



# The Role of Deep Roots in Sorghum Yield Production under Drought Conditions

Xiaofei Chen<sup>1,†</sup>, Qi Wu<sup>1,†</sup>, Yue Gao<sup>1,2</sup>, Jiao Zhang<sup>1</sup>, Yitao Wang<sup>3</sup>, Ruidong Zhang<sup>1,4</sup>, Yufei Zhou<sup>1,\*</sup>, Muji Xiao<sup>1</sup>, Wenjuan Xu<sup>1</sup> and Ruidong Huang<sup>1</sup>

- <sup>1</sup> College of Agronomy, Shenyang Agricultural University, Shenyang 110866, China; chenxiaofei1992@163.com (X.C.); wuqi19921992@163.com (Q.W.); gylina9003@163.com (Y.G.); mrs\_zhang2011@126.com (J.Z.); sxnkyzrd@126.com (R.Z.); mujixiao@163.com (M.X.); w.xu-syau@126.com (W.X.); r\_huang@126.com (R.H.)
- <sup>2</sup> Crops Resources Research Institute, Jilin Academy of Agricultural Science, Gongzhuling 136100, China
- <sup>3</sup> Liaoning Agricultural Development Service Center, Shenyang 110032, China; wangyitao-happy@163.com
- <sup>4</sup> Institute of Economic Crop, Shanxi Academy of Agricultural Sciences, Fenyang 032200, China
- \* Correspondence: zhouyufei2002@aliyun.com or zhouyufei@syau.edu.cn
- + These authors contributed equally to this work.

Received: 23 March 2020; Accepted: 21 April 2020; Published: 24 April 2020



**Abstract:** Root function plays a vital role in maintaining crop production. However, the role of deep roots in yield production and their effects on photosynthetic performance in sorghum remain unclear. This study aimed to provide theoretical supports for establishing highly efficient root systems of sorghum to achieve more yield under certain conditions. In this study, two sorghum (Sorghum bicolor L. Moench) cultivars, Jiza127 and Jiza305, were cultivated in soil columns as experimental materials. Three treatments (no roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60) were carried out under drought conditions during the filling stage. The root bleeding intensity, endogenous substances in the root bleeding sap, photosynthetic characteristics, dry matter accumulation, and yield were measured. The results showed that R30 and R60 significantly reduced yield in both sorghum cultivars, and the effect of R30 on yield was greater than that of R60. The contributions of roots below 30 cm to the yield of both sorghum hybrids were notably higher than those below 60 cm. R30 significantly reduced the dry matter weights (DMWs) of leaves, stems, sheaths, and panicles. R60 significantly reduced the DMW of panicles but had no significant effect on the DMWs of leaves and stems. R30 significantly reduced the photosynthetic level and PSII reaction center activity; however, the effect of R60 was not significant. Although both R30 and R60 significantly reduced root activity and the soluble sugar, amino acid, gibberellin (GA<sub>3</sub>), and abscisic acid (ABA) contents of the root bleeding sap, some of the above indicators in R60 were significantly higher than those in R30 during the filling stage, indicating that the deeper roots (below 30 cm) had a critical regulatory effect on the physiological processes of the aerial parts in sorghum, which resulted in a stronger effect on yield, especially under drought conditions. In brief, the deep roots of sorghum played a key role in yield production, but the roots in different soil depths regulated yield production in different ways. Our results indicate that deep roots of sorghum deserve consideration as a potential trait for yield improvement especially under drought conditions.

Keywords: sorghum; deep roots; root bleeding sap; photosynthetic performance; yield

# 1. Introduction

Plant roots are important organs that absorb water and nutrients from the soil and are a center for the biosynthesis and transport of plant hormones such as abscisic acid (ABA) [1–4]. The morphological



and physiological characteristics of roots affect the growth of plant aerial parts [1]. The roots and aerial parts of crops are interactive and interdependent [5]. Therefore, maintaining high root vitality is essential [6], and changing root structure is more likely to promote crop growth than changing the stems and leaves [7,8]. Alteration of root traits to adapt to the surrounding environment and optimize resource utilization plays a vital role in the adaptation of wheat to different environments [9]. The study of Sebastian et al. [10] showed that stronger suppression of crown roots actually may benefit crop productivity under a water deficit. Hammer et al. [11] reported that root structure optimization and water utilization may be the basis for historic advances in yield breeding in the U.S. maize belt. Hence, to understand what kind of root system is beneficial to crop growth, it is necessary to explore the role of the root system on crop growth and yield production, especially the role of deep roots under adverse conditions such as drought stress.

Gewin revealed that deep roots play a vital role in mitigating water stress in many crops [12]. Deep roots have a greater ability to absorb water and nutrients than shallow roots and act as the central hub of the water and nutrient cycle [3,13,14]. In addition, deep roots play a key role in crop adaptation to circumstances. Manschadi et al. [15] reported that the roots of drought-tolerant wheat are tighter, more uniform, and longer than those of drought sensitive wheat, and the plants have higher water use efficiency. The deep roots of crops can not only increase yield by improving water and nitrogen uptake but also reduce environmental nitrogen leaching [16–18]. Therefore, breeders strongly emphasize the role of deep roots in absorbing water and nutrients when developing new varieties [19–22]. Chaves et al. [23] revealed that since deeper soil layers have higher moisture contents in arid environments, deep-rooted plants are more likely to survive under these conditions. Deep roots can absorb more substances, including water and nutrients, in arid environments and maintain the function of the shallow roots through material transport [24–26]. Although some functions of the root system are well known, some of the physiological functions of deep roots and their effects on aboveground crop production have rarely been reported.

Sorghum is mainly planted in arid and semiarid regions of Asia and Africa, and the planting area in these regions accounts for 85% of the worldwide planting area according to 2018 data [27]. However, filling stage is the most important period for yield production, at which drought can lead to severe decline of yield in sorghum [28]. Sorghum can feed at least 5 million people in these areas due to its unique drought adaptability [29–31], and sorghum roots play an important role in its drought tolerance [32]. Wang et al. reported that a higher root activity has been found in the stay green sorghum B35 as compared to non-green-stayed sorghum Sanchisan and is considered as one of the drought-resistant mechanisms under drought conditions [33]. Photosynthetic parameters and osmotic adjustment ability are the principal factors associated with sorghum yield under drought conditions [28]. However, research on root systems is time consuming, expensive, and difficult because plant roots are hidden under the ground. Therefore, compared with studies of the aboveground characteristics of plants, there are few studies on the physiological functions of underground roots. In recent decades, although the number of root system studies has significantly increased, these studies have mainly been limited to shallow root systems [34,35] or the effects of different root types on plant productivity [1]. Specifically, little is known about the effects of deep root systems on the aboveground physiology and yield production of sorghum plants [4]. Therefore, the objective of this study was to determine (1) the contribution of roots in different soil depths to yield production in sorghum and (2) the manner in which deep roots affect photosynthesis and yield. Overall, we hypothesize that the deep roots of sorghum play a crucial role in yield production, and that the roots in different soil depth regulate yield production differently.

