The average diameter in the second season (2.9 mm) was reduced by 20% relative to the first season. For all shoot organs (spike, flag leaf, and whole stem), the largest reduction in photosynthetic area was found in the flag leaf, followed by the whole stem and spike, indicating that the flag leaf was the organ most sensitive tow drought stress. Owing to the high rainfall in the first season, the average grain yield was 7946 kg ha^{-1}, 49% greater than that in the second season (5311 kg ha^{-1}) (Fig. 9).

3.3. *Correlations between single-spike weight and morphological traits of photosynthetic organs*

Single-spike weight showed significant positive correlations with the area, length, and width of the five internode segments and with specific photosynthetic modules (above flag leaf node, above EXP node, and above PI node) (Table 3). In particular, the area above the EXP node showed the highest correlations with single-spike weight. Within different internode segments, from apical to basal segments, the correlation coefficients decreased gradually. This finding suggests that the area of apical internode segments has more influence on single-spike weight than that of basal internode segments.

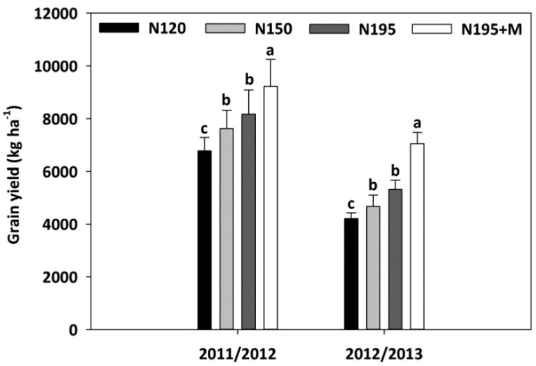

Fig. 9 – Effect of different treatments on grain yield at maturity in two growing seasons. Different letters above bars denote significant differences under different treatments ($P < 0.05$).

Table 3 – Correlations between morphological traits of photosynthetic organs/modules and single spike weight.

Morphological traits	Photosynthetic organ/module										
	Ear	FL	Above flag leaf node	EXP	Above EXP node	ENP	PI	Above PI node	ERI	ESTI	Whole stem
Area	0.556	0.788*	0.761*	0.922**	0.800**	0.853**	0.577	0.762*	0.553	0.486	0.645
Length	0.983**	0.828*		0.840**		0.715*	0.529		0.476	0.455	0.632
Width or diameter	0.542	0.799*		0.871**		0.904**	0.729*		0.721*	0.641	0.782*
Spikelet number	0.815*										

EXP, exposed part of peduncle; ENP, enclosed (by flag leaf sheath) part of peduncle; PI, penultimate internode; ERI, lower internode; ESTI, lowest internode; FL, flag leaf.

Above flag leaf node includes flag leaf and ear; Above EXP node includes flag leaf, ear and EXP; Above PI node includes ear, flag leaf, EXP, ENP, and PI.

Diameter of specific internode segment is the average diameter in the top, middle and bottom positions of a specific internode segment.

* and **, significant at $P = 0.05$ and $P = 0.01$, respectively.

3.4. *Correlations of average grain-filling ratio and grain-filling duration with morphological traits*

Grain-filling ratio and grain-filling duration are two factors influencing yield. For single-spike weight, there was a stronger (significant) correlation with average grain-filling ratio (R^2 = 0.993**) than with grain-filling duration (R^2 = 0.533) (Fig. 10). The morphological traits also showed stronger positive correlations with average grain-filling ratio (Table 4) than with grain-filling duration (Table 5). In particular, the area, length and diameter of EXP showed highly positive correlations with average grain-filling ratio (R^2 = 0.753*, R^2 = 0.726*, and R^2 = 0.786*), whereas only the diameter of PI showed a substantial correlation with the grain-filling duration (Table 4). These results indicate that the morphological traits of EXP play an important role in determining average grain-filling ratio. The correlation coefficients of the average grain-filling ratio and grain-filling duration tended to decrease across the internode segments from apical to basal.

4. Discussion

In wheat genetic improvement, the increasing contribution of ear photosynthesis to grain filling [10] suggests that grain yield can be increased by selection for large spike area. In agreement with a previous study [36], spike length and spikelet number per spike showed significantly positive correlations with single-spike weight, indicating that spike length, as an important component of spike area with high inheritability, is a crucial morphological trait for increasing grain yield.

