

# MAIZE YIELD AND LEAF PHOTOSYNTHETIC CHARACTERISTICS IN RESPONSE TO PLANTING DENSITIES AND APPLICATION OF YUHUANGJIN, AS A NEW PLANT GROWTH REGULATOR

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(Received 1<sup>st</sup> Mar 2019; accepted 21<sup>st</sup> May 2019)

**Abstract.** This study aims to analyze the effects of plant growth regulator on the chlorophyll fluorescence characteristics and yield of spring maize under different planting densities in Northeast China. ‘Dongnong 253’ was used as the material in this experiment and was planted at the densities of 5, 6, 7 and 8 plants m<sup>-2</sup>. The plants were sprayed at the jointing stage with the "Yuhuangjin" plant growth regulator. The results showed that increasing the planting density effectively increased the production of the maize populations, while it led to a decrease in the chlorophyll content, angle between the stem and leaf (ABSL) in the ear leaf, single plant dry weight and photosynthetic efficiency, as well as increases in the plant height, ear height and lodging risk. The application of "Yuhuangjin" decreased the plant height and ear height and increased the dry matter accumulation per plant and lodging resistance, increased the ABSL in the ear leaf, reduced the degree of overshadowing in the prophase, extended the duration of high leaf area index (LAI), delayed the senescence process of the leaf and improved the canopy aeration and transmittance capability. The application of "Yuhuangjin" improved the photosynthetic capacities and maintained higher stable photosynthesis by significantly improving the chlorophyll fluorescence parameters, such as *Fv/Fm*, *Y(II)*, and *qP* and decreasing *Y(NO)* and *qN*. This study showed that the highest yield was 11428.18 kg ha<sup>-1</sup> after the "Yuhuangjin" treatment at the planting density of 7 plants m<sup>-2</sup>. This experiment provided theoretical and experimental evidence for the effect of increased production from the use of plant growth regulator and high-density planting cultivation on spring maize in Northeast China.

**Keywords:** *maize, high-density planting, chlorophyll fluorescence characteristics, photosynthesis rate, Yuhuangjin, yield*

## Introduction

Maize (*Zea mays* L.) plays an important role in ensuring world food security (Dong et al., 2013; Li et al., 2017). Increasing planting density is important to increase maize yields. The demand for maize is increasing with the rapidly increasing demands for food, livestock feed and bio-fuel at the global scale. Previous researchers have conducted extensive work in improving maize yield potentials and increasing yields per unit area (Diallo et al., 2015; Li et al., 2016; Biswas and Ma, 2017; Ren et al., 2017a). Previous studies have shown that increasing population density is an important method to achieve high yields (Rutger and Crowder, 1967; Roy et al., 2014). The analysis of 37 super-high-yield fields between 2006 and 2007 showed that the yields reached 15000 kg ha<sup>-1</sup> in the high yield field, the densities were in the range of 79725~84630 ears ha<sup>-1</sup> (Chen et al., 2008). However, the current density of maize in the United States is generally 8 plants m<sup>-2</sup> or higher. Dense planting is the main measure used to improve maize yields (Zhao et al., 2006; Benari and Makowski, 2016). The

structure of the canopy group becomes poor with the increase in planting density, and increased density can cause the plants to shade each other. Increasing the planting density will also increase the competition for light, water and nutrients between plants (Kang et al., 2011; Farhad and Mehdi, 2015; Ren et al., 2017b). With the planting density increases, the individual plants in the group will shade each other, which will deteriorate the permeability of the canopy, decrease the photosynthetic performance, increase the plant height and increase the risk of lodging (Li et al., 2007; Sangakkara et al., 2012).

The maize in the area of Northeast China experiences low temperatures and rainy environments, which enable the root system to resist falling. Plant growth regulator is a synthetic substance that has the function of plant hormones (Yan et al., 2009). Plant growth regulator can effectively regulate the growth and development of plants and can effectively regulate the structural characteristics of the canopy. It is easy for the root system to fall in the later stages of growth and development, and this will cause many production cuts (Liao et al., 2014). Plant growth regulator technology can lower the height and ear height of maize plants (Meng et al., 2016). Plant growth regulators can enhance the anti-lodging ability, improve the chlorophyll content in leaves and prolong the duration of high leaf area index (*LAI*) values (Cheng and Liu, 2017). Plant growth regulator can truncate the elongation of internode cells, lower the center of the maize plant, increase the roughness of the stalks, improve the anti-lodging capacity (Fan et al., 2017), improve the leaf photosynthetic characteristics, photosynthetic efficiency and the rational distribution of assimilation products (Zhang et al., 2014). Moreover, plant growth regulator can also promote the absorption of the maize root system, promote the delivery of root secretions to the ground and promote the formation of grain yields (Cicchino et al., 2013).

Photosynthesis is the basis of plant growth and development (Inamoto et al., 2015). The photosynthetic rate is an important index that can reflect the photosynthetic capacity of a plant. The process of photosynthesis is composed of a series of complex photophysical and photochemical processes. During these processes, part of the light energy losses is released in the form of fluorescence. Chlorophyll fluorescence is closely related to photosynthesis (Hwang and Choo, 2016; Zhang et al., 2017).

"Yuhuangjin" plays an important role in the increase of maize planting in China. The plant growth regulator "Yuhuangjin" was provided by Fujian Haolun Biology Engineering Technology Co. Ltd, China. Its total effective component contents are the mixture of 3% diethyl aminoethyl hexanoate and 27% ethephon. Maize in Heilongjiang Province faces problems related to how both production and anti-lodging capacity can be synergistically increased. Previous researchers have conducted extensive work on the application of "Yuhuangjin", and it plays an important role in the mining potential of planting. In this experiment, we combined dense planting and plant growth regulator to study the photosynthetic characteristics of maize and the effects on yield. The results also provided an experimental basis for the reasonable application of "Yuhuangjin" in China.

