Longer Duration of Short-day Treatment Is Required to Advance Flowering and Fruiting of Decaploid Strawberry ‘Tokun’

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Additional index words. decaploid strawberry, lateral branch, precocity, runner, short-day treatment

Abstract. Strawberry ‘Tokun’ ($2n = 10x = 70$) is a unique cultivar with special flavors, but its late maturity hampers its extension. To advance flowering and fruiting of this decaploid strawberry, the effects of short-day combined with extra nitrogen (N) nutrition treatments on strawberry ‘Tokun’ plants were studied. Runner plantlets of strawberry ‘Tokun’ were harvested and rooted in tray plugs in June 2016, 2017, and 2018, and established plants were conditioned with short-day (SD; 10 hours) and extra N nutrition. The conditioned plants were transplanted into a tabletop substrate culture system in a plastic greenhouse on 27 Aug., 3 Sept., and 10 Sept. during the 3 years, respectively, and the plants received full-element nutrient solution through the drip tube during the whole experimental period. The number of runners and lateral buds, flowering and fruiting periods, and fruit yield were investigated. Longer duration (6–7 weeks) of the SD treatment (10 hours) could significantly reduce the number of runners and increase the number of lateral buds of strawberry ‘Tokun’, advance flowering and fruiting, and achieve a fruit yield of ≈ 200 g/plant from November to December. The positive effect of extra N nutrition on flowering and fruiting of strawberry ‘Tokun’ was not found. This study is of great practical importance and guiding significance for cultivation and extension of the decaploid strawberry ‘Tokun’.

Strawberry ‘Tokun’ ($2n = 10x = 70$) is an interspecific hybrid (*Fragaria* $\times$ *ananassa* and *Fragaria nilgerrensis*) with pale yellowish orange fruit and the fruit has aroma components of caramel, coconuts, and peach (Noguchi, 2011), which attracts a lot of

consumers. A major drawback of this cultivar is its late maturity, which hampers its extension. Noguchi and Yamada (2015) reported that the harvest of this variety started in February in Japan. We also found a late-harvest start of this variety in the middle of

January when cultivated in Central Yunnan, China (lat. $23^{\circ}19' - 27^{\circ}03'N$; elevation, 1700–2300 m). Studies of the unique cultivar Tokun are needed to find practical ways to advance flowering and fruit harvest, which will benefit both growers and consumers.

Intense studies have been done regarding the effects of temperature and photoperiod on flower induction of octoploid strawberries, and SD and night-chilling treatments have been used to induce flowering and promote early fruit production in autumn (Durner, 2015, 2016; Heide and Sønsteby, 2007; Ruan et al., 2011; Serge and Hancock, 2005; Sønsteby et al., 2009). For ‘Tokun’, SD combined with night-chilling treatments are also found to be effective at inducing flower buds and promoting early fruit production in Japan (Noguchi and Yamada, 2015). Aside from Japan, however, no reports were found regarding regimes to promote ‘Tokun’ flowering and fruiting. Practical techniques to advance flowering and fruiting of ‘Tokun’ should be developed according to the local climate and production systems.

Plant nutrient status, especially N status, is another factor that influences flower bud induction (Guttridge, 1985). In general, less N can promote flower bud induction (Seo et al., 2014), especially for vigorous cultivars. However, SD treatment and timing of N fertilization have strong interactive effects on flower bud induction. The right timing of N fertilization can promote flower bud initiation for some octoploid strawberries such as ‘Korona’ (Sønsteby et al., 2009, 2013). For the decaploid strawberry ‘Tokun’, the effects of timing of N fertilization relative to start of SD treatments on flower induction need to be studied.

The Central Yunnan area, with its high elevation and low latitude, has the mildest climates in China (Wu et al., 2017). The perpetual spring-like weather provides ideal growth conditions for strawberries (Wang et al., 2018). In this area, 4 weeks of SD treatments in natural summer temperatures is enough for octoploid strawberries ‘Akihime’ and ‘Seolhyang’ to induce flower budding and to harvest in autumn (Ruan et al., 2015). There are no reports of flower bud induction of ‘Tokun’ in the Central Yunnan area regarding SD treatment and N fertilization. Thus, in our study, we investigated the parameters of flowering and fruiting, as well as some morphological traits, of ‘Tokun’ plants conditioned with 5, 6, and 7 weeks of SD treatment supplemented by different timing of N fertilization. A practical regime was developed and recommended to advance flowering and fruiting for decaploid strawberry ‘Tokun’ in the Central Yunnan area.

