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Brassinosteroids promote seed development and physiological maturity of oilseed rape (*Brassica napus* L.)

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Abstract: Long developmental stage and late harvest time of winter rapeseed (*Brassica napus* L.) have great negative effects on rice planting of rice-rapeseed farming system in China. Early maturity improvement of rapeseed is necessary. 'Zhongshuang 11', an elite winter rapeseed cultivar, was used in consecutive field experiments during 2010-2012. At initial flowering stage, plants were consecutively sprayed with 0.1 mg/L 2-4-Epibrassinolide (BR) for 3 d. Two hundred sampling pods from different plants were randomly collected to measure seed related indexes with a 4 d interval from 7 to 47 d after peak anthesis (DAPA). Seed color turned light brown at 31 or 35 DAPA after BR treatment, seed dry weight (DWT) was increased while seed moisture content (SMC) was decreased during seed development. DWT almost reached the maximum value when SMC was 33.20% at 31 DAPA in 2010-2011 and 35.29% at 35 DAPA in 2011-2012 growing season after BR treatment. Similarly, the maximum values of standard germination test (SGT), accelerated aging test (AAT) and cold test (CT) were observed at 31 or 35 DAPA after BR treatment respectively. The high yield and seed oil content appeared at 31 or 35 DAPA accompanied with rapid decrease in total non-structural carbohydrate (TNC) in stems and leaves. Our study indicated that BR application advanced maturity of winter rapeseed by 4 to 8 days.

Keywords: winter oilseed rape (*B. napus* L.), brassinosteroids, early maturity, seed production, seed quality

Introduction

Winter rapeseed (*Brassica napus* L.) is cultivated worldwide and plays an important role in edible oil supply. Moreover, the increasing demand is fueled by its growing popularity as renewable energy source in recent years (Adamsen and Coffelt, 2005; Durrett et al., 2008). In China, large growing region and harvesting time of winter rapeseed often hamper following rice plantation in basin areas along Yangtze River. Therefore, farmers were unwilling to plant rapeseed and the acreage was decreased dramatically in recent years. Therefore, it becomes urgent to develop winter rapeseed with high yield and early maturity. It's reported that physiological maturity of crop seeds occurred when seed dry matter accumulation reached

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maximum. Thus, at physiological maturity, crop could be timely harvested with high yield and high quality (Eastin et al., 1973; Elias and Copeland, 2001; Gesch et al., 2005; Wang et al., 2006; Berti and Johnson, 2008; Wang et al., 2008). Seed color, weight and moisture are important factors determining winter rapeseed harvest. When plant physiology reached maturity, seed weight reached the maximum and moisture level decreased (Hill et al., 2005). Canola physiological maturity occurred at approximately 5 to 6 weeks after anthesis (Elias and Copeland, 2001).

Growth and essential yield production of rapeseed can be altered by application of plant growth regulators. Many efforts have been devoted to evaluating physiological characteristics and seed production after exogenous application. Brassinosteroids (BRs) are reported to occur at low concentrations in all plants tested to date and are considered essential for growth and development (Mandava, 1988; Sakurai and Fujioka, 1993; Clouse and Sasse, 1998; Khripach et al., 2000; Ma et al., 2009). BR-induced plant growth had also been documented to be associated with increased metabolic processes, such as photosynthesis (Braun and Wild, 1984; Sairam, 1994; Yu et al., 2004; Ma et al., 2009; Xia et al., 2009). Hua reported that BR treatment increased photosynthetic activity of silique wall and seed oil content (Hua et al., 2012).

So far limited information is available on regulating early maturity of winter rapeseed with exogenous BR application. In previous experiments, foliar application with different contents of BR and other phytohormones were conducted and content was optimized (Wan et al., date not shown). Objective of this study is to investigate influence of foliar BR application on morphological and physiological changes during seed development. Meanwhile, seed production and quality at different harvest time were determined.