## Materials and methods

### *Experimental materials and treatments*

The present experiment was carried out from 2013 to 2014 at the Xiang Fang experimental base, Northeast Agricultural University, Heilongjiang Province at 45°42'N

latitude and 126°36'E longitude. The soil is a typical black soil (typic hapludoll in USDA soil taxonomy). The soil contained organic matter (25.25 g kg<sup>-1</sup>), total nitrogen (N) (1.70 g kg<sup>-1</sup>), available potassium (K) (179.35 mg kg<sup>-1</sup>), rapid available phosphorus (P) (65.34 mg kg<sup>-1</sup>), and alkali hydrolysable nitrogen (N) (118.21 mg kg<sup>-1</sup>). The climate in the region is temperate continental monsoon. The weather data from 2013 and 2014 are provided in *Table 1*.

**Table 1.** Daily mean values of the weather variables at the experimental site during the six months of the maize growing season in 2013 and 2014

Month	Average temperature (°C)		Precipitation (mm)		Sunshine hours (h)	
	2013	2014	2013	2014	2013	2014
April	4.4	10.3	10.8	6.1	202.3	267.0
May	17.9	14.3	73.5	91.4	240.7	127.5
June	21.4	22.9	86.4	56.8	151.7	216.8
July	23.9	23.1	198.0	115.5	195.1	159.9
August	22.5	21.9	125.7	83.8	163.7	208.1
September	15.8	15.5	31.5	32.2	230.1	184.4
Total <sup>a</sup>	17.7	18	525.9	385.8	1183.6	1163.7

<sup>a</sup>Precipitation and sunshine are monthly sums, while temperature is a monthly mean of daily means

Maize was hand-sown at 7 cm depth and 70 cm row distance on May 1 in 2013 and May 1 in 2014 in a plot size of 84 m<sup>2</sup>. The supply trial variety was 'Dongnong 253'. The tested planting densities were 5 plants m<sup>-2</sup>, 6 plants m<sup>-2</sup>, 7 plants m<sup>-2</sup> and 8 plants m<sup>-2</sup>. Using the same density of rope, 20 ml of "Yuhuangjin" was added to 30 kg of water. The solution was sprayed on six leaves and one bud, and water was used as a control. Diammonium phosphate (250 kg), urea (75 kg), and potassium sulfate (150 kg) were used as seed fertilizer, and nitrogen fertilizer was applied per hectare during the jointing stage.

#### **Measurement of leaf area index (LAI)**

The leaf area was measured using three randomly selected maize plants in each plot at the silking stage. The length and maximum width of each leaf were measured with a ruler. Leaf area index (LAI) = single plant leaf area × the number of plants per unit of land ÷ unit land area.

#### **Measurement of chlorophyll content (SPAD)**

The CCM-200+ (Beijing Aozuo Ecology Instrument Co, Ltd) chlorophyll meter was used to measure the relative chlorophyll content in leaves. In each treatment phase, three plants with uniform leaf ages were selected and marked. The determination time was between 8:30 and 10:00 am, and the ear-leaf content of each reproductive period was measured. The leaf tip, leaf center and leaf base of the leaves were determined, and the mean values were calculated as the determination value of the blade. The three measured values of each treatment were calculated, and the average value was taken as the SPAD value of the treatment. In the subsequent measurements, the marked plants were selected for measurement.

### ***Measurement of net photosynthetic rate of leaves (Pn)***

The net photosynthetic rate of the leaves was determined between 9:00 and 11:00 am, and a hand-held photosynthetic rate meter instrument CI-340 (Zealquest Scientific Technology Co., Ltd.) was used to measure the ear-leaf content. The mean value of the three measured values was used as the measured value for the treatment.

### ***Measurement of chlorophyll fluorescence parameters***

The middle of the selected plant was measured to determine the chlorophyll fluorescence parameters, and the measurement was collected between 8:30 and 10:00 am. First, a leaf clip was used to hold the middle of the ear leaf, and the ear leaf was kept in the dark treatment for 20 minutes; then, the leaf clip operation steps were followed. The mean value of the three measurements was taken as the measured value. The *PSII* maximum quantum yield ( $F_v/F_m$ ), actual quantum yield ( $Y(II)$ ), non-regulating performance dissipation quantum yield ( $Y(NO)$ ), non-photochemical quenching parameters ( $qN$ ), and photochemical quenching parameters ( $qP$ ) were calculated. The measurement instrument was a portable modulated chlorophyll fluorometer (PAM-2500) (KANGGAOTE Science and Technology Co., Ltd).

### ***Measurement of biomass and yield***

To determine the biomass and yield, two consecutive four-meter lengths were selected for each process. The rows per ear, number of ear, kernels row, number of row and hundred grain weight were calculated. After harvest, 20 ears were selected to calculate the kernels row, number of row, hundred grain weight, and the theoretical yield. The biomass was determined by the drying method, and the stalks and grains were dried separately. Each process was repeated three times, and the results were averaged.

### ***Statistical analyses***

According to the analysis of variance principles, data were statistically analyzed following standard statistical methods using Microsoft Excel 2010 and SPSS 12.0. Differences between treatments were determined by a posteriori Tukey's test at a significance level of 0.05.

## **Results**

### ***Effects of plant growth regulator on leaf area index of spring maize in different planting densities***

During the reproductive process, the *LAI* first showed an increasing then a decreasing trend in both treatments (*Table 2*). The *LAI* peaks appeared around the time of silking, and the *LAI* also increased with the increase in planting density. In the "Yuhuangjin" treatment, the peak *LAI* values were 0.89% (5 plants m<sup>-2</sup>), 3.95% (6 plants m<sup>-2</sup>), 1.23% (7 plants m<sup>-2</sup>) and 8.74% (8 plants m<sup>-2</sup>) lower than the peak *LAI* values in the water treatment. From the silking stage to the dough stage, the *LAI* values were 3.75% (5 plants m<sup>-2</sup>), 6.47% (6 plants m<sup>-2</sup>), 7.14% (7 plants m<sup>-2</sup>) and 14.60% (8 plants m<sup>-2</sup>) lower than the peak values in the water treatment.

