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Effects of Waterlogging at Flowering Stage on the Grain Yield and Starch Quality of Waxy Maize

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Effects of Waterlogging at Flowering Stage on the Grain Yield and Starch Quality of Waxy Maize

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Abstract: Waterlogging is a common abiotic stress in global maize production. Maize flowering stage (from tasseling to silking) is more fragile to environmental stresses among different growth stages, and this stage frequently overlapped the plum rain season in the middle and lower reaches of Yangtze river in China and affect the yield and quality of spring-sown maize severely. The effects of waterlogging at flowering stage on the grain yield and starch quality of waxy maize were studied using Suyunuo5 (SYN5) and Yunuo7 (YN7). The grain yield was reduced by 14.7% for YN7 and 29.1% for SYN5 with waterlogging due to the decreased grain weight and numbers. The grain starch content in YN7 was decreased by 9.4% when plants suffered waterlogging at the flowering stage, whereas the content in SYN5 was only decreased in 2014 and unaffected in 2015. The size of starch granules and proportion of small-molecule amylopectin with waterlogging at flowering stage increased in SYN5 and decreased in YN7. The starch crystalline structure was not changed by waterlogging, whereas the relative crystallinity was reduced in SYN5 and increased in YN7. The pasting viscosities were decreased, and pasting temperature was unaffected by waterlogging in general. The gelatinization enthalpy was unaffected by waterlogging in both hybrids in both years, whereas the retrogradation enthalpy and percentage in both hybrids were reduced in 2014 and unaffected by waterlogging in 2015. Our study indicated that waterlogging at the flowering stage reduced the grain yield, restricted starch accumulation, and deteriorated the pasting viscosity of waxy maize. Between the two hybrids, YN7 and high pasting viscosities and low retrogradation percentage than SYN5, indicated its advantages on produce starch for viscous and less retrograde food. Results provide information for utilization of waxy maize grain in food production.

Keywords: waterlogging; waxy maize; grain yield; starch granule size; pasting viscosity

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1. Introduction

In the current climate change scenario, agriculture faces increasing instability with extreme weather events, leading to considerable yield losses. Waterlogging is a prevalent environmental adversity that suppresses maize productivity and degrades grain quality; it is predicted to increase in magnitude and frequency along with global warming [1]. In South and Southeast Asia alone, 18% of total maize growing area is frequently affected by waterlogging conditions, which constitute 25%–30% of annual production losses [2]. Maize is a rainfed crop and considered vulnerable to waterlogging when the field soil moisture content is higher than 80%, thereby affecting plant growth and development [3]. Among different growth stages, the flowering stage (from tasseling to silking) is sensitive to environmental stresses. Waterlogging around the flowering stage induces the increase in lodging risk by decreasing the stem diameter, rind penetration strength, and transverse bending strength of the third base internode [3]. Waterlogging also decreases the chlorophyll and carbohydrate contents [4] and the nitrogen content in different organs at silking and maturity stages [5]. Waterlogging also disturbs carbon-nitrogen metabolism, breaks plant endogenous hormone balance, accelerates leaf senescence, eventually resulting in a significant reduction in photosynthetic capacity and maize grain yield [6–8].

Waterlogging also affects grain starch formation, deposition, and structural and functional properties. Waterlogging starting at the anthesis stage decreases the amyloplast number in the wheat endosperm, and advances the programmed cell death in endosperm cells [9]. Post-anthesis waterlogging suppresses the activities of ADP glucose pyrophosphorylase and soluble starch synthase in wheat grains, increases the starch granule size, and reduces the viscosity parameters [10,11]. Meanwhile, waterlogging during grain filling all decrease the grain weight and affect the milling quality, although changes in protein composition may increase or maintain the gluten strength.[12] However, short duration of waterlogging and shading decreases the size of starch granules and increases the peak viscosity [13]. In rice, the structural and pasting properties of starch are affected by flooding irrigation, but the influence varies among cultivars [14]. In comparison with alternate wetting and drying, constant-flooding irrigation decreases the peak and breakdown viscosities, gelatinization temperature, setback viscosity, and gelatinization enthalpy; however, the effect on starch granule size and amylopectin chain length distribution has different trends [15]. In peanut, the oil, unsaturated fatty acids, and starch contents in grain increased, while crude protein, soluble sugar, saturated fatty acids, essential amino acids, non-essential amino acids, and total amino acids contents decreased under waterlogging. [16] Waxy maize is a special maize type with starch is composed of nearly pure amylopectin, which endows high viscosity, low retrograde, and high stability than normal maize starch [17]. Our previous study reported that waterlogging during grain filling stage has significantly affect the starch physicochemical properties [18,19]. However, limited information is available regarding the starch deposition and functional properties of waxy maize that suffered waterlogging at flowering stage. We hypothesize that waterlogging at the flowering stage affects starch accumulation and changes the starch structure, thereby affecting the starch physicochemical properties, such as the pasting and thermal properties of waxy maize. Results could provide a fundamental basis for using stressed starch based on the utilization of waxy maize.