Materials and methods

Experimental design

Two rounds of field trials in 2010-2011 and 2011-2012 were conducted at Yangluo Experimental Station of Oil Crops Research Institute in Wuhan, Hubei, China (30°6′N, 114°1′E), located approximately in the middle of Yangtze River basin. Soil of this area is all paddy soil. Soil samples from 0-20 cm layer were collected prior to experiments and air dried, grounded and analyzed for pH, alkaline-digested N, total N, available phosphorus, available potassium and available B (Table 1). Average monthly rainfall and temperature from September 2010 to May 2011 and from September 2011 to May 2012 were listed in Figure 1. In 2011-2012, monthly rainfall was higher and temperature was particularly lower from February to May.

'Zhongshuang 11' was used in this study. Experiments were arranged in complete randomized block design with 2 treatments, *i.e.*, exogenous BR application and control. All analysis replicated 3 times with 3 plots respectively. Each plot was 2×10 m consisting of 30 rows with 33.3 cm spacing between rows. One meter spacing was left between plots to avoid fertilizer contamination. Seeds were sown on 24th September 2010 and 28th September 2011 with seeding rate of 2.25 kg/ha and planting density of 37.5×10^4 plants/ha. For fertilization, 225 kg/ha N, 90 kg/ha P_2O_5, 120 kg/ha K_2O and 9 kg/ha Borax were applied to each plot. At initial flowering stage, when

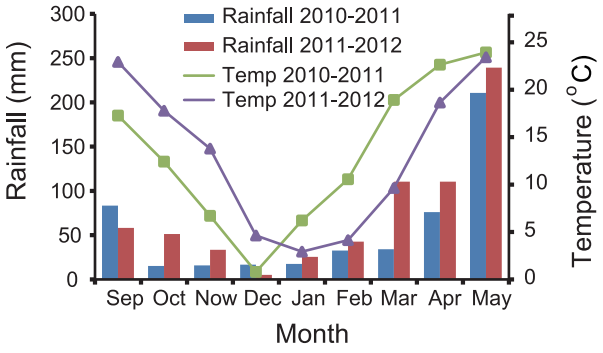

Figure 1. Average monthly temperature and rainfall during 2010-2011 and 2011-2012 at Yangluo Experimental Station.

Table 1. Soil properties during 2010-2011 and 2011-2012 growing seasons

Parameter	2010-2011	2011-2012
pH	6.82	6.65
Dissolved organic carbon (mg/kg)	104.3	85.1
Total N (g/kg)	1.51	1.69
Alkaline digested N (mg/kg)	78.2	74.3
Available phosphorus (mg/kg)	46.2	45.7
Available potassium (mg/kg)	63.1	60.3
Available B (mg/kg)	0.35	0.51

25% of main racemes were flowered, foliar application of 0.1 mg/L BR in 0.1% ethanol and 0.1% Triton X was selected to spray for 3 consecutive days in field according to Ma's report (Ma et al. 2009). Distilled water was used as control.

Determination of morphological and physiological parameters

To evaluate seed development, approximately 50 plants from each plot were randomly chosen and tagged at peak anthesis on March 20th, 2011 and March 22th, 2012 respectively. Two hundred pods, developed from tagged flowers on main inflorescences of primary branches were randomly harvested at 4 d intervals from 7 to 47 d after peak anthesis (DAPA). More than 2,000 pods were collected for each treatment. Subsequently, seed color was recorded according to Munsell's report at each sampling date (Munsell, 1977). Fresh and dry weight was measured in 1,000 seeds from marked pods. Average seed moisture content (SMC, %) and dry seed weight (DWT) were calculated according to Elias's report with some modification (Elias and Copeland, 2001).

Moisture content (%) = [(fresh weight- dry weight)/ fresh weight] × 100% (1)

Growth degree days (GDDs) index from 7 to 47 DAPA was defined as described by Wang (Wang et al. 2008).

$$GDDs = \sum[(T_{max}+T_{min})/2-T_b] \quad (2)$$

T_{max} ——maximum daily temperature, °C

T_{min}——minimum daily temperature, °C

T_b——base temperature, 5°C

Residual seeds from every sampling date in each treatment were air dried in laboratory for approximately 1 month before seed quality determination.

