

DROUGHT-HARDENING IMPROVES FLOODING TOLERANCE OF MAIZE AT SEEDLING STAGE UNDER DROUGHT-FLOOD ABRUPT ALTERNATIONS

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Abstract. In recent years, the frequency and intensity of drought–flood abrupt alterations due to global climate change have drastically increased. In contrast, plant response to the drought-flood abrupt alternations remains unclear. In the current study to reveal maize’s physiological response to drought-flood abrupt alternations, these were carried out in two stages in this study, i.e., three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity) in stage I, and then five durations of flooding (75%-WDi, 65%-WDi and 55%-WDi) in stage II. The results indicated that the 65%-WDi treatment significantly decreased the leaf area and biomass of maize seedlings in stage I, while maintaining plant growth and reaching the control level in stage II. Abscisic acid (ABA) concentration in maize leaves and roots of 0-5 cm depth was sensitive to drought stress. Under drought–flood abrupt alternations, the root system in 55%-WD₅ and 65%-WD₇ exhibited strong ABA signal, which was sensitive to drought-flood abrupt alternations. Therefore, at the maize seedling stage, proper control of soil moisture is beneficial for improving stress tolerance at later stages, but stress tolerance after drought–flood abrupt alterations is related to the degree of drought exposure at the early stage.

Keywords: leaf area, biomass, ABA concentration, stomatal conductance *e*, transpiration

Introduction

Climate change is an ongoing issue that affects global agricultural development. Drought, flooding, and their secondary hazards are the primary problems that limit agricultural development, and severely affect crop growth and yield formation (Ashraf et al., 2010; Kim et al., 2018; Kakanur et al., 2021; Kerdee et al., 2021; Mehar et al., 2021). It was demonstrated that drought stress in the early crop growth stages can promote the emergence of certain metabolites and structural functions which are beneficial for the improvement of crop resistance to stresses in the later growth stages (Reddy et al., 2004). Drought stress in crop seedling stage causes an extension of the root system to the deep layers of soil (Li et al., 2020) and alters crop’s biological performances (Wang et al., 2020), etc. An essential physiological response to drought stress in plants is the change in the abscisic acid (ABA) concentration (Hussain et al., 2012), which is produced in plant tissues after drought and transported to regulate the leaf stomatal conductance, thereby maintaining water balance in the plant (Gil et al., 2007; Ren et al., 2018). The degree of crop damage due to water stress depends on soil water content, crop species, growth stages, etc. Timely replenishment of water after drought helps to restore the crop growth, while the drought-flood abrupt alternations cause the crop to experience flooding

after drought, and flooding weakens the respiration of crop roots. It causes weakening or loss of leaf stomatal conductance of the crop, which is not conducive to healthy growth (Gil et al., 2007). Most drought-stricken fields can be irrigated in time to alleviate drought conditions. Still, it is challenging to control flooding in drought-flood abrupt alternations due to the uncertainty of farm infrastructure and rainfall intensity and duration. Furthermore, there is a lack of reliable prevention and control technology. In most areas of China, maize seedlings are susceptible to uncertain drought and flood stresses. For effective disaster prevention and mitigation, in this study, we aimed to answer two key questions: (1) Does drought in maize seedlings improve resilience to drought-flood abrupt alternations? (2) What are the limits to which maize can withstand the drought-flood abrupt alternations? The results of this study are intended to provide potential solutions for disaster mitigation and prevention in response to the drought-flood abrupt alternations.

Materials and Methods

Experimental design

The experiment was conducted in a temperature-controlled greenhouse with a photosynthetic photon flux density of 400~600 $\mu\text{mol m}^{-2}\cdot\text{s}^{-1}$, 14 h photoperiod, and day and night temperature of 19/24°C. Maize variety sweet corn F1 Early Sunglow, commercialized seeds acquired from the lab in the Environment Centre, Lancaster University, were planted in black cylinders made of polyvinyl chloride (PVC) plastic, with an outer diameter of 6.6 cm, a thickness of 2 mm, and a height of 24.5 cm. The bottom of each cylinder was sealed with a stainless-steel net to facilitate drainage and stabilize the cylinder. The cultivated organic loam John Innes No. 2 was used as experimental soil. This soil type is traditional British soil compost containing loam, sphagnum peat, grit, humus, and fertilizers that promote the healthy growth of plants. Owing to its good performance, John Innes No. 2 has been widely used in the UK for more than 60 years. Each cylinder was filled with 1050 g soil, which was divided into four equal parts and compressed in the cylinder orderly, the bulk density was determined as 1.55 g cm^{-3} . Two maize seeds were sown in each cylinder and watered with 50 mL of water. The water amount was determined with a measuring cup. As evaporation from topsoil cannot be measured precisely, in order to reduce soil evaporation and accurately determine the daily transpiration of maize plant in each pot, the pots were covered with tinfoil in stage I, and wrapped with plastic film to prevent water leaching and evaporation in stage II, respectively (Fig. 1a).