#### 2. Materials and Methods

## 2.1. Plant Materials and Experimental Design

This experiment was conducted at the experimental base of Shenyang Agricultural University from 2015 to 2016. Average temperature and sunshine duration are similar in both experimental years (Figure 1). The sorghum hybrids Jiza127 (J127) and Jiza305 (J305) were selected as experimental materials which are mainly cultivated in northeastern China. Before the test, PVC tubes (tube length 1 m, inner diameter 30 cm) were cut into two sections of the same size longitudinally, fixed tightly with pipe hoops (Figure 2), and placed in a 1 m deep soil pit. The original soil layer was placed into the PVC tubes to ensure the spatial distribution of the original soil layer. After that, the soil was compacted by watering. The tubes were arranged in two rows with a large ridge (ridge spacing 66 cm, row spacing 33 cm), and the planting density was 6 plants m<sup>-2</sup>. Five uniformly sized seeds were sown in each soil column on May 8. One seedling was left at the five-leaf stage. Surface soil samples were taken at 5 cm soil depth. Soil was silt loam with a pH of 7.0, organic matter content of  $30.82 \text{ g kg}^{-1}$ , alkali hydrolysable N of 114.52 mg kg<sup>-1</sup>, available P of 78.33 mg kg<sup>-1</sup>, available K of 102.92 mg kg<sup>-1</sup>. At 45 cm soil depth, soil was loam with organic matter content of 14.82 g kg<sup>-1</sup>, alkali hydrolysable N of 59.21 mg kg<sup>-1</sup>, available P of 12.61 mg kg<sup>-1</sup>, available K of 51.46 mg kg<sup>-1</sup>. At 80 cm soil depth, there were organic matter content of 13.19 g kg<sup>-1</sup>, alkali hydrolysable N of 45.35 mg kg<sup>-1</sup>, available P of 12.53 mg kg<sup>-1</sup>, available K of 75.85 mg kg<sup>-1</sup> in the loam soil. Diammonium phosphate (2.46 g) was used as seed fertilizer, and urea (3.33 g) was applied to each column at the jointing stage based on local sorghum cultivation. Three treatments (no roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60) were carried out during the filling stage. There were 60 columns per treatment. When the roots were removed, the pipe hoop was opened, and the roots were cut with a saw at the desired depth. Meanwhile, the soil was not moved and the pipes were closed again. A 30-d drought period was started at the same time as the root removal, and the soil moisture was controlled to produce moderate drought conditions (the soil water content was 45%–50% of the maximum water holding capacity in the field). The soil columns were protected by a mobile rain shelter from the rain during the drought-stressed period. The soil water content was measured daily at soil depth of 10 cm using the soil water sensor ML2x (DELTA-T, United Kingdom). To ensure that the soil water content remained constant, an automatic equipment of drip irrigation was used to supplement the soil water to maintain the aimed soil water content. After root removal (from the beginning of the filling stage, August 17), plant aerial part and root bleeding sap from three plants in each treatment were collected every 10 d, and photosynthetic parameters and fluorescence parameters from ten plants per treatment, biomass and substances in root bleeding sap from three plants in each treatment were measured. The plants in all treatments were rewatered from 30 d until maturity and harvested on September 28 in the two years. All the parameters in the experiment except yield were collected in 2015.



Figure 1. Average temperature and sunshine duration during growth period in 2015 and 2016.



Figure 2. The cultivation of soil columns used in this study.

#### 2.2. Determination of Photosynthetic Parameters, Fluorescence Parameters and SPAD Values.

The net photosynthetic rate (Pn), stomatal conductance (Gs), and transpiration rate (Tr) of the second leaf from the top were measured using an LI-6400 portable photosynthetic system analyzer (LI-COR Inc., Lincoln, NE, USA) following the method of Chang et al. [36]. Ten plants were randomly selected for each treatment. Chlorophyll fluorescence parameters, including the initial fluorescence (Fo), maximum photosynthetic efficiency (Fv/Fm), photochemical quenching coefficient (qL) and electron transfer efficiency (ETR), were determined on the same leaf selected for determination of photosynthetic parameters by using a Junior-PAM fluorometer (Walz, Effeltrich, Germany) following the method described by Khoshbakht et al. [37]. The SPAD value (relative chlorophyll content) of the same leaf was determined with a SPAD chlorophyll meter; the upper, middle and lower parts of the leaf were measured, and the average value was calculated.

## 2.3. Collection of Root Bleeding Sap

Three uniform plants were selected in each treatment, and the plants were quickly cut with branch shears at the middle of the second stem node above the ground. After the incision was washed with distilled water and covered with preweighed absorbent cotton, the incision was placed in a valve bag and sealed tightly with waterproof tape. The root bleeding sap was collected from 6:00 am. to 18:00 pm., and the root bleeding intensity was determined by weighing (g·plant<sup>-1</sup>·12 h<sup>-1</sup>).

### 2.4. Determination of Osmotic Adjustment Substance and Hormone Content in Root Bleeding Sap

The soluble sugar content of the root bleeding sap was determined with the anthrone-sulfuric acid method described by Quan et al. [38]. The soluble protein content was measured with the Coomassie Brilliant Blue G-250 method described by Guzel and Terzi [39]. The free amino acid content was determined using the ninhydrin colorimetry method according to Sun et al. [40]. The ABA and gibberellin (GA<sub>3</sub>) contents were determined using enzyme-linked immunoassay (ELISA). The kit was provided by the Crop Chemical Control Research Center of China Agricultural University.

#### 2.5. Determination of Dry Matter Weight and Yield

Three plants were sampled from each treatment on the 10th, 20th, and 30th days from the initiation of root removal, and the dry matter weights (DMWs) of different organs (leaves, stems, sheaths, and panicles) were measured. The plant samples were killed at 105 °C for 30 min and dried at 80 °C to a constant weight. At harvest, 142 days after seeding, 10 plants were taken from each treatment, and the biological yield and grain yield were determined after air-drying.

## 2.6. Contribution of Roots of Different Depths to Yield

Contribution of roots below 30 cm to yield  $=\frac{\text{yield of CK-yield of R30}}{\text{yield of CK}} \times 100\%$ Contribution of roots below 60 cm to yield  $=\frac{\text{yield of CK-yield of R60}}{\text{yield of CK}} \times 100\%$ 

### 2.7. Statistical Analysis

This experiment was conducted as a complete randomized design. Significance of main effects of root removal was determined using one way analysis of variance in software SPSS 18.0. Means were separated by Duncan's multiple range test at p < 0.05. SPSS 18.0 software was also used for regression analysis. The data are presented as the means  $\pm$  standard deviation from all replications. Different characters indicate significant differences.