Seed quality determination

Germination percentage and germination rate were conducted according to Elias and Wang's method (Elias and Copeland, 2001; Wang et al., 2008). Tests were replicated 3 times with 100 seeds. Germinated seeds were counted every day. Standard germination test (SGT) was conducted on moist blotter paper in growth cabinet at 20°C for 7 d (AOSA, 1988). According to ISTA handbook, accelerated aging test (AAT) was done by aging seeds at 42°C for 48 h (Wellington, 1969). For cold test (CT), samples were placed on moist blotter paper at 5°C for 5 d and then transferred to 22°C for 5 d. Germination rate was calculated as:

$$\text{Germination rate} = \Sigma\ (Gi/Di) \quad (3)$$

Gi —— number of normal seedlings germinated on the day tested

Di —— number of days until the measurement (Maguire, 1962)

Analysis of seed production, oil content and total non-structural carbohydrate (TNC) concentration

Five BR treated plants and control plants from 23 to 47 DAPA were selected respectively. Seed was carefully harvested to measure production then dried to 9% humidity. Oil content was determined referring to Chinese National Standard GB/T 14488.1-93. Stems and leaves were dried at 105°C for 30 min and then at 70°C for 24 h and grounded into fine powder. TNC was measured according to Takai's report with some modifications (Takai et al., 2006): 0.5 g dry material was added into 20 mL water and heated at 100°C for 30 s. After cooling, 0.5 mg amyloglucosidase and 1.5 mg *a*-amylase were added in 20 mL buffer (containing 0.1 g/L $NaNO_3$, 7.96 g/L $Na_2HPO_4 \cdot 12H_2O$ and 12.08 g/L KH_2PO_4) and incubated at 40°C for 24 h to disbranch TNC into monosaccharides. Subsequently, samples were filtered and residuals were dried at 80°C for 3 h and weighted. Weight difference between initial sample and residuals was used to calculate TNC.

Statistical analysis

All data were presented as means ± standard error (SE) of 3 replicates. Multiple factors analyses were performed using SPSS statistical software package (Version 16.0) and variance (P<0.05) of data were analyzed by one-way ANOVAs test (Duncan's test).

Results

Morphological characteristics of seed

Gradual changes in seed color were observed with

pod development (Table 2). In 2010-2011, at very early days of pod growth, seeds were transparent at 11 DAPA in BR treated group and from 11 to 15 DAPA in control group. It turned light green at 15 DAPA after BR treatment and at 19 DAPA in control sample. Then, seeds turned green from 19 to 27 DAPA in BR treated group and from 23 to 31 DAPA in control group. With further development, seeds turned greenish brown and finally black from 31 to 47 DAPA in BR treated group, whereas it delayed 8 d in control group. In 2011-2012, seeds were transparent at 11 to 15 DAPA and turned from green to dark green or brownish green at 19 to 31 DAPA after BR treatment, whereas it turned from light green to brownish green at 23 to 35 DAPA in control group. It turned greenish brown and then black at 35 to 47 DAPA in BR treated group, whereas at 39 to 47 DAPA for control group (Table 2).

Physiological characteristics of seed

DWT increased and SMC decreased significantly in BR treated and control seeds in both two experimental periods (Figure 2). In 2010-2011, DWT increased rapidly from 1.2 to 4.4 g per 1,000 seeds from 11 to 31 DAPA and increased steadily from 35 to 47 DAPA in BR treated group. Conversely, DWT reached high value of 4.6 g at 39 DAPA in control group, which was 8 d later when compared with BR treatment. In contrast to DWT trend, SMC value decreased sharply. Application of BR led to rapid loss of SMC compared to control ones. It decreased rapidly from 75.87% to 33.20% during 11 to 31 DAPA after BR treatment and during 11 to 35 DAPA in control (Figure 2A). In 2011-2012, DWT increased from 1.1 to 4.8 g per 1,000 seeds from 11 to 35 DAPA and then increased slowly from 39 to 47 DAPA after BR foliar application. DWT reached a maximum value of 4.7 at 39 DAPA and increased steadily from 43 to 47 DAPA in control group. SMC in BR treated group decreased rapidly from 77.64% to 35.29% during 11 to 35 DAPA (Figure 2B).