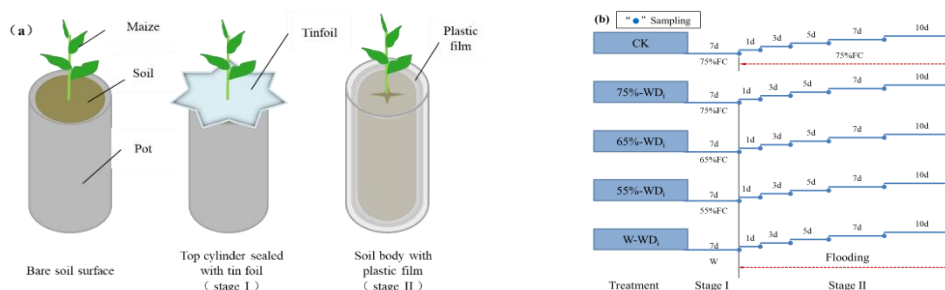


Figure 1. Schematic diagram of (a) covering pots and (b) experimental design. In stage I, 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity), and W-WD means the waterlogging-alone treatment. In stage II, WD_i represents the five durations of flooding. CK means the treatment of water control at 75% of the field capacity

Based on the weight of dry soil in each pot and the field capacity, soil moisture was determined with daily changes in pot weight, which was weighed with an electronic balance at 8:00 a.m. Soil water was controlled from the three-leaf stage of maize, and the experiment was divided into two stages, i.e., Stage I (7 days) and Stage II (10 days). The soil water content was kept at 55%, 65%, and 75% of the field capacity in stage I. In the waterlogging-alone (W-WD) treatment, a water layer of 1 cm was maintained on the soil surface. In stage II, the treatments at stage I were subjected to waterlogging for 1, 3, 5, 7, and 10 days (55%-WD_i, 65%-WD_i, and 75%-WD_i, where *i* is the duration of the plant subjected to waterlogging, *Table 1* and *Fig. 1b*). The treatment of water control at 75% of the field capacity (sufficient soil moisture for maize growth) was used as CK. There were 17 treatments in total, each repeated five times.

Table 1. Experimental design for maize under drought-flood abrupt alternations

Treatment	Treatment count	Stage I (7 days)	Stage II (≤10 days)
CK	1	75% FC	75% FC (10 days)
75%-WD _i	5	75% FC	Waterlogging, <i>i</i> = 1, 3, 5, 7, 10
65%-WD _i	5	65% FC	Waterlogging, <i>i</i> = 1, 3, 5, 7, 10
55%-WD _i	5	55% FC	Waterlogging, <i>i</i> = 1, 3, 5, 7, 10
W-WD _i	1	Waterlogging	Waterlogging, <i>i</i> = 1, 3, 5, 7, 10

Note: FC is the field capacity

Measurement set-up

During the experiment, the top of each cylinder was sealed with tin foil to expose only the maize plants. The total mass of the cylinders was weighed every morning from 8:00 to 9:00 to calculate the daily water loss owing to plant transpiration. The lost water was then supplemented. Maize seedlings were harvested at the end of stage II. Aboveground parts of the seedlings were cut and separated into stem and leaf in each pot. The soil in each pot with the root system was divided into 0~5 cm, 5~10 cm, 10~15 cm, and 15~20 cm from the soil surface. The roots in each segment were then washed. The leaf area was measured by an automatic leaf area meter (Licor Model 3100 Area Meter, Cambridge, UK). Stem, leaf, and root samples were dried at 65°C for 72 h, and their dry matter was then determined.

ABA concentration in leaves and roots was measured by radioimmunoassay using the monoclonal antibody AFRC MAC 252 (Quarrie et al., 1988). The youngest and fully expanded leaflet was harvested for ABA measurement. The sampling was conducted simultaneously (10:00~10:30) on each harvesting day to avoid diurnal effects on foliar ABA concentration. Leaflets and roots (of the same plant) were sampled, frozen in liquid nitrogen, freeze-dried for 48 h, and then finely ground. Small amounts of samples (10~15 mg dry weight for leaflets, and 30~40 mg dry weight for roots) were used for ABA analysis. The powdered samples were diluted with deionized, distilled water (1:70 for leaflets and 1:25 for roots) and placed on a shaker in a cold room (4°C) overnight to extract ABA. A standard curve was determined with standards in a serial dilution of synthetic unlabeled (±)-cis, trans-ABA (Sigma Let., Dorset, UK). ABA concentration was calculated by reference to the standard curve after linearization using the ‘logit’ transformation.

Statistical analysis

All data were analyzed by one-way analysis of variance (ANOVA). A less significant difference (LSD) and Duncan test were carried out ($P < 0.05$) using Statistica 10.0 (StatSoft, Inc., Tulsa, OK, USA).

Results

Changes in leaf area and biomass

Leaf area is directly related to crop respiration, photosynthesis, and biomass formation, can markedly affect crop yield improvement. The changes in leaf area at the end of the two stages are presented in Fig. 2. At the end of stage I, there was no significant difference in maize leaf area between 65%-WD and 75%-WD, while 55%-WD and W-WD exhibited significantly lower leaf area, which was 24.66% and 75.68% lower than that of 75%-WD, respectively (Fig. 2). At the end of stage II, compared with CK, the leaf area of 65%-WD₁₀ was slightly lower, but the difference was not significant, while the leaf areas of 75%-WD₁₀, 55%-WD₁₀, and W-WD₁₀ were reduced by 14.23%, 32.34%, and 49.38%, respectively, which was significantly different.