Materials and Methods

Plant materials. Plug plants of ‘Tokun’ were used in this experiment for SD and extra N nutrition conditioning. Runner plantlets with two to three leaves were collected on 1 June to 3 June in 2016, 2017, and 2018 from mother plants of ‘Tokun’, that were cultivated in a tabletop substrate culture system as

Table 1. Calendar of treatments showing short-day treatments and nitrogen (N) nutrition treatments.

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The letter A represents the formula of nutrient solution A, the letter B represents the formula of nutrient solution B, and the shaded part represents short-day treatment. Solution A contains 30 g compound fertilizer 'SoluFeed' dissolved in 100 L water containing N (99.6 mg·L⁻¹), phosphorus (13 mg·L⁻¹), potassium (99.6 mg·L⁻¹), and other middle and trace elements. Solution B was increased with N nutrition by adding 50 g ammonium nitrate to 10 L solution A, resulting in an N content increase of 272.8 mg·L⁻¹.

described in previous work (Ruan et al., 2017), at the farm (lat. 24°18'N; elevation, 1760 m) of the National Engineering and Technology Research Center for Ornamental Horticulture located in Yuxi city, Yunnan province. The runner plantlets were put in trays (32 plugs 50 cm in length, 24 cm in width, and 9 cm in height) filled with 80% peatmoss and 20% perlite (by vol.) for rooting, and local standard management practices were adopted. On 11 July, the plugged plants were fully established under natural sunshine conditions and the plants were ready for conditioning.

SD conditioning. Trays of 'Tokun' were separated randomly into seven groups. Each group included three trays, with 32 plugged plants per tray. Group 1 to group 6 (namely, T1–T6) were conditioned by SD (10 h) and group 7 (namely, T7) was conditioned under the natural sunshine (NS; 13–14 h, varying with season progression) (Table 1). All plants in the seven groups received nutrition treatments as depicted in Table 1. SD treatments started on 24 July and ceased on 27 Aug., 3 Sept., and 10 Sept., resulting, respectively, in 5, 6, and 7 weeks of SD conditioning for each of T1 to T6. T7 plants were also planted on 27 Aug., 3 Sept., and 10 Sept. as control plants for each planting. The SD treatment were carried out in a tunnel-shaped frame covered by black and white polyethylene (PE) film (0.1 mm, Taekwang Company) as described in previous work (Ruan et al., 2011).

N nutrient conditioning. Each individual tray plant received 50-mL nutrition solutions

on a thrice-weekly basis from 10 July to 26 Aug. There were two nutrition solutions (Table 1). In solution A, 30 g compound fertilizer, 'SoluFeed', was dissolved in 100 L water containing N (99.6 mg·L⁻¹), phosphorus (13 mg·L⁻¹), potassium (99.6 mg·L⁻¹), and other middle and trace elements. Solution B was increased with N nutrition by adding 50 g ammonium nitrate to 100 L solution A, resulting in an N content increase to 272.8 mg·L⁻¹. Solution B was used 3 consecutive weeks to increase N nutrition of plants. It started at various weeks before and during the SD period (T1–T5). The control groups T6 and T7 received solution A only (Table 1).

Plant establishment and performance. At the end of the SD treatments in 2016, 2017, and 2018, the conditioned plants were planted, respectively, on 27 Aug., 3 Sept., and 10 Sept. (planting times P1, P2, and P3, respectively) in a tabletop substrate culture system for evaluation of flowering and fruiting, along with other morphological traits. Each treatment by planting included four replicates, with eight plants per replicate, and the plants of each replicate for each treatment by planting were planted randomly in the tabletop substrate culture system as described. Plant management after planting was the same as in previous studies (Ruan et al., 2011, 2017).