Two-way ANOVA analysis indicated that seed development stage, also named as seed age (SA), and treatment individually affected DWT and SMC significantly (Table 3). Based on changing trends of DWT and SMC, optimum harvest time was 31 or 35 DAPA for BR treated group and 39 DAPA for control group. In other words, the accumulated GDDs (growth degree days) during these periods were from 361 to 638 in BR treated plants in 2010-2011 and approximately 406 to

Table 2. Seed color variation during development after BR treatment in two experimental years

DAPA	GDD		2010-2011		2011-2012	
	2010-2011	2011-2012	BR treatment	Control	BR treatment	Control
11	71	50	Transparent 7.5GY(8/6)	Transparent 7.5GY(8/6)	Transparent 7.5GY(8/6)]	Transparent 7.5GY(8/2)
15	117	93	Light green 7.5GY(8/8)	Transparent 7.5GY(8/6)	Light green 7.5GY(8/8)	Transparent 7.5GY(8/6)
19	158	113	Green 7.5GY(6/8-6/10)	Light green 7.5GY(8/8)	Green 7.5GY(6/8-6/10)	Light green 7.5GY(8/8)
23	253	207	Dark green 7.5GY(5/8)	Green 7.5GY(6/8-6/10)	Green 7.5GY(6/8-6/10)	Green 7.5GY(6/8-6/10)
27	303	267	Dark green to brownish green 5Y(5/4-5/6)	Dark green 7.5GY(5/8)	Dark green 7.5GY(5/8)	Green 7.5GY(6/8-6/10)
31	361	337	Greenish brown to light brown 2.5Y(5/4-5/6)	Dark green to brownish green 5Y(5/4-5/6)	Dark green to brownish green 5Y(5/4-5/6)	Dark green 7.5GY(5/8)
35	426	406	Dark brown 2.5Y(3/6)	Dark green to brownish green 5Y(5/4-5/6)	Greenish brown to light brown 2.5Y(5/4-5/6)	Dark green to brownish green 5Y(5/4-5/6)
39	493	485	Brown to black 5YR (3/2-3/4)	Greenish brown to light brown 2.5Y(5/4-5/6)	Dark brown 2.5Y(3/6)	Greenish brown to light brown 2.5Y(5/4-5/6)
43	556	545	Brown to black 5YR (3/2-3/4)	Dark brown 2.5Y(3/6)	Brown to black 5YR (3/2-3/4)	Dark brown 2.5Y(3/6)
47	638	626	Brown to black 5YR (3/2-3/4)	Brown to black 5YR (3/2-3/4)	Brown to black 5YR (3/2-3/4)	Brown to black 5YR (3/2-3/4)

Notes: DAPA, days after peak anthesis; GDD, growth degree days.

Table 3. Seed dry weight (DWT) and seed moisture content (SMC) of 'Zhongshuang 11'

Source of variation	Df	DWT	SMC
Year (Y)	1	*	*
Treatment (T)	1	**	*
Y×T	1	*	*
Seed age (SA)	9	**	**
Y×S	9	*	*
T×S	9	*	*
Y×T×S	9	*	*

Notes: *, Significant at $P < 0.05$ level; **, Significant at $P < 0.01$ level.