# 3. Results

## 3.1. Effects of Root Removal on Photosynthetic Performance

The photosynthetic parameters and SPAD values of plants in the different treatments were ranked as follows: CK > R60 > R30 (Table 1). In most measurement periods, R60 showed no significant difference as compared to CK, while R30 showed significantly reduced photosynthetic parameters and SPAD values. The greater reduction of the photosynthetic parameters as compared to CK was measured in variety J127. At 30 d after root removal, the Pn, Gs, Tr, and SPAD values of R30 in J127 were reduced by 32.15%, 53.85%, 18.18%, and 25.56% compared with those of CK in J127, respectively. The relative decreases at 10 or 20 d after root removal were similar, but slightly less pronounced decreases as compared to those at 30 days.

Fo in different treatments was highest in R30, followed by R60 and then CK, and R30 showed a significantly increased Fo compared with CK at all measurement periods (Table 2). In contrast, Fv/Fm, qL, and ETR were ranked as follows: CK > R60 > R30. At 30 d after root removal, both varieties in the R30 treatment showed significantly reduced Fv/Fm, qL, and ETR, which were decreased by 10.00%, 19.05%, 33.79% in J127 and 11.43%, 31.25%, and 17.85% in J305, respectively. R60 showed no significant difference from CK. The relative decreases at 10 or 20 d after root removal were similar, but slightly less pronounced decreases as compared to those at 30 days.

Cultivar	Days after Root Removal	Treatment	$Pn \\ (\mu mol \cdot m^{-2} \cdot s^{-1})$	Gs (mmol·m <sup>-2</sup> ·s <sup>-1</sup> )	Tr (mmol·m <sup>-2</sup> ·s <sup>-1</sup> )	SPAD Value
J127	10 d	R30	$20.29 \pm 2.51b$	$0.20 \pm 0.024b$	$3.77 \pm 0.79b$	$45.07 \pm 2.02b$
		R60	$27.16 \pm 0.20a$	$0.22 \pm 0.014$ ab	$5.90 \pm 0.41a$	$49.40 \pm 3.58a$
		СК	$30.00 \pm 1.85a$	$0.25\pm0.015a$	$6.73 \pm 0.48a$	$55.65 \pm 4.26a$
	20 d	R30	$17.97 \pm 0.31$ b	$0.11 \pm 0.004b$	$3.05 \pm 0.49c$	35.13 ± 2.86b
		R60	$23.18 \pm 2.55a$	$0.13 \pm 0.027a$	$4.10\pm0.20\mathrm{b}$	$37.93 \pm 1.16 \mathrm{b}$
		CK	$25.37 \pm 2.40a$	$0.24\pm0.025a$	$4.96\pm0.37a$	$47.30 \pm 2.61a$
	30 d	R30	$9.16 \pm 2.40b$	$0.06 \pm 0.016b$	$1.35 \pm 0.37b$	$29.03 \pm 2.78b$
		R60	$11.39 \pm 2.13ab$	$0.09 \pm 0.014$ ab	$1.64 \pm 0.28a$	$31.60 \pm 4.92$ ab
		CK	$13.50 \pm 2.92a$	$0.13\pm0.014a$	$1.65 \pm 0.28a$	$39.00 \pm 4.40a$
	10 d	R30	$22.26 \pm 1.81b$	$0.20 \pm 0.031b$	$4.51\pm0.51\mathrm{b}$	$48.63 \pm 1.70b$
		R60	$32.25 \pm 2.79a$	$0.25\pm0.016ab$	$5.38 \pm 0.25$ ab	$54.50 \pm 3.95$ ab
J305		CK	$32.81 \pm 2.44a$	$0.30 \pm 0.050a$	$6.12 \pm 0.99a$	$57.66 \pm 6.39a$
	20 d	R30	21.24 ± 1.98b	$0.18 \pm 0.017$ b	$3.52 \pm 0.85a$	$43.33 \pm 6.64b$
		R60	$24.45 \pm 2.25ab$	$0.23 \pm 0.045a$	$4.35 \pm 0.83a$	$49.37 \pm 1.30$ ab
		СК	$27.88 \pm 3.98a$	$0.27 \pm 0.011a$	$5.09 \pm 0.72a$	$54.36 \pm 6.21a$
	30 d	R30	$10.54 \pm 2.52b$	$0.10 \pm 0.012b$	$1.59 \pm 0.25b$	$32.00 \pm 2.98b$
		R60	$12.47 \pm 1.77a$	$0.16 \pm 0.008a$	$1.76 \pm 0.15a$	$38.03 \pm 3.07 \mathrm{ab}$
		CK	$13.17 \pm 1.39a$	$0.17 \pm 0.010a$	$1.81\pm0.20a$	$42.18 \pm 3.76a$

Table 1. Effects of root removal in different soil depths on photosynthetic parameters and SPAD values.

No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60; the net photosynthetic rate, Pn; stomatal conductance, Gs; transpiration rate, Tr; chlorophyll relative content, SPAD value. Different characters within the same column indicate significant differences at p < 0.05. The data are represented as the means  $\pm$  standard deviation (n = 10).

# 3.2. Effects of Root Removal on Root Bleeding

Root bleeding intensity was significantly affected by root excision (Figure 3). R30 and R60 significantly reduced root bleeding intensity in J127 at 10 and 20 d after root excision. The root bleeding intensity was decreased by 42.69% and 35.30% in R30 and R60 at 10 d after root removal and 41.22% and 33.64% at 20 d after root removal compared with that in CK, respectively. In J305, R30 and R60 showed significantly reduced root bleeding intensity at 20 and 30 d after root removal. Compared with that in CK, the root bleeding intensity was decreased by 46.52% and 24.46% in R30 and R60 at 20 d after root removal and 33.99% at 30 d after root removal, respectively.