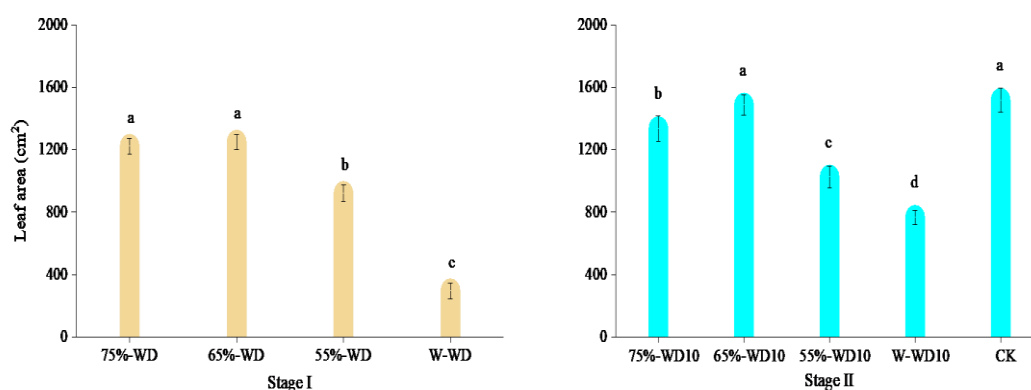


Figure 2. Effects of drought and flood stress on leaf area of maize seedlings. Different letters indicate significant difference between the treatments at $P < 0.05$. In stage I, 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity), and W-WD means the waterlogging-alone treatment. In stage II, WD_i represents the five durations of flooding. CK means the treatment of water control at 75% of the field capacity

Fig. 3 depicts the changes in biomass of each part of maize seedlings at the end of stage I. The leaf biomass was significantly higher in the 75%-WD and 65%-WD treatments than that in the 55%-WD and W-WD treatments, and the leaf, stem, and root biomass was severely suppressed in W-WD. The different drought treatments insignificantly affected the stem biomass, and the 55%-WD treatment exhibited the most developed root system, with significantly higher biomass and lower shoot-root ratio than the other treatments. In contrast, the W-WD treatment exhibited a lower shoot to root ratio than the drought treatments (Fig. 3). The results in Fig. 3 indicated that different degrees of drought exposure at the maize seedling stage affected the biomass. The greater the drought intensity is, the more developed the root system and aboveground inhibition are. However, drought-affected maize seedlings to a weaker extent than flood stress.

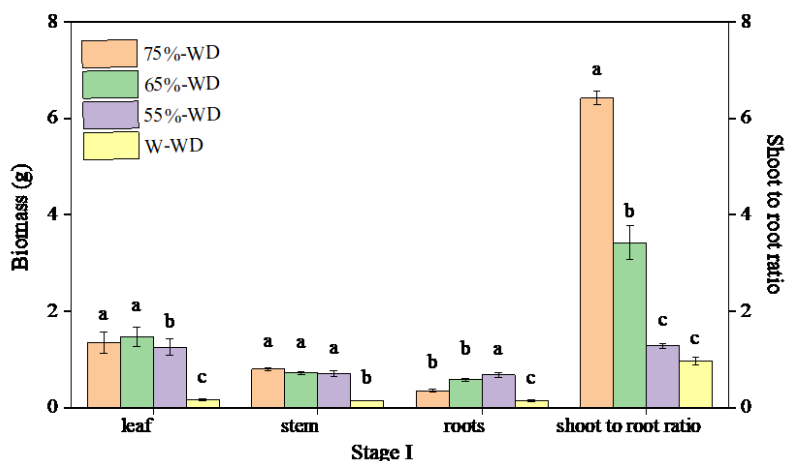


Figure 3. Biomass of leaf, stem, roots, and the shoot to root ratio of maize seedlings at the end of stage I. Different letters indicate significant difference between the treatments at $P < 0.05$. 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity), and W-WD means the waterlogging-alone treatment

The most direct effect of stress on plants is the inhibition of crop growth. In the present study, the drought-flood abrupt alternations affected maize biomass at the seedling stage (Fig. 4a-d). Differences in the biomass of maize organs were already evident at 3 d of stage II, where the biomass of different parts of maize seedlings under 55%-WD₃ and 65%-WD₃ reached the control level. In contrast, the leaf biomass under 55%-WD₃ exceeded the control by 10.72% (Fig. 4a). Under the 75%-WD₃ and W-WD₃ treatments, the biomass of leaves and roots was significantly lower than that of CK (Fig. 4a and c). At 5 d of stage II, the biomass of leaves, stems, and roots under 65%-WD₅ maintained rapid growth and reached the control level, while the biomass under other treatments exhibited significant inhibition and the shoot to root ratio of the treatments did not differ significantly (Fig. 4a-c). At 7 d of stage II, the differences in maize organs biomass among treatments were similar with the results at 5 d of stage I. Compared with CK, the leaf biomass in 75%-WD₇, 55%-WD₇, and W-WD₇ decreased by 51.24%, 41.23%, and 82.44% (Fig. 4a), respectively; the stem biomass decreased by 42.27%, 25.80%, and 55.66% (Fig. 4b), respectively, which increased the degree of inhibition of biomass of each organ compared with CK at 3 to 5 d of stage II. With the increase in flood duration, the shoot-root ratio was continuously adjusted, and under the 65%-WD₁₀ treatment, it reached the control level (Fig. 4d). Furthermore, the degree of root inhibition was higher under other treatments than in aboveground organs (Fig. 4d). The results indicated that maize plants experienced varying degrees of drought in stage I. The aboveground parts of maize seedlings exhibited significant compensatory growth in stage II, thereby reflecting its biological adaptation to the drought-flood abrupt alternations. Therefore, when maize plants were subjected to drought-hardening with severe stress (55% of the field capacity), the subsequent waterlogging of no more than 3 days had insignificant influence on maize vegetative growth. Under the condition of drought-hardening with moderate stress (65% of the field capacity), the effects of subsequent waterlogging of no more than 7 days on maize vegetative growth were insignificant. However, the maize plants with sufficient water supply (75% of the field capacity) lost the resistance to water lodging, so the plant vegetative growth was limited significantly with water lodging at stage II.