### ***Effects of plant growth regulator on chlorophyll SPAD in the leaves of northeast spring maize under different planting densities***

From the silking stage to the dough stage, the chlorophyll SPAD value decreased in the two treatments (Table 3). In the water treatment, the SPAD value decreased in every growth period with the increase in density. From the silking stage to the dough stage, the SPAD value declined by 16.72% (5 plants m<sup>-2</sup>), 23.52% (6 plants m<sup>-2</sup>), 25.01% (7 plants m<sup>-2</sup>) and 25.73% (8 plants m<sup>-2</sup>).

**Table 2.** The effects of plant growth regulator on the LAI of spring maize under different planting densities in Northeast China

Treatment	Density (plants m <sup>-2</sup> )	Jointing stage	Trumpet stage	Silking	Grain filling stage	Dough stage
Water	5	2.25±0.12 <sup>ab</sup>	3.55±0.06 <sup>d</sup>	4.05±0.09 <sup>d</sup>	3.79±0.07 <sup>d</sup>	3.28±0.07 <sup>f</sup>
	6	2.46±0.62 <sup>ab</sup>	4.13±0.12 <sup>c</sup>	4.39±0.13 <sup>cd</sup>	4.06±0.05 <sup>c</sup>	3.45±0.05 <sup>ef</sup>
	7	2.58±0.03 <sup>ab</sup>	4.73±0.16 <sup>b</sup>	4.89±0.07 <sup>b</sup>	4.49±0.12 <sup>b</sup>	3.72±0.08 <sup>cd</sup>
	8	2.88±0.09 <sup>a</sup>	5.22±0.06 <sup>a</sup>	5.59±0.08 <sup>a</sup>	4.71±0.05 <sup>a</sup>	3.84±0.06 <sup>c</sup>
Plant growth regulator	5	2.12±0.12 <sup>b</sup>	3.52±0.03 <sup>d</sup>	4.02±0.05 <sup>d</sup>	3.84±0.13 <sup>d</sup>	3.40±0.10 <sup>ef</sup>
	6	2.36±0.45 <sup>ab</sup>	4.11±0.22 <sup>c</sup>	4.22±0.23 <sup>d</sup>	4.16±0.07 <sup>c</sup>	3.58±0.12 <sup>de</sup>
	7	2.53±0.48 <sup>ab</sup>	4.71±0.29 <sup>b</sup>	4.83±0.48 <sup>bc</sup>	4.52±0.06 <sup>b</sup>	4.02±0.17 <sup>b</sup>
	8	2.67±0.12 <sup>ab</sup>	4.68±0.23 <sup>b</sup>	5.10±0.50 <sup>b</sup>	4.72±0.15 <sup>a</sup>	4.25±0.14 <sup>a</sup>

The values represent the mean ± SE (n=7). The values with the same letters in the columns are not significantly different at P < 0.05

**Table 3.** The effects of plant growth regulator on the chlorophyll SPAD values of spring maize under different planting densities in Northeast China

Treatment	Density (plants m <sup>-2</sup> )	Silking	Grain filling stage	Dough stage
Water	5	77.14±4.52 <sup>ab</sup>	74.50±4.03 <sup>a</sup>	64.24±9.82 <sup>a</sup>
	6	68.53±2.37 <sup>bcd</sup>	59.09±4.52 <sup>b</sup>	52.41±3.97 <sup>bc</sup>
	7	63.58±6.98 <sup>cd</sup>	59.18±3.49 <sup>b</sup>	47.68±3.66 <sup>c</sup>
	8	61.27±6.16 <sup>d</sup>	53.01±2.62 <sup>c</sup>	45.50±8.98 <sup>c</sup>
Plant growth regulator	5	82.66±9.52 <sup>a</sup>	71.17±1.78 <sup>a</sup>	65.34±2.09 <sup>a</sup>
	6	74.83±4.70 <sup>abc</sup>	63.60±1.95 <sup>b</sup>	62.46±1.19 <sup>ab</sup>
	7	68.57±6.24 <sup>bcd</sup>	65.24±4.62 <sup>b</sup>	60.13±3.95 <sup>ab</sup>
	8	66.64±4.06 <sup>bcd</sup>	62.50±2.86 <sup>b</sup>	55.51±2.71 <sup>abc</sup>

In the "Yuhuangjin" treatment, the SPAD value was higher than that in the water treatment in every growth period. The SPAD value in the "Yuhuangjin" treatment was higher than that in the water treatment by 7.14% (5 plants m<sup>-2</sup>), 9.19% (6 plants m<sup>-2</sup>), 7.85% (7 plants m<sup>-2</sup>) and 8.77% (8 plants m<sup>-2</sup>) at the silking stage. From the silking stage to the dough stage, SPAD value declined by 18.52% (5 plants m<sup>-2</sup>), 16.54% (6 plants m<sup>-2</sup>), 12.30% (7 plants m<sup>-2</sup>) and 16.71% (8 plants m<sup>-2</sup>) in the "Yuhuangjin" treatment. Except for the 5 plants m<sup>-2</sup> density, the "Yuhuangjin" treatment decreased the chlorophyll degradation rate at varying degrees in the later stages of procreation.