Cultivar	Days after Root Removal	Treatment	Fo	Fv/Fm	qL	ETR
J127	10 d	R30	$255.36 \pm 5.66a$	$0.75 \pm 0.050a$	$0.22 \pm 0.009a$	$24.49 \pm 0.90b$
		R60	$230.67 \pm 6.66b$	$0.77 \pm 0.003a$	$0.24 \pm 0.019a$	$28.48 \pm 0.89a$
		СК	$219.66 \pm 5.64b$	$0.79 \pm 0.002a$	$0.25\pm0.029a$	$31.05 \pm 2.32a$
	20 d	R30	278.35±9.10a	$0.69 \pm 0.006b$	$0.18 \pm 0.029a$	$21.53 \pm 1.24 c$
		R60	250.41±4.87b	$0.75 \pm 0.026a$	$0.20 \pm 0.034a$	$25.61 \pm 0.40b$
		СК	236.31±5.00c	$0.77 \pm 0.001a$	0.22±0.022a	$28.24 \pm 0.94a$
	30 d	R30	319.00±11.37a	$0.63 \pm 0.039$ b	0.17±0.015b	$16.05 \pm 2.42b$
		R60	293.463±16.57ab	$0.65 \pm 0.019$ ab	$0.20 \pm 0.034a$	$21.80 \pm 1.48a$
		CK	258.33±15.51b	$0.70 \pm 29a$	$0.21 \pm 25a$	$24.24 \pm 1.95a$
J305	10 d	R30	$236.14 \pm 16.97a$	$0.77 \pm 0.004a$	$0.22 \pm 0.033b$	$28.32 \pm 1.00b$
		R60	$227.20 \pm 7.14ab$	$0.78 \pm 0.001a$	$0.31 \pm 0.054a$	$31.14 \pm 2.92ab$
		CK	$216.46 \pm 6.85b$	$0.79 \pm 0.028a$	$0.32 \pm 0.025a$	$34.66 \pm 2.08a$
	20 d	R30	$261.96 \pm 4.88a$	$0.71\pm0.022\mathrm{b}$	$0.17\pm0.005c$	$24.49 \pm 0.53 \mathrm{b}$
		R60	$238.42 \pm 7.23b$	$0.75 \pm 0.020$ ab	$0.23\pm0.016\mathrm{b}$	$27.48 \pm 0.39$ ab
		СК	$228.67 \pm 6.03c$	$0.76 \pm 0.025a$	$0.27 \pm 0.020a$	$30.03 \pm 2.80a$
	30 d	R30	$292.00 \pm 19.67a$	$0.62 \pm 0.079$ b	$0.11 \pm 0.100b$	$20.06 \pm 0.79b$
		R60	$262.67 \pm 11.50$ ab	$0.67\pm0.023 ab$	$0.14\pm0.149a$	$23.56 \pm 2.02ab$
		CK	$246.67 \pm 11.59b$	$0.70 \pm 0.012a$	$0.16 \pm 0.156a$	$24.42 \pm 0.74a$

Table 2. Effects of root removal in different soil depths on fluorescence parameters.

No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60; initial fluorescence, Fo; maximum photosynthetic efficiency, Fv/Fm; photochemical quenching coefficient, qL; electron transfer efficiency, ETR. Different characters within the same column indicate significant differences at p < 0.05. The data are represented as the means ± standard deviation (n = 10).



**Figure 3.** Effects of root removal in different soil depths on root bleeding intensity. No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60. Different characters at the same stage indicate significant differences at p < 0.05. Vertical bars represent the standard deviation of three replications (n = 3).

The content of osmotic adjustment substances in the root bleeding sap was significantly affected by root removal (Figure 4). R30 led to a significantly reduced soluble sugar content. R60 led to a significantly reduced soluble sugar content in J127 compared with CK at 10 and 20 d after root removal; the decreases were 25.71% and 12.98%, respectively. In J305, R60 also led to significantly decreased soluble sugar contents; the decreases were 20.94%, 17.33%, and 11.78% at 10, 20, and 30 d after root excision, respectively. Furthermore, R30 significantly reduced the soluble protein content of the root

bleeding sap of J305 by 26.71%, 20.09%, and 31.55% at 10, 20, and 30 d after root removal, respectively. In contrast, R60 had no significant effect on the soluble protein content of the root bleeding sap at 10 and 30 d after root removal. R30 significantly reduced the amino acid content of the root bleeding sap by 47.41%, 34.63%, and 32.33% in J127 at 10, 20, and 30 d after root removal, respectively, and by 40.59%, 29.82%, and 30.23% in J305 at 10, 20, and 30 d after root removal, respectively. R60 significantly reduced the amino acid content in J127 by 30.94%, 20.65%, and 25.95% at 10, 20, and 30 d after root excision, respectively, and by 28.42% and 18.76% in J305 at 10 and 30 d after root excision, respectively.



**Figure 4.** Effects of root removal in different soil depths on soluble sugar content, soluble protein content, and amino acid content in root bleeding sap. No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60. Different characters at the same stage indicate significant differences at p < 0.05. Vertical bars represent the standard deviation of three replications (n = 3).

The GA<sub>3</sub> content of the root bleeding sap was significantly affected by root excision. R30 significantly reduced the GA<sub>3</sub> content by 36.81%, 39.21%, and 49.04% in J127 at 10, 20, and 30 d after root removal and by 25.11%, 35.02%, and 37.87% in J305, respectively, compared with that in CK. R60 significantly reduced the GA<sub>3</sub> content in J127 by 22.25%, 25.67%, and 36.66% at 10, 20, and 30 d



after root removal, respectively, and by 20.44% and 30.13% in J305 at 20 and 30 d after root removal (Figure 5).

**Figure 5.** Effects of root removal in different soil depths on gibberellin (GA<sub>3</sub>) content and abscisic acid (ABA) content in root bleeding sap. No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60. Different characters at the same stage indicate significant differences at p < 0.05. Vertical bars represent the standard deviation of three replications (n = 3).

At 10 d after root excision, compared with that of CK, the ABA content in the root bleeding sap of R30 was significantly increased, while that of R60 showed no significant difference from CK. R30 significantly reduced the ABA content in the root bleeding sap of J127 at 20 and 30 d after root removal. R60 significantly reduced the ABA content in the root bleeding sap of J127 at 20 and 30 d after root after root removal and that of J305 at 30 d after root removal (Figure 5).

### 3.3. Effects of Root Removal on Dry Matter Accumulation and Yield

Compared with CK, the dry weight of leaves, stems, sheaths, and panicles in J127 decreased by 33.10%, 46.88%, 43.08%, and 24.63% under R30 at 30 d after root removal, respectively (Table 3). R60 had no significant effect on the dry weights of leaves and stems in J127 but significantly reduced those of sheaths and panicles by 30.11% and 13.15% compared with CK, respectively. In J305, R60 had no significant effects on the dry weights of the leaves, stems, and sheaths but significantly decreased the dry weight of the panicles, which was reduced by 14.27% compared to that of CK. The relative decreases at 10 or 20 d after root removal were similar, but slightly less pronounced decreases as compared to those at 30 days.