Grain filling is the final growth stage in cereals and is an important stage in determining economic yield. Average grain-filling ratio and grain-filling duration are two important components influencing grain filling [37]. In the present study, both average grain-filling ratio and grain-filling duration showed positive correlations with single-spike weight. The stronger correlation of single-spike weight with grain-filling ratio (R^2 = 0.993**) than with grain-filling duration (R^2 = 0.533) agrees with previous reports that the rate of grain filling was more important in determining final grain yield than the length of the grain-filling period [37,38]. On the Loess Plateau, drought always occurs during the winter wheat growing season, particularly in the grain-filling period [39]. Drought stress accelerates the senescence of winter wheat, shortening the duration of grain filling [40]. Varieties with high grain-filling ratio are more likely to achieve high yield.

In this study, the quantity of N used in the N195 + M treatment was equivalent to N286 (N195 as chemical fertilizers and N91 as ox manure). Under N195 + M, plants

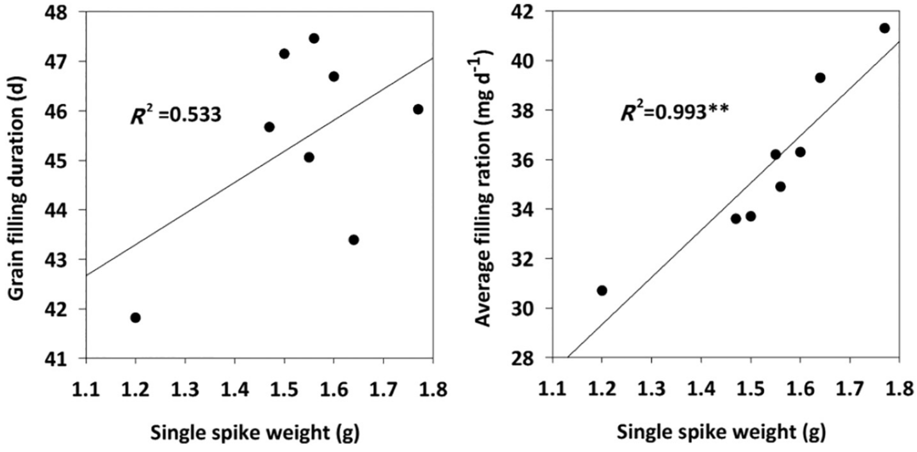

Fig. 10 – Relationship between single spike weight and grain-filling duration (left) and average grain-filling ratio (right). ** Significant at $P < 0.01$.

Table 4 – Relationship between morphological traits of different photosynthetic organs/modules and average grain-filling ratio.

Morphological traits	Photosynthetic organ/module										
	Ear	FL	Above flag leaf node	EXP	Above EXP node	ENP	PI	Above PI node	ERI	ESTI	Whole stem
Area	0.691	0.491	0.668	0.753*	0.815*	0.727*	0.416	0.790*	0.379	0.348	0.489
Length	0.899**	0.736**		0.726*		0.575	0.413		0.324	0.326	0.484
Width or diameter	0.406	0.679		0.786*		0.798*	0.549		0.576	0.490	0.634
Spikelet number	0.659										

EXP, exposed part of peduncle; ENP, enclosed (by flag leaf sheath) part of peduncle; PI, penultimate internode; ERI, lower internode ESTI, lowest internode; FL, flag leaf.

Above flag leaf node includes flag leaf and ear; Above EXP node includes flag leaf, ear, and EXP; Above PI node includes ear, flag leaf, EXP, ENP, and PI.

Diameter of specific internode segment is the average diameter in the top, middle, and bottom positions of a specific internode segment.

* and **, significant at $P = 0.05$ and $P = 0.01$, respectively.

produced the highest yield, followed by N195 and N150, whereas yield was lowest in N120. In agreement with previous studies on the Loess Plateau of China, grain yield increased with N input [41]. The size of photosynthetic organs is an important factor determining the extent of photosynthesis [42], which strongly influences HI [1]. The N195 + M treatment can help plants produce larger photosynthetic organs and increase single-spike weight. In the present study, the areas of all photosynthetic organs showed positive correlations with single-spike weight. A positive correlation between flag leaf area and yield was also reported by Simon [6]. A larger leaf area will improve light interception and thereby increase biomass and yield [43].