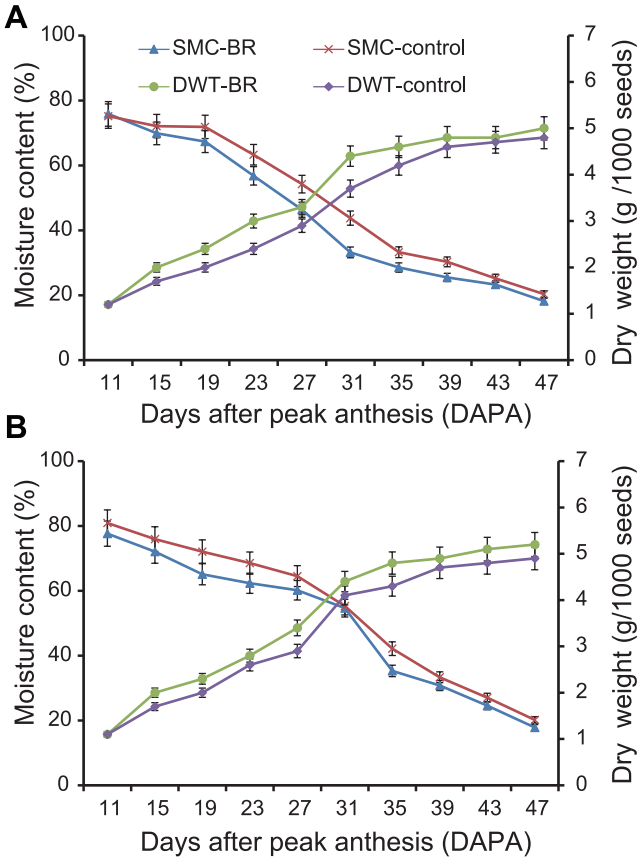

Figure 2. Seed dry weight (DWT) and moisture content (SMC) of 'Zhongshuang 11' during seed development in 2010-2011 and 2011-2012 after BR treatment. (A). DWT and SMC in 2010-2011; **(B).** DWT and SMC in 2011-2012. Error bar indicates standard error of 3 replicates.

626 in 2011-2012 respectively (Table 2). BR treatment significantly accelerated seed development and dehydration process of winter rapeseed.

Quality characteristics of seed

To obtain further evidence for BR treatment effect on seed development, seed quality was investigated by SGT. Germination percentage and germination rate of winter rapeseed in BR treated and control groups were significantly affected by sampling date (Tables 4, 5). Seeds showed no germination ability and vigor from 7 to 19 DAPA (data not shown).

Germination percentage was very low from 23 to 27 DAPA in seeds from BR treated plants, and increased significantly and almost reached 100% at 35 DAPA in 2010-2011 (Table 4). In addition, there was no significant difference from 35 to 47 DAPA. SGT was low from 23 to 35 DAPA in control group, although it increased steadily from 39 to 47 DAPA. Changing trend of AAT was similar to that of SGT despite lower values. Compared to control, AAT increased after BR treatment and the maximum value was observed earlier than control. It was very low from 23 to 27 DAPA and increased significantly at 31 DAPA in seeds from BR treated plants. A significantly higher AAT was obtained at 39 DAPA in control plants. Significantly increased CT was recorded after BR treatment. CT varied during seed development but remained without significant change from 35 to 47 DAPA in BR treatment group. It was significantly higher in control group from 39 to 47 DAPA in comparison to other developmental stages. Similar results were obtained in 2011-2012.

In BR treatment group, germination rate was very low from 23 to 27 DAPA in 2010-2011, although it increased rapidly and was significantly higher at 31 DAPA, leveling off from 35 to 47 DAPA (Table 5). SGT increased significantly at 35 DAPA in control. Germination rate calculated from AAT increased following peak anthesis. Sampling date had no effect on ATT from 35 to 47 DAPA after BR treatment. It showed a significant increase at 39 DAPA in control. The trend of CT was consistent with that of SGT and AAT, which was significantly higher at 31 DAPA after BR treatment and at 39 DAPA in control. Trend of germination rate was similar to that in 2010-2011.

ANOVA results showed that treatment, SA and the year all had significant effects on SGT, AAT and CT respectively. Additionally, interactions between SA and the year, treatment and the year, SA and treatment as well as SA, treatment and the year were highly significant for germination and germination rate (Table 6).

Table 4. Germination percentage (%, mean±1.0SE) of 'Zhongshuang 11' during seed development in 2010-2011 and 2011-2012 by SGT, AAT and CT analysis after BR treatment.