30 d

R60

CK

Cultivar	Days after Root Removal	Treatment	Leaf (g∙plant <sup>-1</sup> )	Stem (g∙plant <sup>-1</sup> )	Sheath (g∙plant <sup>-1</sup> )	Panicle (g∙plant <sup>-1</sup> )
J127	10 d	R30	$21.67 \pm 3.94a$	$40.18 \pm 4.76b$	$14.95 \pm 1.72a$	$75.25 \pm 3.53b$
		R60	$24.41 \pm 2.22a$	$57.73 \pm 9.56a$	$16.92 \pm 2.22a$	$87.37 \pm 6.85a$
		СК	$26.22 \pm 1.94a$	$61.14 \pm 5.57a$	$19.08 \pm 3.65a$	$98.31 \pm 6.30a$
	20 d	R30	18.53 ± 0.71b	35.35 ± 1.32b	$12.05 \pm 0.78c$	70.39 ± 5.39b
		R60	$22.59 \pm 1.85a$	$48.85 \pm 7.88a$	$14.87 \pm 0.70 b$	78.71 ± 9.31ab
		CK	$24.32 \pm 1.54a$	$56.94 \pm 6.93a$	$17.51 \pm 0.66a$	$87.72 \pm 8.78a$
	30 d	R30	$14.15 \pm 1.97b$	27.31 ± 13.34b	$8.43 \pm 0.34c$	63.98 ± 2.61c
		R60	$21.07 \pm 0.74a$	$40.87 \pm 4.19$ ab	$10.35 \pm 0.67b$	73.73 ± 3.13b
		CK	$21.15 \pm 1.41a$	$51.41 \pm 3.79a$	$14.81\pm0.77a$	$84.89 \pm 2.88a$
J305	10 d	R30	$25.70 \pm 2.98a$	$63.77 \pm 7.72b$	$19.12 \pm 6.13a$	$89.17 \pm 8.81b$
		R60	$28.76 \pm 1.24a$	$83.02 \pm 4.04a$	$19.73 \pm 2.50a$	$106.48 \pm 1.73a$
		CK	$29.23 \pm 1.68a$	$85.83 \pm 10.71a$	$21.61 \pm 3.31a$	$115.24 \pm 11.30a$
	20 d	R30	$21.98 \pm 2.05b$	53.44 ± 10.77b	$14.52 \pm 2.67a$	84.18 ± 2.93b
		R60	$26.46 \pm 2.51a$	$79.43 \pm 2.91a$	$17.44 \pm 2.53a$	$102.95 \pm 7.12a$
		СК	$28.27 \pm 1.27a$	$80.87 \pm 6.90a$	$20.59 \pm 1.13a$	$109.80 \pm 5.87a$
		R30	$19.76 \pm 1.84b$	$50.41 \pm 5.23b$	$12.81 \pm 2.45b$	$70.06 \pm 1.58c$

Table 3. Effects of root removal in different soil depths on dry matter accumulation.

No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60. Different characters within the same column indicate significant differences at p < 0.05. The data are represented as the means  $\pm$  standard deviation (n = 3).

 $67.64 \pm 8.78a$ 

 $73.15 \pm 5.33a$ 

 $25.22 \pm 2.13a$ 

 $25.49 \pm 2.95a$ 

 $16.77 \pm 3.87a$ 

 $19.60 \pm 1.60a$ 

 $81.56 \pm 0.23b$ 

 $95.14 \pm 2.82a$ 

Both R30 and R60 significantly reduced grain yield (Table 4). The two-year average grain yield of R30 and R60 decreased by 53.46% and 36.04% compared with that of CK in J127 and by 48.16% and 33.74% in J305. The biological yield was also significantly affected by root removal. The average biological yield of R30 and R60 in J127 decreased by 19.41% and 9.40% compared with that of CK and by 31.42% and 19.86% in J305, respectively. The yield trend of the two-year experiment was consistent.

Year	Cultivar	Treatment	Grain Yield (g/plant)	Biological Yield (g/plant)
2015	J127	R30	$60.34 \pm 1.00c$	$161.18 \pm 8.30c$
		R60	$72.99 \pm 3.64b$	$175.39 \pm 0.71b$
		СК	$102.43 \pm 9.24a$	$197.71 \pm 11.47a$
	J305	R30	$83.34 \pm 4.82b$	$193.44 \pm 3.83b$
		R60	$94.77 \pm 4.21b$	$213.13 \pm 5.56b$
		СК	$125.37 \pm 7.72a$	$231.10 \pm 6.36a$
2016 -	J127	R30	$54.48 \pm 0.45 \mathrm{c}$	$184.39 \pm 25.07c$
		R60	$84.82 \pm 2.41b$	$243.04 \pm 9.08b$
		СК	$144.30 \pm 2.28a$	$295.32 \pm 36.83a$
	J305	R30	$42.72 \pm 10.88b$	$164.21 \pm 9.34b$
		R60	$66.37 \pm 6.97b$	$204.78 \pm 20.35b$
		СК	$117.82 \pm 13.33a$	$290.40 \pm 49.96a$

Table 4. Effects of root removal in different soil depths on grain and biological yield in 2015 and 2016.

No roots removed, CK; roots removed at 30 cm underground, R30; roots removed at 60 cm underground, R60. Different characters within the same column indicate significant differences at p < 0.05. The data are represented as the means  $\pm$  standard deviation (n = 10).

### 3.4. Correlation of Root Bleeding with Grain Yield and Pn

A significant positive relationship ( $R^2 = 0.6072$ , p < 0.01) was found between the Pn and root bleeding intensity, indicating that the increasing root bleeding intensity had a positive influence on Pn.

Similarly, there was a significant positive relationship ( $R^2 = 0.5862$ , p < 0.01) between grain yield and root bleeding intensity, indicating that the increasing root bleeding intensity had a positive influence on grain yield (Figure 6).



**Figure 6.** The correlation between root bleeding intensity and grain yield, Pn (net photosynthetic rate). The data was from two cultivars and included every treatment and replicates; p < 0.01 represents extremely significant correlation.