Although the morphological traits of all internode segments showed positive correlations with single-spike weight, the correlation coefficients for the different internode segments with single-spike weight varied. The two apical internode segments showed higher correlations with single-spike weight than did the other internodes. This finding suggests that the peduncle could be the primary source of assimilates for grain filling [4,44]. Among the other three internodes examined in this study, PI seemed to act as the most important source organ supporting grain filling [45], and the two basal internodes (ERI and ESTI), which form earlier than the top internode segments, may store enough structural carbohydrates to support the development of the plant.

The role of morphological traits above the flag leaf node plays a role not only in determining wheat grain yield [23] but also in drought resistance in durum wheat [46]. Of the three different photosynthetic features (above flag leaf node, above EXP node, and above PI node), a significant positive correlation between the area above flag leaf node and single-spike weight was found. The highly significant associations between single-spike weight and above EXP node ($R^2 = 0.800^{**}$) rather than above flag leaf node ($R^2 = 0.761^{*}$), indicates that the area above EXP node plays a more important role in contributing to the final grain yield. On the Loess Plateau, winter wheat is most likely to encounter drought stress during grain filling. Leaves tend to be damaged earlier under drought stress than EXP [47]. EXP probably has superior performance in photosynthesis during grain-filling under drought conditions.

Both diameter and length of the stem showed positive correlations with single-spike weight. The diameter and length of the stem not only determine the volume of internode segments, which can influence photosynthesis in non-foliar organs, but also are useful in accumulating and remobilizing WSC to the developing kernels [48]. The diameter

Table 5 – Relationship between the morphological traits of different photosynthetic organs/modules and grain-filling duration.

Morphological traits	Photosynthetic organ/module										
	Ear	FL	Above flag leaf node	EXP	Above EXP node	ENP	PI	Above PI node	ERI	ESTI	Whole stem
Area	0.386	0.597	0.565	0.704	0.597	0.643	0.613	0.597	0.594	0.553	0.633
Length	0.557	0.593		0.575		0.630	0.496		0.565	0.521	0.614
Width or diameter	0.570	0.628		0.586		0.605	0.716*		0.651	0.623	0.673
Spikelet number	0.683										

EXP, exposed part of peduncle; ENP, enclosed part (by flag leaf sheath) of peduncle; PI, penultimate internode; ERI, lower internode; ESTI, lowest internode; FL, flag leaf.

Above flag leaf node includes flag leaf and ear; Above EXP node includes flag leaf, ear, and EXP; Above PI node includes ear, flag leaf, EXP, ENP, and PI.

Diameter of specific internode segment is the average diameter in the top, middle, and bottom positions of a specific internode segment.

* Significant at $P = 0.05$.

of the internode, which was highly correlated with stem WSC concentration [18], also plays an important role in lodging resistance [49]. The longer the internode, the more potential there is for larger reserve accumulation [18,50], and the level of WSC stored in the stem has been proposed as a potential selection criterion for drought tolerance in wheat [51]. The area of photosynthetic organs and the length and diameter of internode segments are important factors determining the capacity for reserve accumulation and remobilization in wheat.

5. Conclusions

The relationships between single-spike weight, grain-filling characteristics, and morphological traits of photosynthetic organs were characterized. With greater N input, higher grain yield was realized in parallel with an increase in photosynthetic area. The significant correlations between single-spike weight and the area of non-foliar organs of the above EXP node indicate the important role of the photosynthetic area above the EXP node in determining final grain yield under Loess Plateau conditions. Moreover, the morphological traits of EXP (particularly the diameter) and single-spike weight showed significant positive correlations with the average grain-filling ratio, illustrating the important role of the diameter of EXP in single-spike weight by its effect on grain-filling ratio. EXP morphological traits, particularly diameter, could be a good indicator of grain yield for breeding purposes in terminal drought areas. Further evaluation in a range of environments with more cultivars may validate this correlation.