DAPA	SGT				AAT				CT			
	2010-2011		2011-2012		2010-2011		2011-2012		2010-2011		2011-2012	
	BR	Control	BR	Control	BR	Control	BR	Control	BR	Control	BR	Control
23	26±1^{d}	16±1^{d}	21±1^{d}	18±1^{d}	17±1^{d}	15±2^{d}	12±2^{d}	9±4^{d}	27±5^{c}	14±2^{d}	19±2^{d}	14±2^{d}
27	65±3^{c}	50±3^{c}	49±1^{c}	28±2^{d}	34±2^{c}	20±1^{d}	21±1^{c}	13±1^{d}	50±2^{bc}	35±4^{cd}	40±3^{c}	21±1^{d}
31	89±2^{ab}	56±2^{c}	78±4^{b}	43±1^{c}	77±1^{b}	34±3^{c}	47±1^{b}	22±1^{c}	67±2^{b}	44±5^{c}	52±1^{b}	43±3^{c}
35	95±3^{a}	79±4^{b}	95±1^{a}	74±3^{b}	86±4^{ab}	51±7^{b}	84±2^{a}	54±5^{b}	90±1^{a}	72±3^{b}	86±1^{a}	70±8^{b}
39	97±2^{a}	94±1^{a}	95±2^{a}	91±1^{a}	91±2^{a}	93±1^{a}	90±4^{a}	87±2^{a}	93±2^{a}	92±1^{a}	92±4^{a}	90±1^{a}
43	99±1^{a}	100±2^{a}	98±0^{a}	97±2^{a}	95±1^{a}	95±1^{a}	94±2^{a}	92±1^{a}	98±1^{a}	95±2^{a}	95±2^{a}	93±1^{a}
47	99±1^{a}	99±1^{a}	100±1^{a}	98±1^{a}	97±1^{a}	96±0^{a}	95±1^{a}	94±1^{a}	99±1^{a}	97±1^{a}	99±2^{a}	98±0^{a}

Notes: SGT, standard germination test; AAT, accelerated aging test; CT, cold test; DAPA, days after peak anthesis; GDD, growth degree days. The means followed by same letter are not significantly different at P=0.05. BR means BT treated group.

Table 5. Germination rate (number/d, mean±1.0SE) of 'Zhongshuang 11' during seed development in 2010-2011 and 2011-2012 by SGT, AAT and CT analysis after BR treatment.

DAPA	SGT				AAT				CT			
	2010-2011		2011-2012		2010-2011		2011-2012		2010-2011		2011-2012	
	BR	Control	BR	Control	BR	Control	BR	Control	BR	Control	BR	Control
23	9±1^{cd}	4±0^{d}	4±1^{c}	4±1^{c}	5±1^{c}	3±1^{c}	4±1^{c}	4±1^{b}	8±1^{c}	6±0^{c}	10±1^{c}	5±1^{c}
27	15±1^{c}	10±3^{cd}	11±1^{c}	11±2^{bc}	9±1^{c}	5±2^{c}	7±1^{c}	4±2^{b}	16±1^{bc}	11±3^{bc}	12±1^{c}	8±2^{c}
31	28±2^{b}	11±1^{cd}	20±1^{b}	14±1^{b}	11±1^{c}	7±1^{c}	6±2^{c}	5±1^{b}	25±2^{b}	11±1^{bc}	13±1^{c}	10±1^{bc}
35	32±1^{b}	18±1^{c}	24±1^{ab}	14±1^{b}	21±2^{ab}	12±1^{b}	16±1^{b}	9±2^{b}	30±1^{ab}	15±1^{b}	21±1^{ab}	16±1^{b}
39	37±2^{ab}	29±0^{ab}	35±2^{a}	24±2^{ab}	25±1^{a}	19±2^{a}	23±1^{ab}	21±2^{a}	34±1^{a}	22±1^{ab}	28±2^{a}	20±1^{ab}
43	42±1^{a}	30±1^{a}	37±1^{a}	31±1^{a}	27±1^{a}	19±1^{a}	29±3^{a}	23±1^{a}	34±1^{a}	24±1^{ab}	29±1^{a}	20±1^{ab}
47	44±1^{a}	34±2^{a}	35±1^{a}	32±0^{a}	28±1^{a}	23±1^{a}	30±3^{a}	22±1^{a}	36±1^{a}	30±1^{a}	31±1^{a}	25±1^{a}

Notes: SGT, standard germination test; AAT, accelerated aging test; CT, cold test; DAPA, days after peak anthesis; GDD, growth degree days. The means followed by same letter are not significantly different at P=0.05. BR means BT treated group.