## 4. Discussion

Root activity reflects the strength of root metabolic capacity and has an important role in promoting the formation of yield [41,42]. Munns et al. [43] reported that a decline in osmotic adjustment substances increased the osmotic potential of plant root cells, resulting in a decrease in the water absorption capacity of the root system. In this study, R30 and R60 significantly reduced root bleeding intensity and osmotic adjustment substance content, indicating that roots below 30 cm still had strong water absorption capacity under drought conditions during the filling stage. Meanwhile, some indicators in the root bleeding sap of R60, such as soluble protein and soluble sugar, were significantly higher than those of R30 during the filling process, indicating that deeper root systems (below 30 cm soil depth) hid high physiological activity and strong regulatory effect on the aboveground parts. With respect to signaling substances, root excision significantly reduced the  $GA_3$  content in the root bleeding sap. This result indicates that the reduction of GA<sub>3</sub> synthesis in the root system should lead to a decrease of the available  $GA_3$  in the aerial part.  $GA_3$  is an important hormone in plants that can promote plant growth and material accumulation [44]. These results suggested that the decrease in synthesis and output of GA<sub>3</sub> may be one of the reasons for the reduction in aboveground dry matter, and this effect was more obvious in R30. The changes in ABA were more complicated due to root excision and plant senescence. On the one hand, the change trend in ABA content in the root bleeding sap of CK reflects a response to drought conditions and senescence during the filling stage. A similar change trend of ABA was also found in the results of Abid et al. [45]. The changes in ABA content in R30 and R60 may indicate a stress response in the root system in the early stage due to root excision. This damage and loss in the root system decreased the ability of the roots to synthesize ABA, which significantly reduced the ABA content exported from the roots to the shoots. Lower ABA content in the leaves could reduce plant drought resistance [46], ultimately leading to a reduction in yield. The correlation of the root bleeding intensity with Pn and grain yield also implied that higher root activity could better support the development of photosynthetic potential that ultimately resulted in higher yield.

Song et al. [47] and Qi et al. [48] reported that deep roots play a critical role to improve root vitality, maintain root nutrition and water supply to the aerial part, promote the production of photosynthetic

materials and grain filling, and increase yield. In the present study, regardless of R30 or R60, the yield of sorghum was significantly reduced by root removal, reflecting the important role of deep roots in yield production. Root excision at 30 cm underground significantly reduced leaf chlorophyll content (SPAD value), photosynthetic level, Fv/Fm, qL, and ETR and increased Fo. An increased Fo and a decreased Fv/Fm indicate that the PSII reaction center is damaged [49]. These results revealed that root excision directly reduced the function of the PSII reaction center, and photosynthetic activity and electron transfer were affected. As a result, the photosynthetic capacity was reduced, and the yield was significantly reduced. In addition, the decreased photosynthetic capacity reduced the overall biomass of the aerial parts, which manifested as decreased dry weights of the sorghum leaves, stems, sheaths, and panicles. However, the effect of root excision at 60 cm underground on the photosynthetic parameters was not significant, indicating that R60 had little effect on photosynthetic performance. From the perspective of root system distribution, the root system was shown to be mainly distributed in the upper 0–30 cm of topsoil, which contained up to approximately 95% of the total number of roots in cotton and maize, respectively [50,51], indicating that roots of taproot and fibrous root crops are mainly distributed in shallow layers, and the surface root system was directly related to photosynthesis [52]. However, in this study, the contribution of roots below 60 cm accounted for more than 30% of the yield, indicating that deeper root activity, which was reflected by the root bleeding intensity and component content, played a critical role in yield production under drought conditions during the filling stage. Schittenhelm et al. [53] indicated that root depth was also a significant feature of drought resistance in sorghum. Common farming fertilization is mainly concentrated in the plow layer, and drought also usually occurs in the cultivated layer, which reduces nutrient availability. Although the root system can penetrate deeper soil depths to obtain water under drought conditions, nutrients are relatively scarce in these deeper layers, leading to a degree of spatial dislocation between nutrients and water. As the root system tends to grow toward fertilizer and water, the spatial distribution of the root system changes accordingly; thus, the role of deep roots will be more obvious under drought conditions.

In brief, R30 and R60 significantly reduced the biological and grain yield of sorghum, especially grain yield. R30 mainly reduced yield by reducing photosynthetic level and root activity, while R60 had no significant effect on photosynthetic characteristics. Although root activity was significantly reduced in R60, it was still significantly higher than that of R30, indicating that the greater vitality of deeper root systems (soil depths below 30 cm) was the reason for their greater contribution to yield, especially under drought conditions. In conclusion, the deep roots of sorghum play a crucial role in yield production, but the roots in different soil depths regulate yield production differently. Our results support the hypothesis that deep roots of sorghum greatly contribute to yield production and thus merit investigation as a potential agronomic trait for more yield under drought conditions.

Author Contributions: Conceptualization, Q.W. and Y.Z.; Investigation, Q.W., Y.G., J.Z. and Y.W.; Project administration, Q.W.; Supervision, R.Z., Y.Z., M.X., W.X. and R.H.; Validation, Y.Z. and R.H.; Writing—original draft, X.C.; Writing—review and editing, Y.Z., X.C. All authors have read and agreed to the published version of the manuscript.

Funding: This research and the APC was funded by the China Agriculture Research System (CARS-06).

Acknowledgments: This work is financially supported by the China Agriculture Research System (CARS-06).

Conflicts of Interest: The authors declare no conflict of interest.

#### References

- Arai-Sanoh, Y.; Takai, T.; Yoshinaga, S.; Nakano, H.; Kojima, M.; Sakakibara, H.; Kondo, M.; Uga, Y. Deep rooting conferred by DEEPER ROOTING 1 enhances rice yield in paddy fields. *Sci. Rep.* 2014, *4*, 5563. [CrossRef] [PubMed]
- Rachmilevitch, S.; Lambers, H.; Huang, B. Root respiratory characteristics associated with plant adaptation to high soil temperature for geothermal and turf-type Agrostis species. *J. Exp. Bot.* 2006, 57, 623–631. [CrossRef] [PubMed]