2. Materials and methods

2.1. Experimental design

A pot trial was conducted on the Experimental Farm in Agricultural College of Yangzhou University, China in 2014–2015. Seeds were sown on March 15 in both years. The two seedlings at one-leaf stage were transplanted to pots (38 cm height and 43 cm diameter), and one seedling at the four-leaf stage was left. The plants were supplied with 10 g of compound fertilizer ($N:P_2O_5:K_2O$=15%:15%:15%) at transplanting time and 6 g of urea (N=46%) at jointing stage[18]. The weeds in the pot were manually removed. The mean temperature, rainfall, and sunlight durations during plant growth in 2014 and 2015 were 21.73 and 21.00 °C, 426 and 730 mm, and 599 and 500 h, respectively.

Soil moisture content was controlled by a negative-pressure water supply and controlling pot device (Chinese Patent 200510123976) by setting the water supply tension of the device at different values [20]. Before the tasseling, the soil relative moisture content was set at 75%. At the flowering stage (from tasseling to silking), the soil relative moisture content for control (CK) and waterlogging (WS) was set at 80% and over 100% (10 mm water level aboveground). Stress was terminated after the ears were manually pollinated. During treatment, the plants were covered with a transparent canopy that was 5 m high aboveground to avoid the influence of rainfall. After treatment, the soil relative moisture content was reset to approximately 75% until maturity.

2.2. Grain yield

The grains were harvested at the maturity (about 40 days after pollination), and grain number per ear was counted. The grains were manually stripped from the cobs, and grain weight (mg) and grain yield (g/plant) were determined after sun drying.

2.3. Starch content

Starch content in grains was determined with anthrone–sulfuric acid method [21].

2.4. Starch isolation

The grains were steeped in 1 g/L $NaHSO_3$ solution at room temperature for 2 days. The starches were isolated using the method described by Lu et al. [20]. The samples were rinsed with distilled water, and then ground using a blender for 2.5 min. The suspensions were passed through a 100-mesh sieve. The residues on the screen were again homogenized for 1.5 min and then passed through the same sieve. The starch–protein slurry was collected in a 1000mL wide-neck flask and allowed to stand for 4 h. The supernatant was suctioned, and the settled starch layer was collected in 50 mL centrifuge tubes and centrifuged at 3000 ×*g* for 10 min. The upper non-white layer was scooped. The white layer was resuspended in distilled water and stirred for 30 min before centrifugation. The isolation procedures were repeated thrice. The starch was then collected and dried in an oven at 40°C for 48 h. The protein and ash contents in the isolated starch were determined by using the International Methods 46-10.01 and 08-17.01 of AACC. These contents were lower than 0.3% and 0.2%, indicating that the purity of starch was up to the Chinese National Standard (GB/T 8885-2017).

2.5. Starch granule size

The average starch granule size (μm) was expressed in terms of the volume of equivalent spheres. The size distributions of starch granules were estimated with a laser diffraction particle size analyzer (Mastersizer2000, Malvern, Worcestershire, England) following a procedure described in the study of Lu et al. [20]. The disperse phase was absolute ethyl alcohol. Instrument accuracy was verified by using Malvern standard glass particles. The instrument, which follows the principle of laser diffraction, can measure sizes of 0.1 and 2000 μm.

2.6. Starch molecular weight

For isoamylase debranched starch granules, starch (5 mg) was dissolved in 5 mL of distilled deionized water in a boiling water bath for 60 min. Sodium azide solution (10 μL 2%w/v), acetate buffer (50 μL, 0.6 M, pH 4.4), and isoamylase (10 μL, 1400 U, EC 3.2.1.68, Sigma) were added to the starch dispersion. The mixture was incubated in a water bath at 37 °C for 24 h. The hydroxyl groups of the debranched glucans were reduced by treatment with 0.5% (w/v) of sodium borohydride under alkaline conditions for 20 h. The preparation about 600 μL was dried in vacuo at room temperature and allowed to dissolve in 20 μL of 1 M NaOH for 60 min. The solution was diluted with 580 μL of distilled water.

Molecular weight distribution was analyzed using a PL–GPC 220 high-temperature chromatograph (Agilent Technologies UK Limited; Shropshire, UK) with three columns (PL110–6100, 6300, and 6525) and a differential refractive index detector [22,23]. The eluent system used dimethyl sulfoxide containing 0.5 mM $NaNO_3$ at a flow rate of 0.8 mL/min. The temperature of the column oven was controlled at 80 °C.