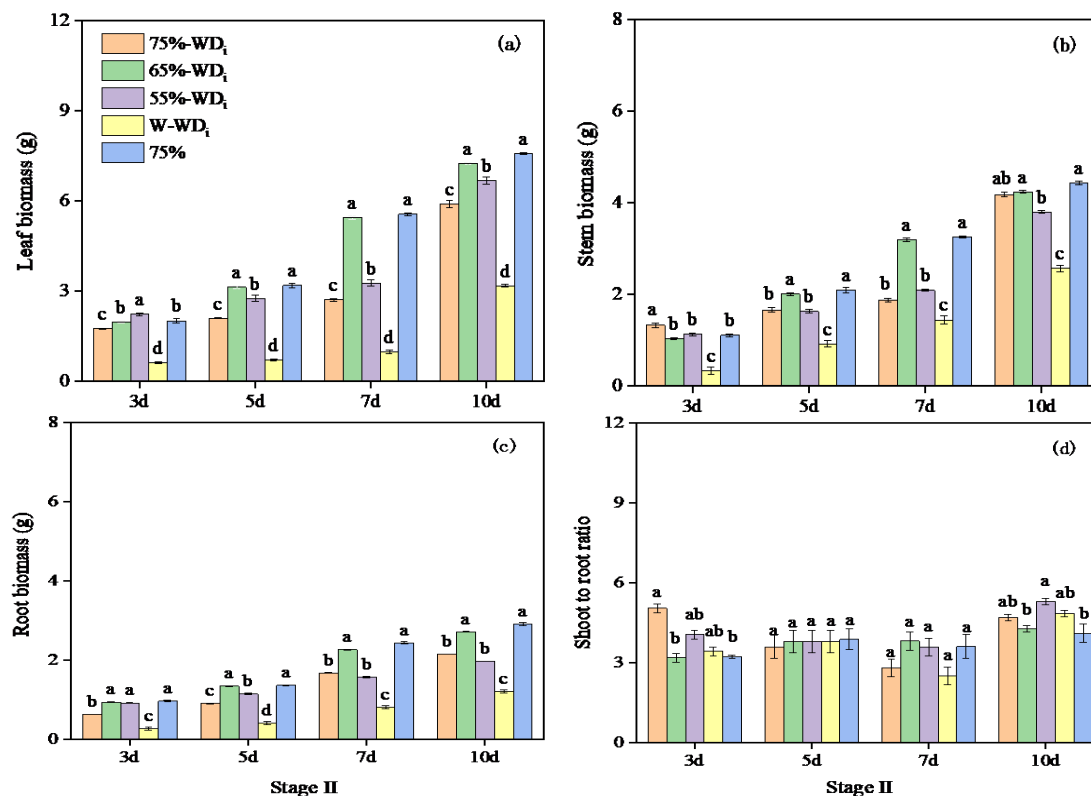


Figure 4. Biomass of (a) leaf, (b) stem, (c) roots, and (d) the shoot to root ratio of maize seedlings during stage II. Different letters indicate significant difference between the treatments at $P < 0.05$. 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity), and W-WD means the waterlogging-alone treatment. CK means the treatment of water control at 75% of the field capacity

Changes in ABA concentration

The changes in ABA concentration in maize leaves and roots at the end of stage I are presented in Fig. 5. ABA concentration in leaves was significantly higher than in roots under all treatments. ABA concentration in each treatment changed gradually from maize leaves to roots, and gradually decreased from maize leaves to roots. In stage I, the greater the degree of water stress is, the greater the ABA concentration in the leaves is, with the maximum ABA concentration in maize leaves of the 55%-WD treatment being significantly higher than the 65%-WD and 75%-WD treatments, and significantly higher than the W-WD treatment ($P < 0.05$). Compared to 75%-WD, leaf ABA concentration increased by 21.10% and 33.81% for 65%-WD and 55%-WD, respectively, and decreased by 24.65% for W-WD (Fig. 5), indicating that the leaf ABA concentration is highly sensitive to drought stress. At the end of stage I, the greater the degree of water stress is, the greater the ABA concentration in maize roots in 0-5 cm depth is. The root ABA concentration was significantly higher in 55%-WD than in 75%-WD and W-WD. Compared with 75%-WD, ABA concentration of roots in 0-5 cm depth was 12.05% and 28.75% higher in 65%-WD and 55%-WD, respectively, and 3.59% lower in W-WD. ABA concentration in the 5-20 cm depth remained around $70 \text{ ng g}^{-1} \text{ DW}$, with gradually decreasing differences between different treatments (Fig. 5).

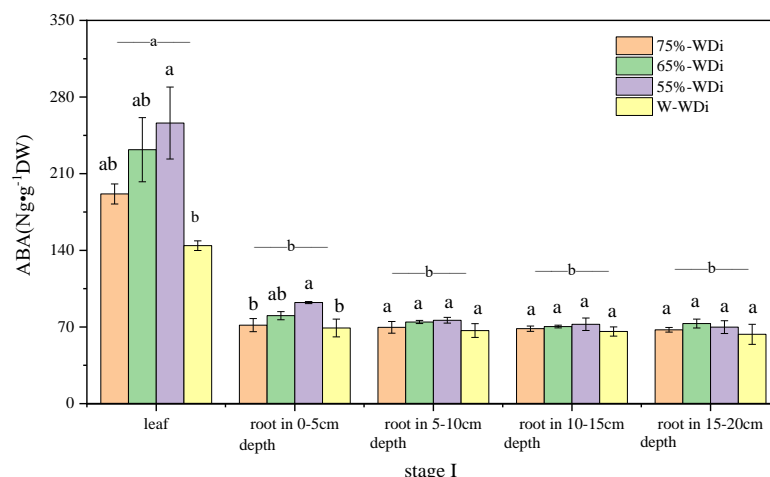


Figure 5. ABA (abscisic acid) concentration in leaf and root of maize seedlings at the end of stage I. Different letters indicate significant difference between the treatments at $P < 0.05$. 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity), and W-WD means the waterlogging-alone treatment. — a—and—b— indicates the difference in ABA between maize leaf and roots in different layers at $P < 0.05$