### ***Effects of plant growth regulator on the net photosynthetic rate (Pn) of leaves of northeast spring maize under different planting densities***

From the silking stage to the dough stage, the net photosynthetic rate of the leaves decreased gradually in both treatments (Table 4). In the water treatment, the net photosynthetic rate was 43.29  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (5 plants  $\text{m}^{-2}$ ), 40.42  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (6 plants  $\text{m}^{-2}$ ), 38.79  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (7 plants  $\text{m}^{-2}$ ) and 37.59  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (8 plants  $\text{m}^{-2}$ ) at the silking stage. When the planting density increased, the net photosynthetic rate decreased. Compared to the 5 plants  $\text{m}^{-2}$  density, the net photosynthetic rate in the other densities reduced by 6.64% (6 plants  $\text{m}^{-2}$ ), 10.40% (7 plants  $\text{m}^{-2}$ ) and 13.17% (8 plants  $\text{m}^{-2}$ ). At various densities, the net photosynthetic rate decreased by 46.23% (5 plants  $\text{m}^{-2}$ ), 44.47% (6 plants  $\text{m}^{-2}$ ), 44.72% (7 plants  $\text{m}^{-2}$ ) and 49.70% (8 plants  $\text{m}^{-2}$ ) from the silking stage to the dough stage.

**Table 4.** The effects of plant growth regulator on the Pn of spring maize leaves under different planting densities in Northeast China

Treatment	Density (plants $\text{m}^{-2}$ )	Jointing stage	Trumpet stage	Silking	Grain filling stage	Dough stage
Water	5	15.93±0.09 <sup>b</sup>	29.40±1.80 <sup>b</sup>	43.29±1.31 <sup>bc</sup>	35.05±0.88 <sup>b</sup>	23.28±0.88 <sup>cd</sup>
	6	14.50±0.16 <sup>d</sup>	26.53±0.91 <sup>de</sup>	40.42±0.32 <sup>d</sup>	32.22±0.42 <sup>e</sup>	22.44±0.43 <sup>de</sup>
	7	13.97±0.07 <sup>e</sup>	25.23±0.17 <sup>ef</sup>	38.79±0.41 <sup>ef</sup>	31.02±0.18 <sup>f</sup>	21.44±0.17 <sup>e</sup>
	8	13.61±0.22 <sup>e</sup>	24.58±0.44 <sup>f</sup>	37.59±1.21 <sup>f</sup>	29.90±0.66 <sup>f</sup>	18.91±0.66 <sup>f</sup>
Plant growth regulator	5	16.55±0.41 <sup>a</sup>	32.52±1.31 <sup>a</sup>	46.81±0.88 <sup>a</sup>	37.23±0.62 <sup>a</sup>	26.48±0.88 <sup>a</sup>
	6	15.99±0.09 <sup>b</sup>	29.28±0.70 <sup>b</sup>	44.28±0.32 <sup>b</sup>	34.80±0.32 <sup>bc</sup>	25.28±0.32 <sup>b</sup>
	7	15.84±0.16 <sup>bc</sup>	28.41±0.49 <sup>bc</sup>	42.65±0.66 <sup>c</sup>	33.66±0.74 <sup>cd</sup>	24.18±0.38 <sup>c</sup>
	8	15.60±0.22 <sup>c</sup>	27.16±0.96 <sup>cd</sup>	39.99±0.22 <sup>de</sup>	32.52±1.05 <sup>de</sup>	19.87±0.69 <sup>f</sup>

The net photosynthetic rate was higher in the "Yuhuangjin" treatment than in the water treatment in every growth period. The peak values were 46.81  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (5 plants  $\text{m}^{-2}$ ), 44.28  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (6 plants  $\text{m}^{-2}$ ), 42.65  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (7 plants  $\text{m}^{-2}$ ) and 40.00  $\mu\text{mol m}^{-2} \text{s}^{-1}$  (8 plants  $\text{m}^{-2}$ ) at the silking stage. The net photosynthetic rates in the "Yuhuangjin" treatments were higher than those in the water treatment by 8.15% (5 plants  $\text{m}^{-2}$ ), 9.55% (6 plants  $\text{m}^{-2}$ ), 9.95% (7 plants  $\text{m}^{-2}$ ) and 6.39% (8 plants  $\text{m}^{-2}$ ).

From the silking stage to the dough stage, the net photosynthetic rate declined by 43.42% (5 plants  $\text{m}^{-2}$ ), 42.90% (6 plants  $\text{m}^{-2}$ ), 43.29% (7 plants  $\text{m}^{-2}$ ) and 50.33% (8 plants  $\text{m}^{-2}$ ). The net photosynthetic rate in the "Yuhuangjin" treatment was higher than that in the water treatment by 13.79% (5 plants  $\text{m}^{-2}$ ), 12.64% (6 plants  $\text{m}^{-2}$ ), 12.80% (7 plants  $\text{m}^{-2}$ ) and 5.08% (8 plants  $\text{m}^{-2}$ ) at the dough stage.

### ***Effects of plant growth regulator on chlorophyll fluorescence parameters of the leaves of northeast spring maize under different planting densities***

From the silking stage to the dough stage, the PSII maximum quantum yield showed a declining trend in the two treatments (Table 5). The  $F_v/F_m$  declined by 3.16% (5 plants  $\text{m}^{-2}$ ), 3.40% (6 plants  $\text{m}^{-2}$ ), 3.19% (7 plants  $\text{m}^{-2}$ ) and 3.41% (8 plants  $\text{m}^{-2}$ ) from the silking stage to the dough stage in the water treatment. The  $F_v/F_m$  declined by 3.07% (5 plants  $\text{m}^{-2}$ ), 3.28% (6 plants  $\text{m}^{-2}$ ), 2.65% (7 plants  $\text{m}^{-2}$ ) and 2.17% (8 plants  $\text{m}^{-2}$ ) from the silking stage to the dough stage in the "Yuhuangjin" treatment. Compared

with the water treatment, the *PSII* maximum quantum yield in the "Yuhuangjin" treatment decreased by a small amount. At the same time, the "Yuhuangjin" treatment increased the *PSII* maximum quantum yield.