2.7. X-ray diffraction

X-ray diffraction patterns of starch were obtained with an X-ray diffractometer (D8 Advance, Bruker–AXS, Germany) operated at 200 mA and 40 kV. The scanning region of diffraction angle (2θ) ranged from 3° to 40° at a step size of 0.04° with a count time of 0.6 s. Relative crystallinity (RC, %) was calculated as the percentage of the sum of total crystalline peak areas to total diffractograms.

2.8. Pasting property

The pasting properties of starch (1.96 g of starch added in 26.04 g of water, total weight of 28 g; 7% *db*, w/w) were estimated using a rapid viscosity analyzer (RVA, Model 3D; Newport Scientific, Warriewood NSW, Australia) following the method of Lu and Lu[24]. A sample suspension was equilibrated at 50 °C for 1 min, heated to 95 °C at 12 °C/min, maintained at 95 °C for 2.5 min, cooled to 50 °C at 12 °C/min, and maintained at 50°C for 1 min. The paddle speed was set at 960 rpm for the first 10 s and then decreased to 160 rpm for the rest of the analysis.

 doi:10.20944/preprints202311.0320.v1

2.9. Thermal property

The gelatinization properties of starch were estimated by differential scanning calorimetry (DSC, Model 200 *F*3 Maia, NETZSCH, Germany) following the method of Lu and Lu [24]. Each sample (5 mg, dry weight) was loaded into an aluminum pan (25/40 microliters, D = 5 mm) and distilled water was added to achieve a starch-water suspension containing 66.7% water. Samples were hermetically sealed and allowed to stand for 24 h at 4 °C before heating in the DSC. The DSC analyzer was calibrated using an empty aluminum pan as a reference. Sample pans were heated at a rate of 10 °C/min from 20 to 100 °C. Thermal transitions of starch were defined as onset temperature (T_o), peak gelatinization temperature (T_p), conclusion temperature (T_c), and gelatinization enthalpy (ΔH_{gel}). Samples were stored at 4 °C for 7 days after thermal analysis for retrogradation investigations. Retrogradation enthalpy (ΔH_{ret}) was automatically calculated and retrogradation percentage (%R) was computed as $\%R = 100 \times \Delta H_{ret} / \Delta H_{gel}$.

2.10. Statistical design

Data presented in tables and figures are the mean of three repetitions. Analysis was performed using ANOVA and Duncan's test at a significance level of $p < 0.05$ with the data processing system (version 7.05).

3. Results and discussion

3.1. Grain yield

The grain weight was significantly reduced by waterlogging in both hybrids in both years, with decrease was 8.4% and 23.5% for YN7 and SYN5, respectively (Figure 1). The grain number was also reduced by waterlogging in both hybrids in both years (6.7% for YN7 and 7.5% for SYN5), and the decrease was significant for SYN5 in 2015 and for YN7 in 2014.The reduced grain weight and number induced the yield loss in both years, and the decrease was 14.7% for YN7 and 29.1% for SYN5, indicated that YN7 was more tolerant to waterlogging than that of SYN5. Yield penalty due to waterlogging at the flowering stage has been widely reported in maize and is mainly related to the decreased grain number and weight. These yield penalties may related to the decreases in dry matter accumulation, redistribution of stored photosynthate to the grain, and the conversion capacity from carbohydrate to starch in grain [25]. This results were caused by decreased the activities of ribulose bisphosphate carboxylase and phosphoenolpyruvate carboxylase, decreased the contents of zeatin riboside, indole-3-acetic acid and gibberellic acid but increased abscisic acid in leaf, and reduced photosynthetic rate by disordered the leaf gas exchange parameters and chlorophyll fluorescence parameters [6–8], which resulted in the decreased antioxidative enzyme activities, accelerating leaf senescence, and ultimately leading to decreased biomass accumulation [25]. The yield for YN7 under different treatments was higher than that for SYN5, this genotypic difference was also reported in various studies [6].