At 1 d of stage II, leaves under the 75%-WD₁, 65%-WD₁, and 55%-WD₁ treatments exhibited significantly higher ABA concentrations than those under the corresponding treatments in stage I. Under the 55%-WD₁ and 65%-WD₁ treatments, the mean ABA concentration in leaves reached up to 465.32 and 427.18 ng g⁻¹ DW, respectively, which was significantly higher than that in the treatment of W-WD₁ and CK (Fig. 6). At 3-7 d of stage II, leaf ABA concentration in 75%-WD₁, 65%-WD₁, and 55%-WD₁ remained around 200 ng g⁻¹ DW, which was significantly higher than that in W-WD₁ (Fig. 6).

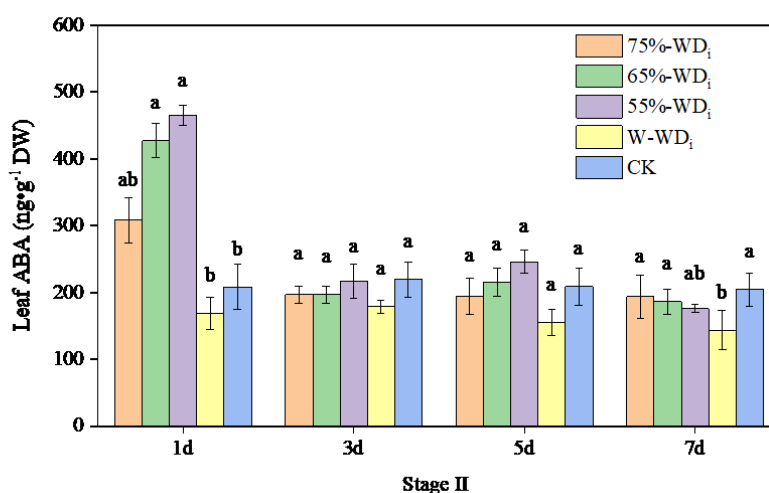


Figure 6. Variations of ABA concentration in maize leaves during stage II. Different letters indicate significant difference between the treatments at $P < 0.05$. 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity) in stage I, and W-WD means the waterlogging-alone treatment. CK means the treatment of water control at 75% of the field capacity

Fig. 7 presents the variation of ABA concentration in the root system of maize seedlings at different depths in stage II. Under the 75%-WD₁ treatment, the ABA concentration of the root system gradually decreased from topsoil to deeper soil and was higher than that of CK, with significant differences at the depth of 0-5 cm. Compared with the 75%-WD₁ treatment, the ABA concentration in all root layers decreased by 21.39-35.39% under the 75%-WD₃ treatment, which was significantly higher than that at 5-7 d of stage II, indicating that the maize leaves and roots still had a strong ABA signal after 1 d of the drought-flood abrupt alternations, and the strong ABA signal was abolished after 3 d of the drought-flood abrupt alternations (*Fig. 7a*).

Under the 65%-WD₁ treatment, the root ABA concentration decreased gradually from topsoil to deeper soil and was higher than that of CK, with significant differences at the depth of 0-5 cm. Compared with the 65%-WD₁ treatment, the ABA concentration of each root layer under the 65%-WD₃ treatment decreased by 27.80% to 47.88%, and after 3 d of the drought-flood abrupt alterations, the ABA concentration of the root system increased as the flood duration increased, and it increased by 6.96% to 65.01% at 3-5 d and 92.06% to 151.15% at 5-7 d. The ABA concentration of each root layer in the 65%-WD₇ treatment was significantly higher than that in CK (*Fig. 7b*).

Compared with the 55%-WD₁ treatment, the root ABA concentration under the 55%-WD₃ treatment decreased by 22.15% to 44.84%, but the difference between the root layers was not significant. During 3-7 d in stage II, the root ABA concentration gradually increased. Compared with the 55%-WD₅ treatment, the root ABA concentration in all layers under the 55%-WD₇ treatment increased by 5.73% to 30.30%, which was significantly higher than that of CK (except the depth of 15-20 cm) (*Fig. 7c*). However, ABA concentration in the root system was significantly lower under the 55%-WD₇ treatment compared with the 65%-WD₇ treatment.

The root ABA concentration under persistently flooding treatment of W-WD_i was relatively stable, with root ABA concentration being around 70 ng g⁻¹ DW at each sampling date (*Fig. 7d*), which was comparable to stage I.

Leaf stomatal and transpiration changes

Stomata is an important water channel for photosynthesis and the transpiration of crops. Stomatal characteristics under different drought-flood abrupt alternations affect crop yield and water use efficiency. The changes in leaf stomata and plant transpiration of maize seedlings under different drought and flood stresses are presented in *Fig. 8*. In stage I, the leaf stomatal conductance of maize seedlings in the 65%-WD, 55%-WD, and W-WD treatments was lower than the CK values. The more severe the drought stress is, the worse the stomatal conductance is. At 1 d of stage II, the stomatal conductance of maize seedlings in drought treatments exhibited an increasing trend, then the stomatal conductance in the 55%-WD_i and 75%-WD_i treatments were decreased by a fluctuating trend with the increase of flooding duration. However, the leaf stomatal conductance in 65%-WD_i increased closer to the value in CK at 3-5 d of stage II, and then decreased gradually. At 3-5 d of stage II, the leaf stomatal conductance in 65%-WD_i were greater than these in 55%-WD_i and 75%-WD_i. During the whole experimental period (stage I and II), CK had greater stomatal conductance than the drought treatments, drought-flood treatments, and the flood treatments. The treatment of W-WD_i had lower stomatal conductance than the drought treatments and the drought-flood treatments (*Fig. 8a*).