With the increase in density, the *Y(II)* of the two treatments showed a decreasing trend (Table 6). From the silking stage to the dough stage, the *Y(II)* in the water treatment reduced by 7.04% (5 plants m<sup>-2</sup>), 47.64% (6 plants m<sup>-2</sup>), 47.40% (7 plants m<sup>-2</sup>) and 57.49% (8 plants m<sup>-2</sup>). The *Y(II)* in the "Yuhuangjin" treatment reduced by 42.44% (5 plants m<sup>-2</sup>), 45.44% (6 plants m<sup>-2</sup>), 46.38% (7 plants m<sup>-2</sup>) and 53.42% (8 plants m<sup>-2</sup>) from the silking stage to the dough stage. The "Yuhuangjin" treatment reduced the decrease in *Y(II)* in the later stages of procreation. Compared with the water treatment, the *Y(II)* in the "Yuhuangjin" treatment increased at varying degrees. In the silking stage, the *Y(II)* values in the "Yuhuangjin" treatment were 0.71% (5 plants m<sup>-2</sup>), 1.32% (6 plants m<sup>-2</sup>), 0.67% (7 plants m<sup>-2</sup>) and 3.80% (8 plants m<sup>-2</sup>) higher than those in the water treatment. In the dough stage, the *Y(II)* values in the "Yuhuangjin" treatment were 9.46% (5 plants m<sup>-2</sup>), 5.58% (6 plants m<sup>-2</sup>), 2.61% (7 plants m<sup>-2</sup>) and 13.73% (8 plants m<sup>-2</sup>) higher than those in the water treatment.

**Table 5.** The effects of plant growth regulator on the *Fv/Fm* of spring maize leaves under different planting densities in Northeast China

Treatment	Density (plants m <sup>-2</sup> )	Silking	Grain filling stage	Dough stage
Water	5	0.799±0.002 <sup>ab</sup>	0.789±0.006 <sup>abc</sup>	0.774±0.009 <sup>a</sup>
	6	0.796±0.002 <sup>ab</sup>	0.787±0.005 <sup>abc</sup>	0.768±0.002 <sup>a</sup>
	7	0.791±0.010 <sup>ab</sup>	0.784±0.003 <sup>abc</sup>	0.766±0.011 <sup>a</sup>
	8	0.785±0.004 <sup>b</sup>	0.780±0.008 <sup>c</sup>	0.758±0.009 <sup>a</sup>
Plant growth regulator	5	0.803±0.004 <sup>a</sup>	0.797±0.008 <sup>a</sup>	0.779±0.001 <sup>a</sup>
	6	0.803±0.013 <sup>a</sup>	0.794±0.010 <sup>ab</sup>	0.779±0.019 <sup>a</sup>
	7	0.793±0.004 <sup>ab</sup>	0.784±0.007 <sup>abc</sup>	0.772±0.013 <sup>a</sup>
	8	0.786±0.012 <sup>b</sup>	0.781±0.006 <sup>bc</sup>	0.770±0.013 <sup>a</sup>

**Table 6.** The effects of plant growth regulator on the *Y(II)* of spring maize leaves under different planting densities in Northeast China

Treatment	Density (plants m <sup>-2</sup> )	Silking	Grain filling stage	Dough stage
Water	5	0.661±0.002 <sup>ab</sup>	0.608±0.024 <sup>ab</sup>	0.350±0.093 <sup>ab</sup>
	6	0.654±0.022 <sup>ab</sup>	0.593±0.024 <sup>abc</sup>	0.343±0.028 <sup>ab</sup>
	7	0.647±0.048 <sup>ab</sup>	0.576±0.030 <sup>c</sup>	0.340±0.049 <sup>ab</sup>
	8	0.615±0.039 <sup>c</sup>	0.532±0.050 <sup>d</sup>	0.261±0.047 <sup>c</sup>
Plant growth regulator	5	0.666±0.005 <sup>a</sup>	0.614±0.016 <sup>a</sup>	0.383±0.048 <sup>a</sup>
	6	0.663±0.007 <sup>ab</sup>	0.610±0.006 <sup>ab</sup>	0.362±0.040 <sup>a</sup>
	7	0.652±0.024 <sup>ab</sup>	0.586±0.014 <sup>bc</sup>	0.350±0.054 <sup>ab</sup>
	8	0.638±0.008 <sup>bc</sup>	0.548±0.024 <sup>d</sup>	0.297±0.032 <sup>bc</sup>

With the progress of procreation, the two *Y(NO)* treatments generally showed upward trends (Table 7). In the same growth period, the *Y(NO)* values of both treatments also increased with increasing density. Compared to the plant growth regulator treatment, the *Y(NO)* in the water treatment increased by 21.01% (5 plants

m<sup>-2</sup>), 24.26% (6 plants m<sup>-2</sup>), 34.38% (7 plants m<sup>-2</sup>) and 41.60% (8 plants m<sup>-2</sup>) from the silking stage to dough stage. The "Yuhuangjin" treatment increased the *Y(NO)* by 19.63% (5 plants m<sup>-2</sup>), 18.78% (6 plants m<sup>-2</sup>), 21.03% (7 plants m<sup>-2</sup>) and 24.50% (8 plants m<sup>-2</sup>) from the silking stage to the dough stage. The "Yuhuangjin" treatment reduced the *Y(NO)* at varying degrees. In the silking stage, the water treatment reduced the *Y(NO)* by 1.07% (5 plants m<sup>-2</sup>), 4.16% (6 plants m<sup>-2</sup>), 4.32% (7 plants m<sup>-2</sup>) and 5.14% (8 plants m<sup>-2</sup>) compared to the "Yuhuangjin" treatment. In the dough stage, the water treatment reduced the *Y(NO)* by 2.20% (5 plants m<sup>-2</sup>), 8.39% (6 plants m<sup>-2</sup>), 13.83% (7 plants m<sup>-2</sup>) and 16.61% (8 plants m<sup>-2</sup>) compared to the "Yuhuangjin" treatment.