Acknowledgments

The authors acknowledge the financial support of the National Basic Research Program of China (2015CB150402), National Natural Science Foundation of China (51479189), Open Foundation of State Key Laboratory, Institute of Water and Soil Conservation, Chinese Academy of Sciences, Ministry of Water Resources (A314021402-1610), and Key Research Foundation of Baoji University of Arts and Sciences (ZK16066) and thank Prof. A. Egrinya Eneji from Agronomy & Honourable Commissioner of the Ministry of Agriculture and Natural Resources in Nigeria for critically reviewing the manuscript. The authors thank Mrs. Frances Brigg at Murdoch University, Perth, Australia for polishing the manuscript.

REFERENCES

[1] M. Parry, M. Reynolds, M. Salvucci, C. Raines, P. Andralojc, X. Zhu, G. Price, A. Condon, R. Furbank, Raising yield potential of wheat. II. Increasing photosynthetic capacity and efficiency, J. Exp. Bot. 62 (2011) 453–467.

[2] X.G. Zhu, S.P. Long, D.R. Ort, Improving photosynthetic efficiency for greater yield, Annu. Rev. Plant Biol. 61 (2010) 235–261.

[3] J. Pietragalla, Crop morphological traits, in: A. Pask, J. Pietragalla, D. Mullan, M.P. Reynolds (Eds.),Physiological Breeding II: A Field Guide to Wheat Phenotyping, International Maize and Wheat Improvement Center, Mexico, D.F 2012, pp. 106–112.

[4] K.G. Briggs, A. Aytenfisu, Relationships between morphological characters above the flag leaf node and grain yield in spring wheats, Crop Sci. 20 (1980) 350–354.

[5] S.H. Mohiuddin, L.I. Croy, Flag leaf and peduncle area duration in relation to winter wheat grain yield, Agron. J. 72 (1980) 299–301.

[6] M.R. Simon, Inheritance of flag-leaf angle, flag-leaf area and flag-leaf area duration in four wheat crosses, Theor. Appl. Genet. 98 (1999) 310–314.

[7] P.F. Li, Z.G. Cheng, B.L. Ma, J.A. Palta, H.Y. Kong, F. Mo, J.Y. Wang, Y. Zhu, G.C. Lv, A. Batool, Dryland wheat domestication changed the development of aboveground architecture for a well-structured canopy, PLoS One 9 (2014), e95825. .

[8] B. Shen, W.D. Yu, Y.J. Zhu, Y.Y. Fan, J.Y. Zhuang, Fine mapping of a major quantitative trait locus, qFLL6.2, controlling flag leaf length and yield traits in rice (*Oryza sativa* L.), Euphytica 184 (2012) 57–64.

[9] G. Aschan, H. Pfanz, Non-foliar photosynthesis: a strategy of additional carbon acquisition, Flora 198 (2003) 81–97.

[10] M.L. Maydup, M. Antonietta, J.J. Guiamet, C. Graciano, J.R. López, E.A. Tambussi, The contribution of ear photosynthesis to grain filling in bread wheat (*Triticum aestivum* L.), Field Crop Res. 119 (2010) 48–58.

[11] L. Kong, F. Wang, B. Feng, S. Li, J. Si, B. Zhang, The structural and photosynthetic characteristics of the exposed peduncle of wheat (*Triticum aestivum* L.): an important photosynthate source for grain-filling, BMC Plant Biol. 10 (2010) 141–151.

[12] L.A. Kong, Y. Xie, M.Z. Sun, J.S. Si, L. Hu, Comparison of the photosynthetic characteristics in the pericarp and flag leaves during wheat (*Triticum aestivum* L.) caryopsis development, Photosynthetica 54 (2016) 40–46.

[13] Y.P. Li, H.B. Li, Y.Y. Li, S.Q. Zhang, Improving water-use efficiency by decreasing stomatal conductance and transpiration rate to maintain higher ear photosynthetic rate in drought-resistant wheat, Crop J. 5 (2017) 231–239.

[14] J.L. Araus, H.R. Brown, A. Febrero, J. Bort, M.D. Serret, Ear photosynthesis, carbon isotope discrimination and the contribution of respiratory CO_2 to differences in grain mass in durum wheat, Plant Cell Environ. 16 (1993) 383–392.

[15] P. Kriedemann, The photosynthetic activity of the wheat ear, Ann. Bot. 30 (1966) 349–363.