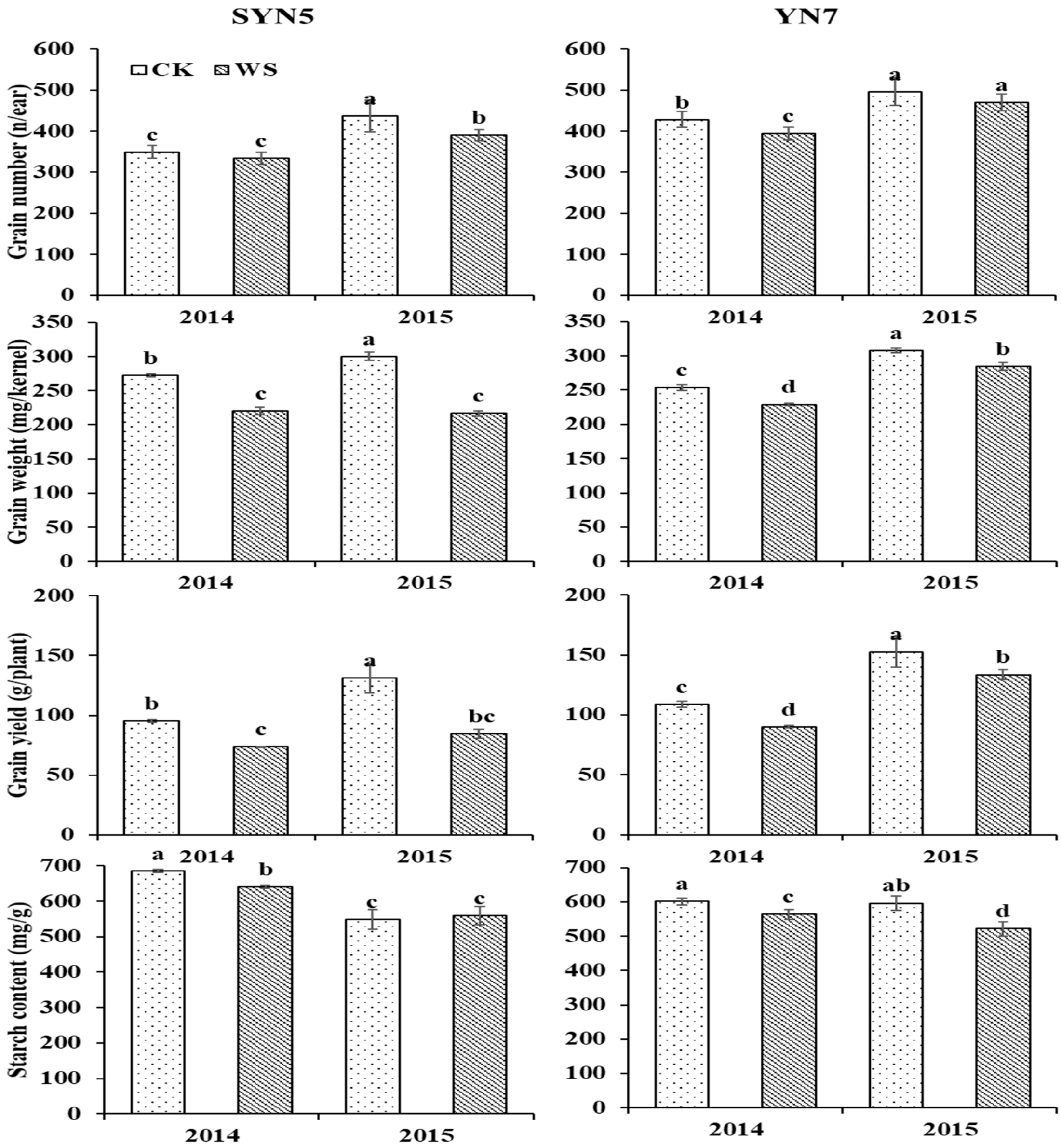

Figure 1. Effects of waterlogging at flowering stage on grain yield and starch content of waxy maize. Mean value in the same column within each hybrid followed by different letters is significantly different ($P < 0.05$). SYN5, Suyunuo5; YN7, Yunuo7; CK, control; WS, waterlogging.

3.2. Starch content

The starch content in YN7 was decreased by waterlogging (6.3% and 12.5% in 2014 and 2015) in both years, and the value in SYN5 was unaffected in 2015 and decreased by 6.7% in 2014 (Figure 1). The unaffected starch content in SYN5 in 2015 may due to the severe yield decrease in this year, and the shrunken sink make the surviving grains received similar source. Our previous study on fresh waxy maize observed that the starch content was increased due to the accelerated grain filling and shortened grain filling duration [26]. Zhou et al. [11] observed that the post-anthesis waterlogging reduced the wheat grain amylose content but did not affect the amylopectin content. The reduced starch content maybe caused by the weakened activities of sucrose synthase and soluble starch synthase [25], and decreased the allocation of nitrogen and carbon assimilates at pre- and post-anthesis to the grains [27]. A study in peanut also reported that the decreased starch content under waterlogging at the flowering stage was mainly caused by the decreased activities of sucrose synthetase and sucrose phosphate synthetase [28].