**Table 7.** The effects of plant growth regulator on the *Y(NO)* of spring maize leaves under different planting densities in Northeast China

Treatment	Density (plants m <sup>-2</sup> )	Silking	Grain filling stage	Dough stage
Water	5	0.319±0.004 <sup>bc</sup>	0.343±0.001 <sup>c</sup>	0.386±0.046 <sup>c</sup>
	6	0.334±0.022 <sup>abc</sup>	0.356±0.016 <sup>c</sup>	0.415±0.024 <sup>bc</sup>
	7	0.357±0.008 <sup>ab</sup>	0.406±0.035 <sup>ab</sup>	0.479±0.006 <sup>ab</sup>
	8	0.367±0.037 <sup>a</sup>	0.414±0.048 <sup>a</sup>	0.520±0.018 <sup>a</sup>
Plant growth regulator	5	0.322±0.010 <sup>c</sup>	0.339±0.006 <sup>c</sup>	0.378±0.002 <sup>c</sup>
	6	0.320±0.030 <sup>bc</sup>	0.349±0.009 <sup>c</sup>	0.381±0.017 <sup>c</sup>
	7	0.341±0.014 <sup>abc</sup>	0.366±0.019 <sup>bc</sup>	0.413±0.022 <sup>bc</sup>
	8	0.348±0.004 <sup>abc</sup>	0.377±0.029 <sup>abc</sup>	0.434±0.019 <sup>b</sup>

With procreation, the *qN* values of the two treatments generally showed upward trends at the different densities (Table 8). From the silking stage to the dough stage, the *qN* value of the water treatment increased by 0.39% (5 plants m<sup>-2</sup>), 0.43% (6 plants m<sup>-2</sup>), 0.43% (7 plants m<sup>-2</sup>) and 0.44% (8 plants m<sup>-2</sup>). The *qN* value of the "Yuhuangjin" treatment increased by 0.37% (5 plants m<sup>-2</sup>), 0.42% (6 plants m<sup>-2</sup>), 0.41% (7 plants m<sup>-2</sup>) and 0.44% (8 plants m<sup>-2</sup>) from the silking stage to the dough stage.

**Table 8.** The effects of plant growth regulator on *qN* of spring maize leaves under different planting densities in Northeast China

Treatment	Density (plants m <sup>-2</sup> )	Silking	Grain filling stage	Dough stage
Water	5	0.038±0.008 <sup>d</sup>	0.125±0.015 <sup>abc</sup>	0.424±0.008 <sup>e</sup>
	6	0.065±0.011 <sup>c</sup>	0.145±0.005 <sup>ab</sup>	0.490±0.008 <sup>cd</sup>
	7	0.102±0.011 <sup>ab</sup>	0.151±0.002 <sup>ab</sup>	0.532±0.025 <sup>ab</sup>
	8	0.113±0.010 <sup>a</sup>	0.156±0.005 <sup>a</sup>	0.554±0.027 <sup>a</sup>
Plant growth regulator	5	0.023±0.009 <sup>d</sup>	0.108±0.008 <sup>c</sup>	0.395±0.044 <sup>e</sup>
	6	0.059±0.004 <sup>c</sup>	0.120±0.020 <sup>bc</sup>	0.475±0.007 <sup>d</sup>
	7	0.084±0.011 <sup>b</sup>	0.124±0.019 <sup>bc</sup>	0.496±0.010 <sup>bcd</sup>
	8	0.086±0.014 <sup>a</sup>	0.131±0.031 <sup>a</sup>	0.523±0.017 <sup>a</sup>

The "Yuhuangjin" treatment reduced the increase of the *qN* value and enhanced the photosynthetic performance of the leaves. With the increase in densities, the *qN* values of the two treatments showed increasing trends. In terms of the plant growth regulator,



the  $qN$  values of the blades declined under the "Yuhuangjin" treatment. In the dough stage, compared with the water treatment, the "Yuhuangjin" treatment resulted in low  $qN$  values of 6.83% (5 plants  $m^{-2}$ ), 3.20% (6 plants  $m^{-2}$ ), 6.75% (7 plants  $m^{-2}$ ) and 5.63% (8 plants  $m^{-2}$ ).

With the increase in densities, the  $qP$  values of the two treatments gradually decreased after the silking stage (Table 9). The water treatment reduced the  $qP$  values by 7.49% (5 plants  $m^{-2}$ ), 37.15% (6 plants  $m^{-2}$ ), 44.84% (7 plants  $m^{-2}$ ) and 57.25% (8 plants  $m^{-2}$ ) from the silking stage to the dough stage. The "Yuhuangjin" treatment reduced the  $qP$  values by 34.91% (5 plants  $m^{-2}$ ), 36.31% (6 plants  $m^{-2}$ ), 42.47% (7 plants  $m^{-2}$ ) and 53.83% (8 plants  $m^{-2}$ ) from the silking stage to the dough stage. Compared with the water treatment, the "Yuhuangjin" treatment exhibited varying degrees of improvement. In the silking stage, the maximum  $qP$  values were 2.05% (5 plants  $m^{-2}$ ), 3.60% (6 plants  $m^{-2}$ ), 4.56% (7 plants  $m^{-2}$ ) and 4.94% (8 plants  $m^{-2}$ ) higher than those in the water treatment.

### ***Effects of plant growth regulator on yield of spring maize in different planting densities in northeast China***

With the increase in densities, the water treatment first exhibited an increasing then decreasing trend in the yield (Table 10). The highest yield was measured in the 7 plants  $m^{-2}$  density (12346.40 kg  $ha^{-1}$ ), which was 34.94%, 12.89% and 9.58% higher than the yields in the 5 plants  $m^{-2}$ , 6 plants  $m^{-2}$  and 8 plants  $m^{-2}$  densities, and these differences were significant.