Table 6. Seed quality of 'Zhongshuang 11' measured by SGT, AAT and CT after BR treatment

Source of variation	Df	Germination percentage			Germination rate		
		SGT	AAT	CT	SGT	AAT	CT
Year (Y)	1	†NS	†NS	†NS	*	*	*
Treatment (T)	1	**	**	**	**	**	**
Y × T	1	**	**	**	**	**	**
Seed age (SA)	6	**	**	**	**	**	**
Y × S	6	**	**	**	**	**	**
T × S	6	**	**	**	**	**	**
Y × T × S	6	**	**	**	**	**	**

Notes: SGT, standard germination test; AAT, accelerated aging test; CT, cold test; *, Significant at P<0.05 level; **, Significant at P<0.01 level; †NS, not significant.

Seed production, oil content and total non-structural carbohydrates (TNC) concentration

Seed production increased rapidly with pod development in both BR treated and control plants from 23 to 47 DAPA (Figure 3A). Moreover, it was significantly higher in BR treated plants than control in both two rounds of experimental years. In 2010-2011, seed production increased significantly at 31 DAPA after BR treatment but remained without significant change from 35 to 47 DAPA. However, seed production increased rapidly from 23 to 39 DAPA and a significant higher value appeared at 39 DAPA in control group. In 2011-2012, seed weight increased significantly at 35 DAPA, whereas it increased significantly at 43 DAPA in control plants. In our study, seed oil content increased rapidly with pod development and the highest values appeared at 31 DAPA (Figure 3B).

TNC of stem and leaf reduced greatly both in BR treated and control plants (Figure 3C). More importantly, in 2010-2011 for BR treated plants, it reduced sharply and lower than control plants from 31 DAPA. In 2011-2012, TNC reduced rapidly from 35DAPA in BR treatment group and from 39 DAPA in control group.

ANOVA analysis showed that not only treatment, SA and the year as the individuals, but also their interactions between SA and the year, treatment and the year, SA and treatment, SA, treatment and the year were significant for seed production, oil content and TNC (Table 7).

Table 7. Variance of seed production, seed oil content and TNC after BR treatment.

Source of variation	Df	Seed production	Seed oil content	TNC
Year (Y)	1	*	†NS	*
Treatment (T)	1	**	†NS	**
Y × T	1	**	*	**
Seed age (S)	6	**	*	**
Y × S	6	**	*	**
T × S	6	**	*	**
Y × T × S	6	**	*	**

Notes: TNC, total non-structural carbohydrate content; *, Significant at $P<0.05$ level; **, Significant at $P<0.01$ level; †NS, not significant.