[16] Z.M. Wang, A.L. Wei, D.M. Zheng, Photosynthetic characteristics of non-leaf organs of winter wheat cultivars differing in ear type and their relationship with grain mass per ear, Photosynthetica 39 (2001) 239–244.

[17] E.A. Tambussi, J. Bort, J.J. Guiamet, S. Nogues, J.L. Araus, The photosynthetic role of ears in C3 cereals: metabolism, water use efficiency and contribution to grain yield, Crit. Rev. Plant Sci. 26 (2007) 1–16.

[18] C.S. Pierre, R. Trethowan, M. Reynolds, Stem solidness and its relationship to water-soluble carbohydrates: association with wheat yield under water deficit, Funct. Plant Biol. 37 (2010) 166–174.

[19] A. Sallam, M. Hashad, E. Hamed, M. Omara, Genetic variation of stem characters in wheat and their relation to kernel weight under drought and heat stresses, J. Crop. Sci. Biotechnol. 18 (2015) 137–146.

[20] F. Breseghello, M. Sorrells, QTL analysis of kernel size and shape in two hexaploid wheat mapping populations, Field Crop Res. 101 (2007) 172–179.

[21] Y. Xiao, S. He, J. Yan, Y. Zhang, Y. Zhang, Y. Wu, X. Xia, J. Tian, W. Ji, Z. He, Molecular mapping of quantitative trait loci for kernel morphology traits in a non-1BL.1RS × 1BL.1RS wheat cross, Crop Pasture Sci. 62 (2011) 625–638.

[22] Y.Q. Wang, W.X. Xi, Z.M. Wang, B. Wang, X.X. Xu, M.K. Han, S. L. Zhou, Y.H. Zhang, Contribution of ear photosynthesis to grain yield under rainfed and irrigation conditions for winter wheat cultivars released in the past 30 years in North China plain, J. Integr. Agric. 15 (2016) 2247–2256.

[23] G. Simpson, Association between grain yield per plant and photosynthetic area above flag leaf node in wheat, Can. J. Res. 48 (1968) 253–260.

[24] P. Hsu, P.D. Walton, Relationships between yield and its components and structures above the flag leaf node in spring wheat, Crop Sci. 11 (1971) 190–193.

[25] B. Ehdaie, G. Alloush, M. Madore, J. Waines, Genotypic variation for stem reserves and mobilization in wheat: II. Postanthesis changes in internode water-soluble carbohydrates, Crop Sci. 46 (2006) (2903–2013).

[26] J.J. Zhang, W. Chen, B. Dell, R. Vergauwen, X.M. Zhang, J.E. Mayer, W. van den Ende, Wheat genotypic variation in dynamic fluxes of WSC components in different stem segments under drought during grain filling, Front. Plant Sci. 6 (2015) 624.

[27] G.P. Xue, C.L. McIntyre, A.R. Rattey, A.F. van Herwaarden, R. Shorter, Use of dry matter content as a rapid and low-cost estimate for ranking genotypic differences in water-soluble carbohydrate concentrations in the stem and leaf sheath of *Triticum aestivum*, Crop Pasture Sci. 60 (2009) 51–59.

[28] S. Guo, H. Zhu, T. Dang, J. Wu, W. Liu, M. Hao, Y. Li, J.K. Syers, Winter wheat grain yield associated with precipitation distribution under long-term nitrogen fertilization in the semiarid loess plateau in China, Geoderma 189–190 (2012) 442–450.

[29] N.Q. Xing, Y.Q. Zhang, L.X. Wang, The Study on Dryland Agriculture in North China, China Agriculture Press, Beijing, 2002 (in Chinese).

[30] H. Dong, J. Bi, G.L. Xia, X.B. Zhou, Y.H. Chen, Effects of irrigation and planting patterns on photosynthetic characteristics of flag leaf and yield at late growth stages of winter wheat, Chin. J. Appl. Ecol. 8 (2014) 2259–2266 (in Chinese with English abstract).

[31] I.D. Teare, C.J. Peterson, Surface area of chlorophyll-containing tissue on the inflorescence of *Triticum aestivum* L, Crop Sci. 11 (1971) 627–628.

[32] S. Qiu, L. Zhai, Assays on the surface area of ear and awn in winter wheat, Acta Agron. Sin. 11 (1985) 138 (in Chinese with English abstract).