 doi:10.20944/preprints202311.0320.v1

3.3. Starch granule size

The size distributions of starch granules all presented dual peaks (Figure 2). The average starch granule size in response to waterlogging differed between the two hybrids; that is, it increased in SYN5 and decreased in YN7 in both years when the plants suffered from waterlogging. A study on rice also observed that the starch granule size in response to constant flooding irrigation management differed between the two cultivars [15]. Waterlogging during wheat heading and anthesis causes damage to endosperm cell structure, decreased the starch granules numbers in endosperms, resulting in the formation of irregular starch granules and increased the starch granule size [11]. Another study reported that short-time waterlogging and shading decreased the starch granules size [13]. Our previous study reported that the starch granule size of the two hybrids was decreased by post-silking waterlogging [18]. In the present study, the discrepancy of the results between the two hybrids may due to the different features of their development; the amyloplast in the endosperm in SYN5 was formed late and less but developed quickly, whereas the starch granules in YN7 formed more number in endosperm cells [29]. Therefore, amyloplast formation in endosperm cells needs further study to clarify the discrepancy on different hybrids.

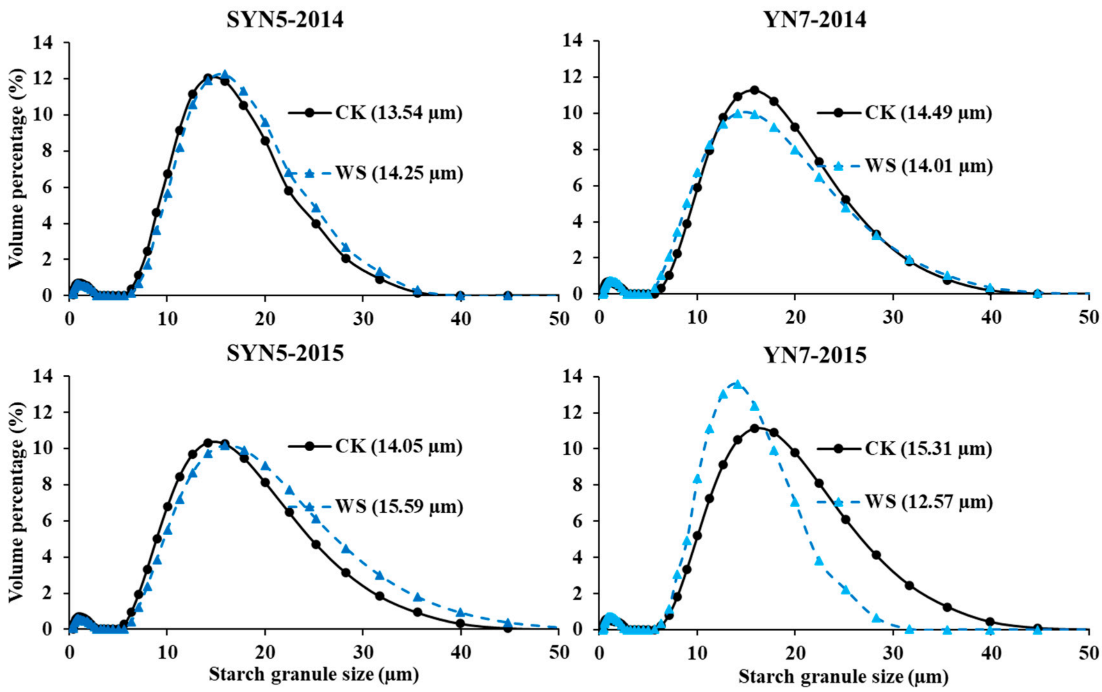

Figure 2. Effects of waterlogging at flowering stage on size distribution of waxy maize starch granules. SYN5, Suyunuo5; YN7, Yunuo7; CK, control; WS, waterlogging. Value in the bracket is the average granule size.

3.4. Starch molecular weight distribution

The molecular weight distribution of isoamylase debranched starch for all the samples presented dual peaks, namely, peak1 and peak2 (Figure 3), consistent with those of different waxy starch resources, indicating the typical waxy starch character [30]. In the GPC profiles of amylopectin, the peak1 fraction contained short starch chains, such as A and B chains (A + B1 chains) and the peak2 fraction consisted of long B chains with high-molecular-weight molecules [31]. The peak1/peak2 value with waterlogging increased in SYN5 but decreased in YN7 in both hybrids, consistent with the change trends of starch granule size; this finding indicated that starch with large granule size has high proportion of small molecular size of amylopectin branch chains [30]. A study on rice observed that the amylopectin chain length in response to constant-flooding irrigation management differed between the two cultivars [15].