For each plant, the following data were collected on a thrice-weekly basis: days from transplanting to the emergence (appearance) of the primary inflorescence seen by the naked eye, start date of the first flowering, and start date of the harvest for the 3 years of experiments. The runners and lateral branches were removed from the plants, and their numbers were recorded for 5 weeks after planting on a once-weekly basis. On a twice-weekly basis, individual marketable fruit (>7 g) was harvested and weighed for the plants planted in 2018, and data were collected until 28 Feb. 2019.

Statistical analysis. Experimental data were subjected to analysis of variance by standard procedures using dps, version 7.05. Duncan's multiple range test was used to determine significant differences among the results. A *P* value of ≤0.05 was considered significant.

Results

Flowering and fruiting of 'Tokun' plants for three seasons. SD treatments significantly promoted the flowering and fruiting of straw-

berry 'Tokun' with regard to days to inflorescence emergence, flowering, and harvest of the first fruit according to mean data across all 3 years of the study (Table 2, Fig. 1). For the T7 plants, days to inflorescence emergence, flowering, and harvest of the first fruit from 27 Aug. were 91, 110.4, and 155.4 d, respectively, which is ≈40 d more than those data for the T1 through T6 plants. Furthermore, the SD treatment significantly decreased the flower number of the first inflorescence from 22.6 (T7) to 18.6 to 21.3 for T1 through T6.

In consideration of the duration of SD treatments, 6 or 7 weeks advanced flowering and fruiting significantly compared with 5 weeks. For plants treated with 5 weeks of SD, days to inflorescence emergence, flowering, and harvest of the first fruit from 27 Aug. were 71.0, 87.9, and 129.1 d, respectively, which is ≈20 d more than those data for SD plants treated for 6 and 7 weeks (Table 2), even though these plants were planted 1 and 2 weeks later. However, the longer duration of the SD treatments decreased significantly the flower number of the primary inflorescence from 23.5 at 5 weeks to 21.0 at 6 weeks to 16.4 at 7 weeks.

In consideration of N nutrition, extra N nutrition only increased significantly flower number of primary inflorescence from 18.6 (T6) to a range of 19.2 (T3) to 21.3 (T5). For different N fertilization timings (T1–T5), no significant differences were detected for any flowering or fruiting date.

Among the three experiment years, days to inflorescence, days to flowering, days to harvest, and flower number of primary inflorescence varied significantly (Table 2, Fig. 1).

Fruit yield of 'Tokun' plants for the 2018 season. Fruit yield data were collected for the 2018 season and are presented monthly (Nov. 2018, Dec. 2018, Jan. 2019, and Feb. 2019) and as a total (Nov. 2018–Feb. 2019) in Table 3. SD treatments (T1–T6) realized fresh fruit production in November (35.0–60.5 g/plant) and December (78.1–91.5 g/plant), whereas T7 plants yielded only 0.3 g/plant in December. In January, even the fruit yields of the plants conditioned with SD (T1–T6) were greater than those of the T7 plants, but were not significant. In February, the fruit yields of the T7 plants were greater than those of the T1 through T6 plants. With regard to

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J.R. conceived and designed the experiment, and wrote the article. G.W. and G.N. performed the experiment and collected the data. C.Y., F.L., and L.T. analyzed the data and prepared the figures. L.W. conceived and designed the experiment, and reviewed the draft of the article. Original data have been saved by J.R.

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Table 2. The reproductive and vegetative growth of strawberry 'Tokun' plants conditioned by different short-day duration treatments (D) and different timing of nitrogen fertilization (T) for the 3-year experiment.