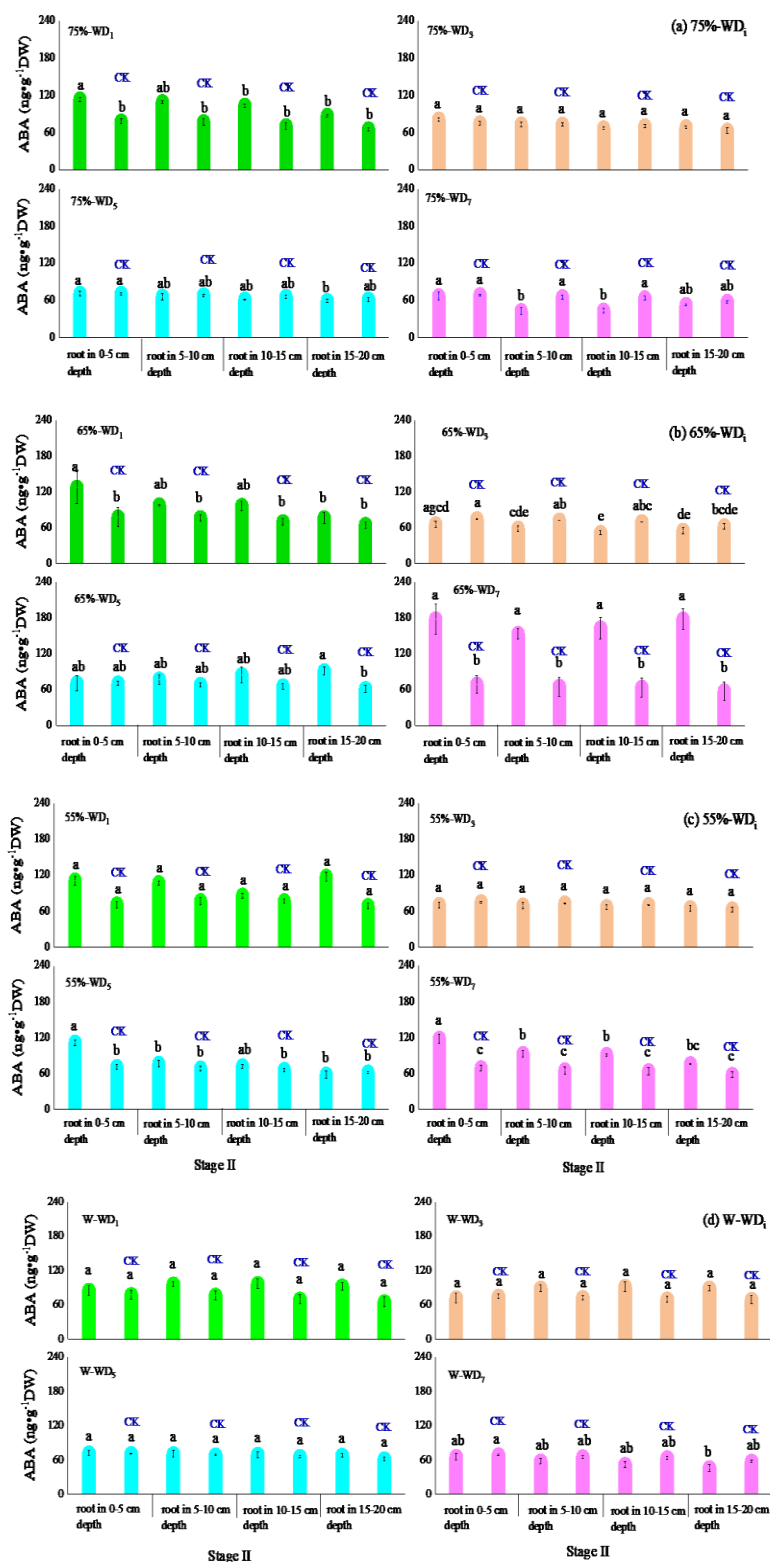


Figure 7. Variations of ABA concentration in maize roots of different treatments during stage II. Different letters indicate significant difference between the treatments at $P < 0.05$. 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity) in stage I, and W-WD means the waterlogging-alone treatment. CK means the treatment of water control at 75% of the field capacity