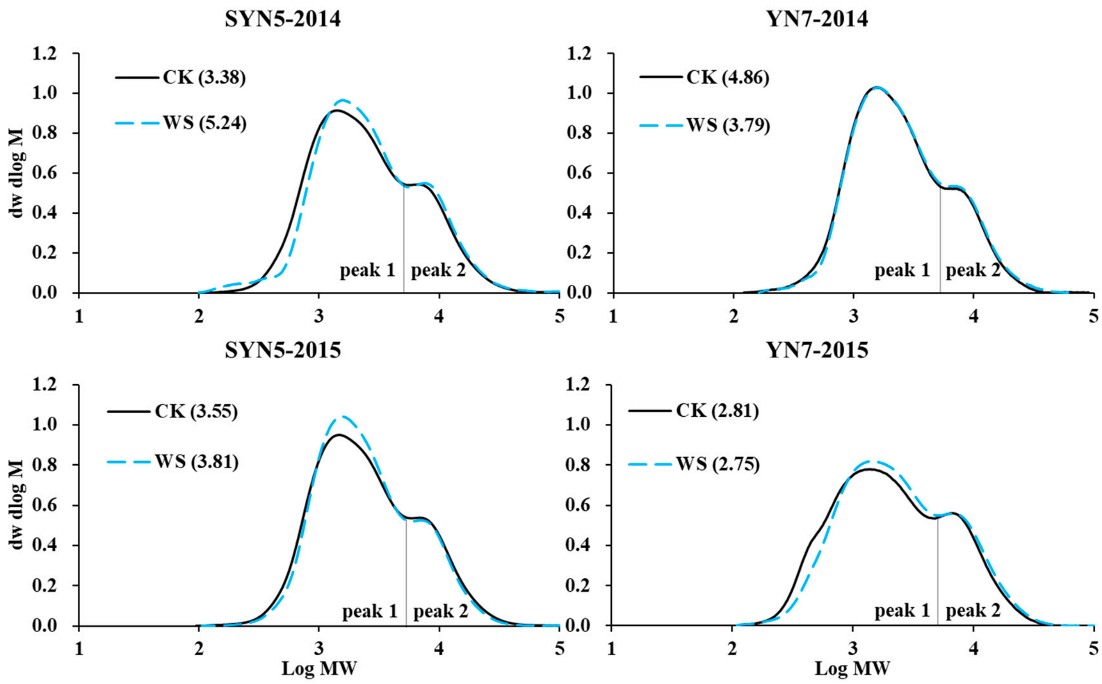

Figure 3. Effects of waterlogging at flowering stage on molecular weight distribution of isoamylase debranched starch in waxy maize. SYN5, Suyunuo5; YN7, Yunuo7; CK, control; WS, waterlogging. Value in the bracket is the ratio of peak1 to peak2.

3.5. Starch X-ray diffraction

The X–ray diffraction (XRD) of starch provided information on the long–range molecular order and was associated with ordered arrays of double helices formed by the amylopectin side chains [32]. All the samples presented refection angles at 15°, 23°, 17°, and 18°, which present a typical A diffraction pattern (Figure 4). The relative crystallinity (RC) with waterlogging decreased in SYN5 but increased in YN7 in both years. The different responses of RC to flooding irrigation was also reported in rice [14]. In the present study, the change trend of RC was contrary to the trend of starch granule size and peak1/peak2 ratio, indicated the starch with large granule size and high molecular-weight has low RC. Our previous study also observed that waxy maize starch with high proportion of medium-sized starch granules has high RC [33]. Hsieh et al. [30] reported that waxy starch with small granule size and high proportion of peak1 has high RC.