**Table 9.** The effects of plant growth regulator on  $qP$  of spring maize leaves under different planting densities in Northeast China

Treatment	Density (plants $m^{-2}$ )	Silking	Grain filling stage	Dough stage
Water	5	0.839±0.039 <sup>abc</sup>	0.754±0.006 <sup>a</sup>	0.524±0.076 <sup>ab</sup>
	6	0.814±0.015 <sup>abc</sup>	0.734±0.036 <sup>a</sup>	0.512±0.042 <sup>ab</sup>
	7	0.798±0.019 <sup>bcd</sup>	0.711±0.045 <sup>a</sup>	0.440±0.031 <sup>bc</sup>
	8	0.755±0.049 <sup>d</sup>	0.647±0.080 <sup>b</sup>	0.322±0.025 <sup>d</sup>
Plant growth regulator	5	0.856±0.002 <sup>a</sup>	0.763±0.010 <sup>a</sup>	0.557±0.099 <sup>a</sup>
	6	0.843±0.024 <sup>ab</sup>	0.758±0.005 <sup>a</sup>	0.537±0.038 <sup>ab</sup>
	7	0.835±0.010 <sup>abc</sup>	0.720±0.029 <sup>a</sup>	0.480±0.065 <sup>ab</sup>
	8	0.792±0.012 <sup>cd</sup>	0.661±0.046 <sup>b</sup>	0.366±0.046 <sup>cd</sup>

**Table 10.** The effects of plant growth regulator on yield factors of spring maize under different planting densities in Northeast China

Treatment	Density (plants $m^{-2}$ )	Rows per ear	Kernels row	Hundred grain weight (g)	Yield (kg $ha^{-1}$ )
Water	5	16.05 <sup>a</sup>	33.95 <sup>bc</sup>	33.97 <sup>a</sup>	7868.41 <sup>d</sup>
	6	15.93 <sup>a</sup>	35.58 <sup>abc</sup>	32.31 <sup>a</sup>	9404.50 <sup>c</sup>
	7	16.39 <sup>a</sup>	33.89 <sup>bc</sup>	32.74 <sup>a</sup>	10618.44 <sup>ab</sup>
	8	16.00 <sup>a</sup>	31.50 <sup>de</sup>	29.47 <sup>b</sup>	9689.59 <sup>bc</sup>
Plant growth regulator	5	15.80 <sup>a</sup>	35.95 <sup>ab</sup>	34.49 <sup>a</sup>	7887.15 <sup>d</sup>
	6	15.95 <sup>a</sup>	33.50 <sup>cd</sup>	34.36 <sup>a</sup>	9450.90 <sup>c</sup>
	7	15.45 <sup>a</sup>	36.50 <sup>a</sup>	34.61 <sup>a</sup>	11428.18 <sup>a</sup>
	8	15.90 <sup>a</sup>	31.28 <sup>e</sup>	31.71 <sup>ab</sup>	10435.83 <sup>abc</sup>

Compared with the water treatment, the production in the "Yuhuangjin" treatment increased, and the highest yield was measured in the 7 plants m<sup>-2</sup> density (13290.95 kg ha<sup>-1</sup>), but this difference was not significant. Compared with the water treatment, the yields in the "Yuhuangjin" treatment increased by 6.14% (5 plants m<sup>-2</sup>), 0.48% (6 plants m<sup>-2</sup>), 7.65% (7 plants m<sup>-2</sup>) and 3.81% (8 plants m<sup>-2</sup>). With the increase in density, rows per ear, kernels row and hundred-grain weight did not obviously change, and the differences were not significant. For the "Yuhuangjin" treatment, the differences between row and kernel numbers of the different densities were not significant, while the hundred-grain weight increased, but not significantly.

## Discussion

The main factors of maize yields include effective ear number, spike number and grain weight per unit area. The effective ear number of the group is easiest to control and is one of the main ways to increase maize yields. With the increase in density, the yield first increased and then decreased, the ear rows and the line grain number decreased, and kilo-grain weight decreased (Gilberto et al., 2013; Li et al., 2014; Niyogi, 2017). In this experiment, the changes in the yields at different densities in the two treatments are consistent with the results of previous studies. The maximum yields in both treatments occurred at the 7 plants m<sup>-2</sup> density. The maximum yields were 10618.44 kg ha<sup>-1</sup> (water treatment) and 11428.18 kg ha<sup>-1</sup> ("Yuhuangjin" treatment). Compared with the water treatment, the "Yuhuangjin" treatment increased the yields by 0.24% (5 plants m<sup>-2</sup>), 0.49% (6 plants m<sup>-2</sup>), 7.63% (7 plants m<sup>-2</sup>) and 7.70% (8 plants m<sup>-2</sup>). The differences in the ear rows and the line grain number were not obvious. The experiment also showed that the "Yuhuangjin" treatment mainly increased the hundred-grain weight, which influenced the yield.

The formation of maize yield is a group production process and not simple individual accumulation. The production of a maize population is related to the *LAI*, canopy characteristics, photosynthetic productivity and assimilation products of the entire population (Khoshbakht et al., 2017). During the growth period, the *LAI* exhibited a curve with a single peak, and the peak *LAI* was reached in the silking stage. With the increase in density, the maize yield mainly depends on the photosynthetic characteristics of the leaf groups, especially the photosynthetic capacity of the upper part and the duration of high light. In this experiment, the change in the *LAI* with density was consistent with the results obtained by Bian (Bian et al., 2011) and the spraying of "Yuhuangjin" reduced the *LAI*. The *LAI* values in the "Yuhuangjin" treatment were 0.89% (5 plants m<sup>-2</sup>), 3.95% (6 plants m<sup>-2</sup>), 1.23% (7 plants m<sup>-2</sup>) and 8.74% (8 plants m<sup>-2</sup>) lower than those in the water treatment. Spraying "Yuhuangjin" can reduce the leaf area of the colony, and it can also reduce the mutual shading between individuals within a group and improve the permeability of the group. The positive effects of the "Yuhuangjin" treatment are greater than the losses caused by the decreases in the leaf area. From the silking stage to the dough stage, *LAI* decreased by 3.75% (5 plants m<sup>-2</sup>), 6.47% (6 plants m<sup>-2</sup>), 7.14% (7 plants m<sup>-2</sup>) and 14.60% (8 plants m<sup>-2</sup>) compared to the water treatment. The aging process slows in the later stages. With the increase in density, the *LAI* of the group also increased. Increased densities also increased the supply levels of the source. The pre-control effect of "Yuhuangjin" on the leaf area of the group increased the permeability of the group and delayed the aging process of the leaf blades.