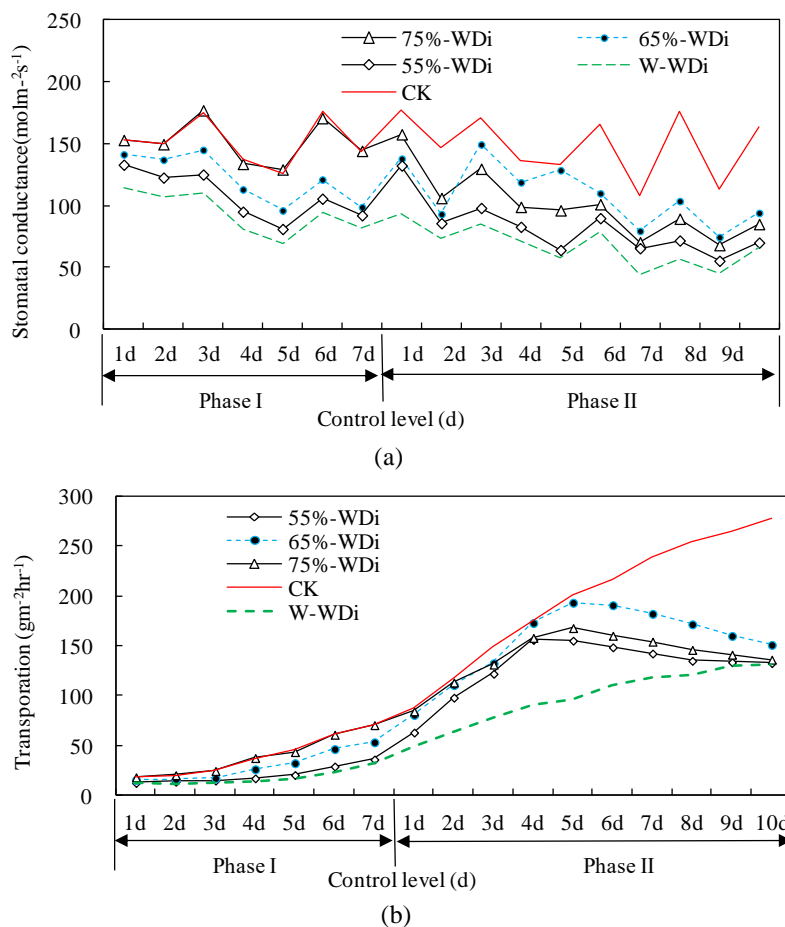


Figure 8. Changes in (a) stomatal conductance and (b) plant transpiration of maize seedlings during stage I and II. 75%-WD, 65%-WD, and 55%-WD represents the three levels of water deficit (WD, i.e., 75%, 65%, and 55% of the field capacity) in stage I, and W-WD means the waterlogging-alone treatment. CK means the treatment of water control at 75% of the field capacity

Daily transpiration in maize seedlings was related to soil moisture. In stage I, the higher the soil moisture, the higher is the daily transpiration, which in descending order was W-WD, 75%-WD, 65%-WD, and 55%-WD; the differences between different treatments gradually increased with crop growth and the duration of moisture control (Fig. 8b). The flood treatments restricted plant transpiration compared with the drought treatments (Fig. 8b). Plant transpiration in 75%-WD_i increased closer to the 65%-WD_i during 1-3 d of stage II, while lower than the 65%-WD_i at 3 d of stage II, and reached the maximum transpiration at 3 d of stage II. The maximum transpiration in 55%-WD_i and 65%-WD_i was measured at 3 d and 5 d of stage II, respectively. The daily transpiration in 55%-WD_i, 65%-WD_i, and 75%-WD_i decreased after the peak, and the difference between the two treatments and CK gradually increased. At 10 d of stage II, the daily transpiration was similar between 55%-WD_i, 75%-WD_i, and W-WD_i, and lower than the value in 65%-WD_i.

Discussion

Drought and flood stress are the leading natural disasters worldwide, and the most direct effect of stress on crops is the inhibition of crop growth and reduction of biomass (Wu et al., 2013; Zhen et al., 2018). Therefore, biomass is often essential for assessing crop resistance under abiotic stresses. In the present study, the leaf area of maize seedlings under 65%-WD₁₀ was not significantly reduced (Figs. 2-4), indicating that maize seedlings that experienced a suitable water stress exhibit better resistance to flood stress at the later stage. Under water stress, a series of metabolite changes will occur in crops, which can regulate the osmotic capacity of crop leaves, thus maintaining the extended growth of cells and their stomatal conduction and photosynthetic production capacity. Therefore, the maize seedlings in 65%-WD_i exhibited high stomatal conductance and transpiration after drought-flood abrupt alternations, and then maintained photosynthate accumulation, which plays an important role in the resistance to flood stress in the late stage. The response of crops to drought stress activates crop resilience function. At stage II, the 65%-WD_i measured high biomass, indicating the facilitating effects of drought hardening in the early stage on the flood resistance at the late stage. However, the strength of flood resistance at late stage was depended on the degree of drought stress at the early stage. Maize seedlings in 55%-WD_i suffered severe drought stress at stage I and had low water potential in the root zone, resulting in low stomatal conductance and limited photosynthate accumulation. At 1-4 d of stage II, the seedlings in 55%-WD_i exhibited a high rate of biomass accumulation and high stomatal conductance, showing the rapid growth of stress resistance. While this response only lasted for 4 days at stage II, indicating that the severe water stress at stage I was not conducive to the response of crops to waterlogging in the late stage. The less capacity of maize seedlings in response to waterlogging in 55%-WD_i than 65%-WD_i confirmed it. The seedlings in 75%-WD_i were supplied by sufficient water and exhibited a good base of biomass at stage I; after 3 d at stage II, however, leaf senescence was measured in 75%-WD_i, and the stomatal conductance and transpiration lower than the treatment of 65%-WD_i (Fig. 8), and the biomass accumulation rate decreased, lower than that in 65%-WD_i. The results indicated that the sufficient water supply at stage I was ineffective for improving crop resistance capacity after drought-flood abrupt alternations. At stage I, the biomass and leaf area of maize seedlings in W-WD_i were significantly restricted by stress. Under waterlogging stress, the stomatal conductance and photosynthetic capacity of crops decrease, and the transport of photosynthate also slows down. The root zone is hypoxic, and the stomatal resistance of CO₂ diffusion increases, which affects their photorespiration. The chlorophyll content decreases, and the leaves' premature senescence and abscission are obvious, limiting their ability to resist waterlogging adversity. Therefore, moderate drought exposure in the early stage is beneficial in alleviating or ameliorating the effects of flood on the biomass in the later stage, which promotes a good material basis for improving crop yield, thereby securing the economic returns (Zhen et al., 2017; Liang et al., 2019). The shoot-root ratio, a marker of the sensitivity of crops to stressful environments, is related to ABA synthesis and is considered as an adaptative mechanism to stress (Boursiac et al., 2013). In this study, the greater the degree of water stress in stage I, the greater the effects on the aboveground stem and leaf growth, and the more developed root system. After 10 d of stage II, the root inhibition resulted in differences in the shoot-root ratio among different treatments (Fig. 4d). Water stress at stage I promoted the root growth and expanded the root functional area, which laid a good physiological foundation for crop survival under late flood stress and exerted a buffering

and regulatory effect on the maintenance of the late root zone flooding damage. Therefore, the stress resistance of crops can be improved to some extent through soil moisture regulation.