- 3. Silva, E.V.D.; Bouillet, J.P.; Gonçalves, J.L.D.M.; Junior, C.H.A.; Trivelin, P.C.O.; Hinsinger, P.; Jourdan, C.; Nouvellon, Y.; Stape, J.L.; Laclau, J.P. Functional specialization of Eucalyptus fine roots: Contrasting potential uptake rates for nitrogen, potassium and calcium tracers at varying soil depths. *Funct. Ecol.* **2011**, 25, 996–1006. [CrossRef]
- 4. Guo, H.; York, L.M. Maize with fewer nodal roots allocates mass to more lateral and deep roots that improve nitrogen uptake and shoot growth. *J. Exp. Bot.* **2019**, *70*, 5299–5309. [CrossRef] [PubMed]
- Kashiwagi, J.; Krishnamurthy, L.; Crouch, J.H.; Serraj, R. Variability of root length density and its contributions to seed yield in chickpea (*Cicer arietinum* L.) under terminal drought stress. *Field Crop. Res.* 2006, 95, 171–181. [CrossRef]
- 6. Liedgens, M.; Richner, W. Relation between maize (*Zea mays* L.) leaf area and root density observed with minirhizotrons. *Eur. J. Agron.* **2001**, *15*, 131–141. [CrossRef]
- 7. Craine, J.M.; Wedin, D.A.; Chapin, F.S.; Reich, P.B. Relationship between the structure of root systems and resource use for 11 North American grassland plants. *Plant Ecol.* **2003**, *165*, 85–100. [CrossRef]
- 8. Vamerali, T.; Saccomani, M.; Bona, S.; Mosca, G.; Guarise, M.; Ganis, A. A comparison of root characteristics in relation to nutrient and water stress in two maize hybrids. *Plant Soil* **2003**, *255*, 157–167.
- 9. Bakhshandeh, S.; Kertesz, M.A.; Corneo, P.E.; Dijkstra, F.A. Dual-labeling with15N and H218O to investigate water and N uptake of wheat under different water regimes. *Plant Soil* **2016**, *408*, 429–441. [CrossRef]
- Sebastian, J.; Yee, M.; Viana, W.G.; Rellan-Alvarez, R.; Feldman, M.; Priest, H.D.; Trontin, C.; Lee, T.; Jiang, H.; Baxter, I.; et al. Grasses suppress shoot-borne roots to conserve water during drought. *Proc. Natl. Acad. Sci. USA* 2016, *113*, 8861–8866. [CrossRef]
- Hammer, G.L.; Dong, Z.; McLean, G.; Doherty, A.; Messina, C.; Schussler, J.; Zinselmeier, C.; Paszkiewicz, S.; Cooper, M. Can Changes in Canopy and/or Root System Architecture Explain Historical Maize Yield Trends in the U.S. Corn Belt? *Crop Sci.* 2009, 49, 299–312. [CrossRef]
- 12. Gewin, V. Food: An underground revolution. Nature 2010, 466, 552–553. [CrossRef] [PubMed]
- 13. Thorup-Kristensen, K.; Rasmussen, C.R. Identifying new deep-rooted plant species suitable as undersown nitrogen catch crops. *J. Soil Water Conserv.* **2015**, *70*, 399–409. [CrossRef]
- 14. Giambelluca, T.W.; Mudd, R.G.; Liu, W.; Ziegler, A.D.; Kobayashi, N.; Kumagai, T.; Miyazawa, Y.; Lim, T.K.; Huang, M.Y.; Fox, J.; et al. Evapotranspiration of rubber (*Hevea brasiliensis*) cultivated at two plantation sites in Southeast Asia. *Water Resour. Res.* **2016**, *52*, 660–679. [CrossRef]
- 15. Manschadi, A.M.; Christopher, J.; de Voil, P.; Hammer, G.L. The role of root architectural traits in adaptation of wheat to water-limited environments. *Funct. Plant Biol.* **2006**, *33*, 823. [CrossRef]
- 16. Palta, J.A.; Chen, X.; Milroy, S.P.; Rebetzke, G.J.; Dreccer, M.F.; Watt, M. Large root systems: Are they useful in adapting wheat to dry environments? *Funct. Plant Biol.* **2011**, *38*, 347–354. [CrossRef]
- 17. Thorup-Kristensen, K.; Cortasa, M.S.; Loges, R. Winter wheat roots grow twice as deep as spring wheat roots, is this important for N uptake and N leaching losses? *Plant Soil* **2009**, *322*, 101–114. [CrossRef]
- Thorup-Kristensen, K.; Dresbøll, D.B.; Kristensen, H.L. Crop yield, root growth, and nutrient dynamics in a conventional and three organic cropping systems with different levels of external inputs and N re-cycling through fertility building crops. *Eur. J. Agron.* 2012, *37*, 66–82. [CrossRef]
- 19. Lopes, M.S.; Reynolds, M.P. Partitioning of assimilates to deeper roots is associated with cooler canopies and increased yield under drought in wheat. *Funct. Plant Biol.* **2010**, *37*, 147–156. [CrossRef]
- 20. Kell, D.B. Breeding crop plants with deep roots: Their role in sustainable carbon, nutrient and water sequestration. *Ann. Bot.* **2011**, *108*, 407–418. [CrossRef]
- 21. Wasson, A.P.; Rebetzke, G.J.; Kirkegaard, J.A.; Christopher, J.; Richards, R.A.; Watt, M. Soil coring at multiple field environments can directly quantify variation in deep root traits to select wheat genotypes for breeding. *J. Exp. Bot.* **2014**, *65*, 6231–6249. [CrossRef] [PubMed]
- 22. Thorup-Kristensen, K.; Kirkegaard, J. Root system-based limits to agricultural productivity and efficiency: The farming systems context. *Ann. Bot.* **2016**, *118*, 573–592. [CrossRef] [PubMed]
- 23. Chaves, M.M.; Maroco, J.P.; Pereira, J.S. Understanding plant responses to drought—From genes to the whole plant. *Funct. Plant Biol.* 2003, *30*, 239–264. [CrossRef]
- 24. Burgess, S.S.O.; Adams, M.A.; Turner, N.C.; Ong, C.K. The redistribution of soil water by tree root systems. *Oecologia* **1998**, *115*, 306–311. [CrossRef] [PubMed]