Chlorophyll is an important material in the photosynthesis process. Chlorophyll is involved in the absorption of light energy by a plant and the conversion process. The change in chlorophyll content can be reflected in the blades during the aging process. The chlorophyll content can also reflect the photosynthetic performance and is closely linked to the yield formation (Ma and Dwyer, 1998). Increasing the planting density can decrease the chlorophyll contents of individual plant leaves in a group, and it can reduce the production capacity of single plants. The "Yuhuangjin" treatment increased the chlorophyll content of the leaves and increased the photosynthetic capacity of the leaves. The living environment of an individual plant worsens with the increase in the number of maize plants number per unit area. These density increases will result in the decrease of the net photosynthetic rate of the group. The results of the "Yuhuangjin" treatment in this experiment show that the net photosynthetic rate of the blades is improved by using "Yuhuangjin". The function time of the blade is extended, which created the conditions necessary for higher yields.

The light energy absorbed by the photosynthetic apparatus is mainly utilized in the following ways: to promote photochemical reactions, to dissipate in the form of heat energy and to emit in the form of fluorescence. These three paths are interrelated with each other. The fluorescence signals emitted by plant bodies contain rich photosynthetic information that can be used to predict crop yield potentials (Gong et al., 2006). With the increase of densities, the relative chlorophyll content (*SPAD*) will decrease, and plant growth regulators can improve the chlorophyll content of maize leaves (Li et al., 2011). This study shows that the *SPAD* value of the plants treated by "Yuhuangjin" increased at different degrees under different densities. The *SPAD* values in the "Yuhuangjin" treatment were 7.14% (5 plants m<sup>-2</sup>), 9.19% (6 plants m<sup>-2</sup>), 7.85% (7 plants m<sup>-2</sup>) and 8.77% (8 plants m<sup>-2</sup>) higher than those in the water treatment. By regulating the canopy structure of the group, "Yuhuangjin" reduced the shading of the plants and increased the light energy intercepted by the ear leaves. These conditions laid the foundation for the synthesis of chlorophyll. With the increase in density, the filling stage was reflected in the *PSII* center of the ear, while the *qP* and *qN* increased. A suitable planting density can help improve the apparent quantum efficiency of maize leaves, and it can increase the primary photochemical efficiency, *PSII* quantum yield, and photochemical quenching coefficient. A suitable planting density can also reduce the photochemical quenching coefficients of the blades (Yang et al., 2009). The chlorophyll fluorescence parameters of the "rod and three leaves" under the two strains showed a unique phenomenon. The "Yuhuangjin" treatment increased the photosynthetic capacity, and the actual quantum yield increased significantly (Xu et al., 2008). With the increase in densities, the *Fv/Fm*, *Y(II)* and *qP* decreased, while the *Y(NO)* and *qN* increased in the water treatment. This result suggested that increasing planting density can lead to worse population canopy conditions and can lower the fluorescence yield. Spraying "Yuhuangjin" increased the *Fv/Fm*, *Y(II)* and *qP*, while it decreased the *Y(NO)* and *qN*. The "Yuhuangjin" treatment improved the maximum quantum yield, improved the ability of the leaves to adapt to strong light, reduced the chemical quenching parameters, enhanced the leaf physiological functions and improved the utilization of energy and the actual quantum yield of the blade. Compared with the water treatment, the "Yuhuangjin" treatment resulted in smaller decreases in *Fv/Fm*, *Y(II)* and *qP*, higher reductions in *Y(NO)*, and reductions in the *qN*. Spraying "Yuhuangjin" delayed the senescence of the leaves and increased the duration of high photosynthetic efficiency, which laid the foundation for higher yields. The changes in

the net photosynthetic rate and chlorophyll fluorescence parameters of the leaf blades showed a unique phenomenon. Increasing the planting density can reduce the photosynthetic capacity of individual plants in the population. Spraying "Yuhuangjin" can improve the photosynthetic capacity of individual plants.

## Conclusion

In the water treatment, the *LAI* increased, and the *SPAD* value of the group decreased with the increase in density. The group yield first increased and then decreased. The maximum yield was 10618.44 kg ha<sup>-1</sup> (7 plants m<sup>-2</sup>). The *Pn* decreased, and the chlorophyll fluorescence parameters *Fv/Fm*, *Y(II)* and *qP* decreased, while *Y(NO)* and *qN* increased. In the "Yuhuangjin" treatment, the *LAI* declined, the *SPAD* increased, the *Pn* increased, the chlorophyll fluorescence parameters *Fv/Fm*, *Y(II)* and *qP* increased, and the *Y(NO)* and *qN* decreased. The maximum yield was 11428.18 kg ha<sup>-1</sup> (7 plants m<sup>-2</sup>). The "Yuhuangjin" treatment slowed the aging process of the leaves, improved the photosynthetic capacity of the leaves and increased production. In maize production, it is important to pay more attention on obtain high yields by increasing the planting density in Heilongjiang Province. We still have a lot of work to do to reveal the effects of plant density and plant growth regulators on the physiological and molecular levels of maize.

**Acknowledgements.** This work was supported by Heilongjiang Provincial Funding for National Key R&D Programs of China (GX18B029), National Key R&D Program of China (2017YFD0300506, 2016YFD0300103) and "Academic Backbone" Project of Northeast Agricultural University (17XG23). We thank the anonymous reviewers and the editor for their valuable comments and suggestions, which distinctly contributed to the improvement of this paper.

**Conflict of Interests.** Authors state no conflict of interests.

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