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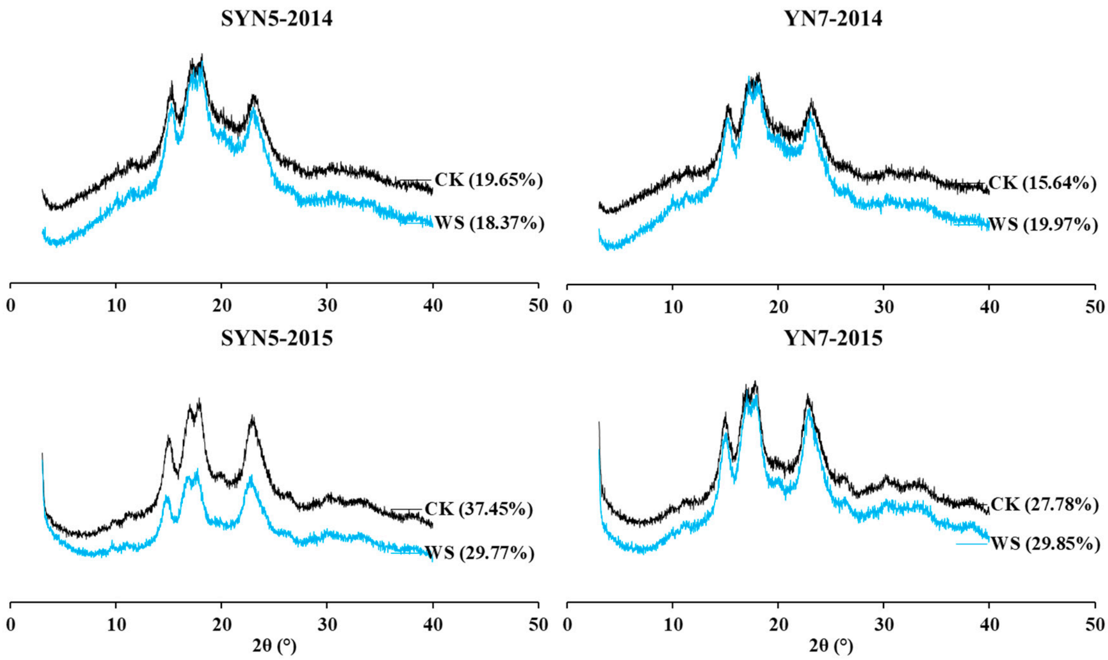

Figure 4. Effects of waterlogging at flowering stage on X-ray diffraction pattern of waxy maize starch. SYN5, Suyunuo5; YN7, Yunuo7; CK, control; WS, waterlogging. Value in the bracket is the relative crystallinity.

3.6. Pasting property

The grain flour peak (PV), trough (TV), final (FV), and setback (SB) viscosities in SYN5 were reduced by waterlogging in both years, whereas the breakdown viscosity (BD) was reduced and unaffected by waterlogging in 2014 and 2015 (Table 1). The pasting viscosities of YN7 were decreased by waterlogging in 2014. The PV and BD were reduced, whereas the TV, FV, and SB were unaffected in 2015. The pasting temperature (P_{temp}) in YN7 was unaffected by waterlogging in both years, whereas the value in SYN5 was unaffected in 2014 but decreased by waterlogging in 2015. In general, the pasting viscosities decreased, and P_{temp} was unaffected by waterlogging. This finding is consistent with the observation on wheat [10,11] and our previous study on waxy maize starch [18,19]. The decreased viscosity is mainly due to the decreased starch content [34]. However, a study on wheat observed that short-time (7 d) waterlogging and shading increased the PV and P_{temp} but decreased the TV and FV [13]. In rice, the pasting viscosity in response to waterlogging differed among the cultivars [14]. YN7 has higher PV, BD, FV, and SB than SYN5, but they have similar TV and P_{temp}. This finding indicated that YN7 has an advantage for producing viscous foods.

Table 1. Effects of waterlogging at flowering stage on the flour pasting property of waxy maize.

Year	Hybrid	water	PV (mPa.s)	TV (mPa.s)	BD (mPa.s)	FV (mPa.s)	SB (mPa.s)	P_{temp} (°C)
2014	SYN5	CK	1384±1d	1256±6a	128±7d	1660±20a	404±14a	76.1±0.4bc
		WS	998±26e	927±24e	71±2e	1181±32e	254±8de	75.3±0.4c
	YN7	CK	1378±42d	1275±34a	103±8d	1692±52a	417±18a	76.1±0.4bc
		WS	1016±2e	983±3de	33±1f	1299±12cd	316±9b	75.3±0.4c
2015	SYN5	CK	1586±20b	1134±4b	452±16c	1427±6b	293±2bc	77.9±0.4a
		WS	1477±29c	1022±22cd	455±7c	1246±22de	224±0e	76.3±0.4bc
	YN7	CK	1791±4a	1075±5bc	717±9a	1344±1bc	270±6cd	76.3±0.5bc

		WS	1659±13b	1061±5c	599±9b	1354±9bc	293±4bc	77.1±0.4ab

Mean value in the same column followed by different letters is significantly different ($P < 0.05$). SYN5, Suyunuo5; YN7, Yunuo7; CK, control; WS, waterlogging; PV, peak viscosity; TV, trough viscosity; BD, breakdown viscosity; FV, final viscosity; SB, setback viscosity; P_{temp}, pasting temperature.