[33] D.J. Miralles, D.F. Calderini, K.P. Pomar, A. D'Ambrogio, Dwarfing genes and cell dimensions in different organs of wheat, J. Exp. Bot. 49 (1998) 1119–1127.

[34] F.J. Richards, A flexible growth function for empirical use, J. Exp. Bot. 10 (1959) 290–300.

[35] Y. Zhang, M.D. Hao, Y.H. Pang, Study on the grain-filling characteristic of the succession of wheat cultivars in the gully areas of the loess plateau, J. Triticeae Crops 28 (2008) 1058–1062 (in Chinese with English abstract).

[36] M. Shahid, F. Mohammad, M. Tahir, Path coefficient analysis in wheat, Sarhad J. Agric. 18 (2002) 383–388.

[37] H.G. Nass, B. Reiser, Grain filling period and grain yield relationships in spring wheat, Can. J. Plant Sci. 55 (1975) 673–678.

[38] R.X. Wang, L. Hai, X.Y. Zhang, G.X. You, C.S. Yan, S.H. Xiao, QTL mapping for grain filling rate and yield-related traits in RILs of the Chinese winter wheat population Heshangmai × Yu8679, Theor. Appl. Genet. 118 (2009) 313–325.

[39] L. He, S. Asseng, G. Zhao, D. Wu, X. Yang, W. Zhang, N. Jin, Q. Yu, Impacts of recent climate warming, cultivar changes, and crop management on winter wheat phenology across the Loess Plateau of China, Agric. For. Meteorol. 200 (2015) 135–143.

[40] J.C. Yang, J.H. Zhang, Grain filling of cereals under soil drying, New Phytol. 169 (2006) 223–236.

[41] L.L. Wang, J.A. Palta, W. Chen, Y.L. Chen, X.P. Deng, Nitrogen fertilization improved water-use efficiency of winter wheat through increasing water use during vegetative rather than grain filling, Agric. Water Manag. 197 (2018) 41–53.

[42] P.J. Welbank, S.A. French, K. Witts, Dependence of yields of wheat varieties on their leaf area durations, Ann. Bot. 30 (1966) 291–299.

[43] D.S. Falster, M. Westoby, Plant height and evolutionary games, Trends Ecol. Evol. 18 (2003) 337–343.

[44] I. Khaliq, N. Parveen, M.A. Chowdhry, Correlation and path coefficient analyses in bread wheat, Int. J. Agric. Biol. 6 (2004) 633–635.

[45] W. Chen, X.P. Deng, A.E. Eneji, L.L. Wang, Y. Xu, Y.J. Cheng, Dry-matter partitioning across parts of the wheat internode during the grain filling period as influenced by fertilizer and tillage treatments, Commun. Soil Sci. Plant Anal. 13 (2014) 1779–1812.

[46] D. Villegas, L.F. Garcia Del Moral, Y. Rharrabti, V. Martos, C. Royo, Morphological traits above the flag leaf node as indicators of drought susceptibility index in durum wheat, J. Agron. Crop Sci. 193 (2007) 103–116.

[47] I.F. Wardlaw, Interaction between drought and chronic high temperature during kernel filling in wheat in a controlled environment, Ann. Bot. 90 (2002) 469–476.

[48] A. Blum, Improving wheat grain filling under stress by stem reserve mobilisation, Euphytica 100 (1998) 77–83.

[49] P.M. Berry, R. Sylvesterbradley, S. Berry, Ideotype design for lodging-resistant wheat, Euphytica 154 (2007) 165–179.

[50] D. Dodig, D. Rančić, B.V. Radović, M. Zorić, J. Savić, V. Kandić, I. Pećinar, S. Stanojević, A. Šešlija, D.V.S. Pekićquarrie, Response of wheat plants under post-anthesis stress induced by defoliation: II. Contribution of peduncle morpho-anatomical traits and carbon reserves to grain yield, J. Agric. Sci. 155 (2016) 475–493.

[51] M.J. Foulkes, R. Sylvester-Bradley, R. Weightman, J.W. Snape, Identifying physiological traits associated with improved drought resistance in winter wheat, Field Crop Res. 103 (2007) 11–24.