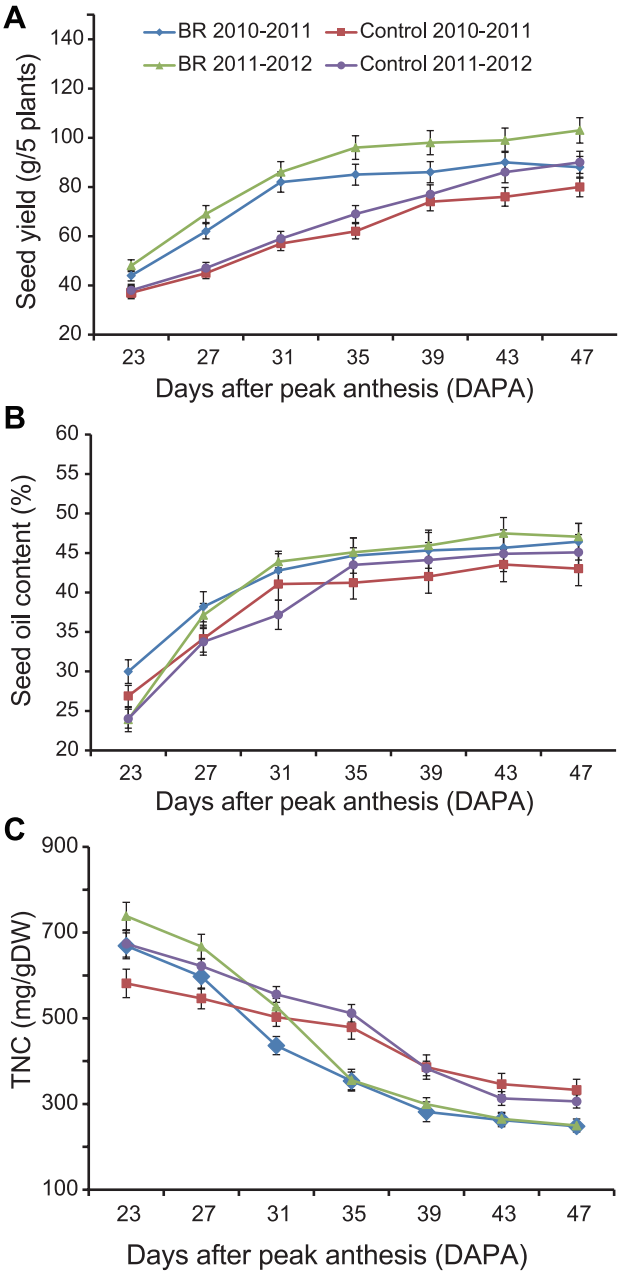

Figure 3. Seed production, oil content and TNC content in BR-treated and control plants assessed on different DAPA. (A). seed yield; **(B).** seed oil content; **(C).** TNC content. TNC, total non-structural carbohydrate; DAPA, days after peak anthesis. Error bar indicates standard error of 3 replicates.

Discussion

In two experimental years, neither the year nor interactions between SA and year were significant. SGT, AAT and CT varied during seed development in BR treatment, but remained without significant change from 35 to 47 DAPA (Table 4 and 5). Our study showed that both low and high temperature decreased percentage of germination and germination rate, which could be resulted from immature seeds being sensitive to

stress. Variation in rainfall and temperature between 2010-2011 and 2011-2012 led to different effects after BR application (Figure 1). Higher temperature in 2010-2011 promoted early maturity of seed, but decreased production and seed quality compared to 2011-2012. These data agreed with Steward's report (Steward et al., 2000). In addition, BR application at initial flowering stage turned seed color to light brown at 31 or 35 DAPA (Table 2), which indicated that BR treatment resulted in earlier physiological maturity for 4-8 d.

A number of studies reported that BR treatment increased growth and production of crops (Sairam, 1994; Clouse and Sasse, 1998). Our study showed that BR treatment at initial flowering stage increased 1,000 seed weight, seed oil content and seed yield, and decreased seed moisture content, promoted seed production and quality (Figure 2 and 3). These results were consistent with our previous study (Ma et al., 2009). Remarkable decrease of TNC could be resulted from BR promoted photosynthesis and rapid translocation of assimilates from source organs (*e.g.* leaves and stems) to sink organs (*i.e.* seeds).

In summary, exogenous application of BR promots earlier maturation of 4-8 days, increases seed production, and elevates seed oil content. It promots seed germination and positively influences assimilation and translocation of TNC. This research is the first to suggest that BR can be used for improving early maturity of oilseed rape.

Abbreviations

BR, Brassinosteroids; DAPA, days after peak anthesis; DWT, dry weight; SMC, seed moisture content; TNC, total non-structural carbohydrate concentration; SGT, standard germination test; AAT, accelerated aging test; CT, cold test; GDDs, growth degree days; SA, seed age.

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