3.7. Thermal property

The gelatinization and retrogradation characteristics of waxy maize flours under different water conditions are presented in Table 2. The ΔH_{gel} was unaffected by waterlogging in both hybrids in both years, whereas the ΔH_{gel} in response to post-silking waterlogging was dependent on hybrids [19]. A study on rice also observed that ΔH_{gel} with constant flooding irrigation was higher than that after alternate wetting and drying irrigation, but it compared with conventional irrigation was fluctuated between cultivar and year [15]. The T_o and T_p in SYN5 were reduced by waterlogging in both years; the two parameters in YN7 were reduced and unaffected by waterlogging in 2014 and 2015, respectively. The T_c in SYN5 was unaffected and decreased by waterlogging in 2014 and 2015, respectively. The T_c in YN7 was unaffected and increased by waterlogging in 2015 and 2014, respectively. Constant-flooding irrigation reduced the transition temperature of starch in rice [15], indicating that waterlogging reduced the stability of the starch structure [17].

Retrogradation occurred after the gelatinized samples were stored at 4 °C for 7 days. The ΔH_{ret} and $\%R$ in both hybrids were decreased by waterlogging in 2014 and were unaffected in 2015. Our previous study observed that the $\%R$ was increased by waterlogging after pollination [19]. The discrepancy may be due to the plants grown in 2015, which has longer rainfall duration during grain filling (211 and 445 mm in 2014 and 2015, respectively); the adequate rainfall erased the influence of waterlogging during flowering. The two hybrids have similar ΔH_{gel} and T_o, but SYN5 has higher T_p, T_c, ΔH_{ret}, and $\%R$ than YN7, which endows the advantage of YN7 to produce low retrograde food.

Table 2. Effects of waterlogging at flowering stage on the flour thermal property of waxy maize.

Year	Hybrid	water	ΔH_{gel} (J/g)	T_o (°C)	T_p (°C)	T_c (°C)	ΔH_{ret} (J/g)	$\%R$ (%)
2014	SYN5	CK	8.84±0.24abc	69.6±0.1d	75.6±0.0de	82.4±0.1c	3.5±0.0a	39.7±0.9a
		WS	8.74±0.22bcd	68.5±0.1e	74.8±0.0f	81.9±0.1c	2.9±0.1b	33.0±0.3b
	YN7	CK	8.53±0.12cd	70.2±0.0cd	75.5±0.0e	82.3±0.0c	2.7±0.2b	31.9±1.5b
		WS	8.23±0.07d	68.3±0.1e	74.6±0.0f	83.1±0.1b	2.1±0.2c	24.9±1.9c
2015	SYN5	CK	8.93±0.07abc	72.5±0.0a	77.8±0.2a	84.7±0.0a	3.6±0.1a	40.8±1.8a
		WS	9.36±0.11a	70.4±0.1cd	76.2±0.1b	83.3±0.2b	4.1±0.2a	43.2±2.2a
	YN7	CK	8.83±0.11abcd	71.4±0.6b	76.0±0.0c	82.9±0.1b	3.6±0.1a	40.6±1.1a
		WS	9.16±0.30ab	70.9±0.2bc	75.8±0.2cd	83.0±0.2b	3.7±0.2a	40.0±0.7a

SYN5, Suyunuo5; YN7, Yunuo7; CK, control; WS, waterlogging; ΔH_{gel}, gelatinization enthalpy; T_o, onset temperature; T_p, peak gelatinization temperature; T_c, conclusion temperature; ΔH_{ret}, retrogradation enthalpy; $\%R$, retrogradation percentage. Mean value in the same column followed by different letters is significantly different ($P < 0.05$).

4. Conclusion

This study showed that waterlogging stress at the flowering stage decreased the grain number and weight, resulting in yield loss. The grain starch content was reduced by waterlogging. The size of starch granules was enlarged and reduced by waterlogging in SYN5 and YN7, respectively. The proportion of high-molecular weight in amylopectin and RC were decreased and increased by waterlogging in SYN5 and YN7. The grain flour pasting viscosities were reduced by waterlogging in general, whereas those parameters in response to waterlogging were inconsistent between the two hybrids across the two years. The ΔH_{gel} was unaffected, and the transition temperatures were reduced

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by waterlogging in general. The ΔH_{ret} and $\%R$ in both hybrids were reduced and unaffected by waterlogging in 2014 and 2015, respectively. Between the two hybrids, YN7 had higher pasting viscosity and low $\%R$ and is superior in producing food with viscous taste and low retrograde. The plants growth in 2015 with adequate rainfall during grain filling achieved higher grain yield, PV, BD, and $\%R$. The results offer the option to choose an optimal waxy maize hybrid under normal and waterlogged conditions based on different food utilizations.

CRediT authorship contribution statement: Huan Yang: Investigation, Formal analysis, Writing–original draft, Writing–review & editing. Xuemei Cai: Investigation, Formal analysis. Dalei Lu: Conceptualization, Funding acquisition, Methodology, Supervision, Validation, Writing–review & editing.

Declaration of competing interest: The authors declare that they have no conflicts of interest.

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