As a phytohormone, ABA plays a significant role in regulating plant growth and development, especially in several physiological processes during abiotic stresses (high salt, low temperature, drought, etc.) (Dong et al., 2015; Li et al., 2017). It has been shown that a variety of abiotic stresses increase ABA concentrations in plants (Shinozaki et al., 2003; Boursiac et al., 2013). The root system is the first site to sense the soil moisture changes under water stress, and plant ABA concentrations are derived from the root system. Furthermore, soil water stress derives from the changes in this physiological characteristic (Puértolas et al., 2013). Therefore, changes in plant ABA concentrations are often adjusted or ameliorated through irrigation, leading to optimal production (Davies et al., 2011). In the present study, the sharp alternation to flooding from stage I to stage II caused a gradual change in ABA concentration in maize leaves and roots, which decreased from the leaves to roots, with the root system originating the drought stress signal, and this signal was heavily aggregated and transmitted upstream to the leaves along with the plant fluid flow, so that, the leaves are the end point of the heavily aggregated ABA signal and exhibit the highest ABA concentration. However, at 1 d of stage II, the leaf ABA concentrations of the 75%-WD₁, 65%-WD₁, and 55%-WD₁ treatments were significantly higher than those of the 75%-WD₁, 65%-WD₁, and 55%-WD₁ treatments at the end of stage I. The reason for this difference may be that the exposure of the crop to a new stress prompted the plant cells to actively regulate the osmotic substances in response to the stress (Wu et al., 2013). The increase in osmoregulatory capacity caused a significant increase in the leaf ABA signal at 1 d of stage 75%-WD₁, 65%-WD₁, and 55%-WD₁ treatments, which gradually attained a stable stage. Due to ABA degradation in the crop caused by the fold stress, the leaf ABA concentration in each treatment in this study dropped to a lower level at 7 d of stage II, but the leaf ABA concentration in each treatment subjected to drought stress in stage I at this time point was still significantly higher than that of the continuous flood stress treatment. It was suggested that the flood stress also leads to a significant increase in ABA concentration in the crop (Chen et al., 2006; Hiroaki et al., 2007). When the leaf stomatal conductance decreases, transpiration rate decreases, and ABA synthesis in roots increases. Furthermore, ABA accumulation increases in the roots under the influence of reduced transpiration pull. Therefore, the root ABA concentration in this study was significantly elevated at 7 d after the drought-flood abrupt alternations, indicating that the physiological resistance signal of maize is strongest at 7 d after the drought-flood abrupt alternations. Studies on other plants revealed that plant physiological functions were recovered to varying degrees at 15 d after rehydration (Song et al., 2012; Wang et al., 2015), and exhibited a compensatory effect on growth (Li et al., 2013; Wang et al., 2015). In this study, at stage I, the leaf ABA concentrations under 55%-WD and 65%-WD were significantly higher than that under 75%-WD, and it increased with an increase in water stress, and leaf stomatal conductance, and then, plant transpiration subsequently decreased. At the end of stage I, leaf area and biomass of the 65%-WD treatment group were not affected. During 1-5 d of stage II, the leaf-root ABA concentration difference increased with an increase in water stress. With regards to the leaf stomatal conductance, plant transpiration and biomass, the recovery ability of each marker under 55%-WD_i was weaker than that under 65%-WD_i. By 7 d of stage II, since the gradient of ABA concentration between leaves-roots became smaller, ABA concentration between leaves-

roots attained a more balanced state; the leaf stomatal conductance and plant transpiration slowly decreased under the 65%-WD₇ and 55%-WD₇ treatments. The rate of dry matter accumulation declined under 65%-WD₇ and was significantly decreased under 55%-WD₇. The results of this study revealed that in stage I, when the water was controlled at 55% of the field capacity, the rehydration effect on biomass occurred within 3 d after the drought-flood abrupt alternation. However, in stage I, when the water was controlled at 65% of the field capacity, the rehydration effect occurred 5 d after the drought-flood abrupt alternations, compensatory growth was observed in all markers, which was similar to the control level. Considering the restoration effect of the crop itself and the relief duration for disaster mitigation and prevention, 65% of the field capacity at the maize seedling stage can be considered as a more suitable water control measure, and the duration of flooding should not exceed 7 d after the drought-flood abrupt alternation.

Conclusions

Maize seedlings undergo a certain degree of water stress (55%-WD_i and 65%-WD_i) and compensatory crop biomass growth occurs after the drought-flood abrupt alternations. Under the condition of drought and flood, ABA concentration in maize leaves was significantly higher than that in roots. The most sensitive site in maize to drought stress was found to be the leaf. After the drought-flood abrupt alternations, there was a strong ABA signal in the roots under 55%-WD₅ and 65%-WD₇. At maize seedling stage, the water level is controlled at 65% of the field capacity, and the duration of flooding should not exceed 7 d after the drought-flood abrupt alternations. Analyses of metabolomics, proteomics, etc. should be carried out in future to further reveal effects of the drought-flood abrupt alternations on plant physiological-biochemical characteristics.

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Data Availability Statement. The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

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