- Domec, J.C.; Warren, J.M.; Meinzer, F.C.; Brooks, J.R.; Coulombe, R. Native root xylem embolism and stomatal closure in stands of Douglas-fir and ponderosa pine: Mitigation by hydraulic redistribution. *Oecologia* 2004, 141, 7–16. [CrossRef] [PubMed]
- 26. Bleby, T.M.; Mcelrone, A.J.; Jackson, R.B. Water uptake and hydraulic redistribution across large woody root systems to 20 m depth. *Plant Cell Environ.* **2010**, *33*, 2132–2148. [CrossRef]
- 27. FAO. Food and Agriculture Organization of the United Nations. 2018. Available online: http://faostat3.fao. org/download/Q/QC/E (accessed on 24 April 2020).
- Wang, N.; Wang, Y.T.; Yu, J.L.; Zhou, Y.F.; Wu, Q.; Gao, Y.; Xu, W.J.; Huang, R.D. Prioritization of feasible physiological parameters in drought tolerance evaluation in sorghum: A grey relational analysis. *Zemdirbyste* 2015, 102, 457–464. [CrossRef]
- 29. Haussmann, B.; Mahalakshmi, V.; Reddy, B.; Seetharama, N.; Hash, C.; Geiger, H. QTL mapping of stay-green in two sorghum recombinant inbred populations. *Theor. Appl. Genet.* **2002**, *106*, 133–142. [CrossRef]
- 30. Mace, E.S.; Tai, S.S.; Gilding, E.K.; Li, Y.H. Whole-genome sequencing reveals untapped genetic potential in Africa's indigenous cereal crop sorghum. *Nat. Commun.* **2013**, *4*, 2320. [CrossRef]
- 31. Paterson, A.H. Genomics of Sorghum. Int. J. Plant Genomics 2008, 2008, 362451. [CrossRef]
- Zhang, D.F.; Zeng, T.R.; Liu, X.Y.; Gao, C.X.; Li, Y.X.; Li, C.H.; Song, Y.C.; Shi, Y.S.; Wang, T.Y.; Li, Y. Transcriptomic profiling of sorghum leaves and roots responsive to drought stress at the seedling stage. *J. Integ. Agric.* 2019, *18*, 1980–1995. [CrossRef]
- 33. Wang, D.Q.; Zhou, Y.F.; Lu, Z.B.; Xiao, M.J.; Xu, W.J.; Huang, R.D. Root morphology and activity of stay green sorghum under water stress. *Agric. Res. Arid Area.* **2012**, *30*, 73–76.
- 34. Smit, A.L.; Bengough, A.G.; Engels, C.; De Noordwijk, V. Root Methods: A Handbook. J. Agron. Crop Sci. 2002, 188, 64.
- 35. Maeght, J.L.; Rewald, B.; Pierret, A. How to study deep roots—And why it matters. *Front. Plant Sci.* **2013**, *4*, 299. [CrossRef] [PubMed]
- Chang, B.; Yang, L.; Cong, W.; Zu, Y.; Tang, Z. The improved resistance to high salinity induced by trehalose is associated with ionic regulation and osmotic adjustment in *Catharanthus roseus*. *Plant Physiol. Biochem*. 2014, 77, 140–148. [CrossRef]
- 37. Khoshbakht, D.; Asghari, M.R.; Haghighi, M. Effects of foliar applications of nitric oxide and spermidine on chlorophyll fluorescence, photosynthesis and antioxidant enzyme activities of citrus seedlings under salinity stress. *Photosynthetica* **2018**, *56*, 1313–1325. [CrossRef]
- 38. Quan, R.; Shang, M.; Zhang, H.; Zhao, Y.; Zhang, J. Improved chilling tolerance by transformation with *betA* gene for the enhancement of glycinebetaine synthesis in maize. *Plant Sci.* **2004**, *166*, 141–149. [CrossRef]
- Guzel, S.; Terzi, R. Exogenous hydrogen peroxide increases dry matter production, mineral content and level of osmotic solutes in young maize leaves and alleviates deleterious effects of copper stress. *Bot. Stud.* 2013, 54, 26. [CrossRef]
- 40. Sun, S.W.; Lin, Y.C.; Weng, Y.M.; Chen, M.J. Efficiency improvements on ninhydrin method for amino acid quantification. *J. Food Compos. Anal.* **2006**, *19*, 112–117. [CrossRef]
- 41. Chu, G.; Chen, T.T.; Wang, Z.Q.; Yang, J.C.; Zhang, J.H. Morphological and physiological traits of roots and their relationships with water productivity in water-saving and drought-resistant rice. *Field Crop. Res.* **2014**, *162*, 108–119. [CrossRef]
- 42. Guan, D.H.; Al-Kaisi, M.M.; Zhang, Y.S.; Duan, L.S.; Tan, W.M.; Zhang, M.C.; Li, Z.H. Tillage practices affect biomass and grain yield through regulating root growth, root-bleeding sap and nutrients uptake in summer maize. *Field Crop. Res.* **2014**, *157*, 89–97. [CrossRef]
- 43. Munns, R.; Passioura, J.; Colmer, T.; Byrt, C. Osmotic adjustment and energy limitations to plant growth in saline soil. *New phytol.* **2019**, *225*, 1091–1096. [CrossRef] [PubMed]
- 44. Zhang, S.; Zhang, D.; Fan, S.; Du, L.; Shen, Y.; Xing, L.; Li, Y.; Ma, J.; Han, M. Effect of exogenous GA<sub>3</sub> and its inhibitor paclobutrazol on floral formation, endogenous hormones, and flowering-associated genes in 'Fuji' apple (*Malus domestica* Borkh.). *Plant Physiol. Biochem.* **2016**, *107*, 178–186. [CrossRef]
- 45. Abid, M.; Shao, Y.H.; Liu, S.X.; Wang, F.; Gao, J.W.; Jang, D.; Tian, Z.W.; Dai, T.B. Pre-drought priming sustains grain development under post-anthesis drought stress by regulating the growth hormones in winter wheat (*Triticum aestivum* L.). *Planta* **2017**, *246*, 509–524. [CrossRef] [PubMed]

- Zhou, Y.F.; Wang, D.Q.; Lu, Z.B.; Wang, N.; Wang, Y.T.; Li, F.X.; Xu, W.J.; Huang, R.D. Effects of Drought Stress on Photosynthetic Characteristics and Endogenous Hormone ABA and CTK Contents in Green-Stayed Sorghum. *Sci. Agric. Sin.* 2014, 47, 655–663.
- 47. Song, R.; Wu, C.S.; Ma, L.Y. Effect of application of combined fertilizers on the root system of maize. *Acta Agron. Sin.* **2002**, *28*, 393–396.
- Qi, W.Z.; Liu, H.H.; Liu, P.; Dong, S.T.; Zhao, B.Q.; So, H.B.; Li, G.; Liu, H.D.; Zhang, J.W.; Zhao, B. Morphological and physiological characteristics of corn (*Zea mays* L.) roots from cultivars with different yield potentials. *Eur. J. Agron.* 2012, *38*, 54–63. [CrossRef]
- Ge, J.L.; Shi, L.; Gu, W.B.; Tang, Y.D.; Zhang, J.Z.; Jiang, C.D.; Ren, D.M. Photosynthetic Characteristics and the Regulation of PhotosystemII Function in Salt-Stressed Sweet Sorghum Seedlings. *Acta Agron. Sin.* 2007, 33, 1272–1278.
- 50. Liang, Y.; He, W.S.; Dai, X.H.; Ma, K.; Hou, X.Q. Effects of Planting Density and Row Spacing on Root-shoot Spatial Distribution and Grain Yield of Spring Maize. *J. Maize Sci.* **2016**, *24*, 97–102.
- 51. Li, Y.S.; Feng, L.P.; Guo, M.L.; Han, X.X. Studies on the growth characteristics of root system and its relation with cultural practices and yield in cotton (*G. hirsutum* L.) the effects of cultural practices on the growth of root system and its relation with above ground parts and yield. *Cotton Sci.* **1992**, *4*, 59–66.
- 52. Zhao, Q.Z.; Qiao, J.F.; Liu, H.; Tian, Z.Q. Relationship Between Root and Leaf Photosynthetic Characteristic in Rice. *Sci. Agric. Sin.* **2007**, *40*, 1064–1068.
- 53. Schittenhelm, S.; Schroetter, S. Comparison of Drought Tolerance of Maize, Sweet Sorghum and Sorghum-Sudangrass Hybrids. J. Agron. Crop Sci. 2014, 200, 46–53. [CrossRef]



© 2020 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).