Treatments	Days to inflorescence	Days to flowering	Days to harvest	No. of flowers	No. of runners	No. of lateral buds
Years (Y)						
2016	72.2 a	85.7 a	124.1 a	29.7 a	6.4 a	1.8 c
2017	62.6 b	76.5 b	123.1 a	19.1 b	4.8 b	2.2 b
2018	43.4 c	58.6 c	93.9 b	12.1 c	3.1 c	6.4 a
SD						
5 weeks	71.0 a	87.9 a	129.1 a	23.5 a	5.6 a	3.3 b
6 weeks	55.0 b	66.1 b	106.2 b	21.0 b	3.8 b	3.7 a
7 weeks	52.2 b	66.9 b	105.8 b	16.4 c	1.9 c	3.5 ab
T						
T1	55.1 b	67.5 b	107.3 b	20.2 bc	3.4 c	4.0 a
T2	54.2 b	68.2 b	107.5 b	20.1 bc	3.3 cd	3.9 a
T3	51.1 b	64.3 b	103.7 b	19.2 cb	3.0 d	3.8 ab
T4	54.8 b	68.8 b	108.0 b	20.1 bc	3.6 bc	3.6 ab
T5	55.5 b	68.5 b	107.6 b	21.3 b	3.9 b	3.9 a
T6	54.1 b	67.5 b	106.6 b	18.6 d	2.9 d	3.4 b
T7	91.0 a	110.4 a	155.4 a	22.6 a	6.3 a	1.6 c
Analysis of variance						
Factors			<i>P</i> value			
Y	***	***	***	***	***	***
D	***	***	***	***	***	***
T	***	***	***	***	***	***
Y × D	***	***	***	***	***	***
Y × T	***	***	***	**	***	***
D × T	**	*	*	NS	***	NS
Y × D × T	NS	NS	NS	***	***	NS

Different letters in the same column mean a significant difference at the level of $P \leq 0.05$ based on Duncan's multiple range test.

NS, *, **, ***Nonsignificant or significant at $P < 0.05$, 0.01, or 0.001, respectively

total fruit yield, from Nov. 2018 to Feb. 2019, the fruit yields of the T1 through T6 plants were significantly greater than those of the T7 plants.

With regard to duration of SD treatment, in Nov. and Dec. 2018, plants treated with SD for 6 and 7 weeks yielded significantly more fruit than plants treated with SD for 5 weeks (Table 3). In Jan. 2019, plants treated with SD for 7 weeks produced a significantly greater yield—113.0 g/plant—than plants treated with SD for 5 and 6 weeks (81.8 and 78.8 g/plant, respectively). In February, 5-week conditioned plants produced 323.8 g/plant of fruit and 6-week conditioned plants produced 303.9 g/plant of fruit; both values are significantly greater than the 7-week conditioned plants (264.4 g/plant). In terms of total fruit yield from Nov. 2018 to Feb. 2019, 7-weeksconditioned plants yielded more fruit than 6-week conditioned plants; 5-week conditioned plants yielded the least fruit.

With regard to N nutrition, only yields in November were significantly different among the T1 through T6 treatments.

Number of runners and lateral branches of 'Tokun' plants for three seasons. SD treatments reduced significantly the number of runners, from 6.3/plant (T7) to a range of 2.8/plant (T3) to 3.9/plant (T5), while significantly increasing the number of branch crowns, from 1.6/plant (T7) to a range of 3.2/plant (T6) to 4.0/plant (T1). The longer duration SD treatments reduced significantly the number of runners, from 5.6/plant at 5 weeks to 3.8/plant at 6 weeks, to 1.9/plant at 7 weeks. However, the number of lateral branches increased: 3.3/plant at 5 weeks to 3.7/plant at 6 weeks to 3.5/plant at 7 weeks.

In terms of N nutrition, extra N nutrition increased the number of runners from 2.9

(T6) to a range of 3.0 (T3) to 3.9 (T5), and increased the number of lateral branches from 3.4 (T6) to a range of 3.6 (T3) to 4.0 (T1) (Table 2). Among different timings of N fertilization (T1–T5), significant differences were found both for the number of runners and lateral branches, but not reaching a conclusion for the effect of timing of N fertilization (Table 2, Fig. 1).

Among the 3 experiment years, the number of runners and lateral branches varied significantly (Table 2, Fig. 1).

Discussion

Flowering, fruiting, and fruit yield. For decaploid strawberries, only one study—by Noguchi and Yamada (2015)—reported that an 8-h SD treatment supplemented with 12 °C night chilling during August and September in Japan could promote flowering and realize a 33-d earlier fruit harvest, advancing the start of fruit harvest from February to January. In our study, we demonstrated that 6 to 7 weeks of a 10-h SD treatment without temperature manipulation in July and August in Central Yunnan, promoted flowering and realized a 2-month earlier fruit harvest, advancing from January to November. Around 200 g/plant of fruit yield in November and December, from the conditioned plants, markedly increased the grower's income and satisfied consumer demand for this peach-like-tasting strawberry.

Temperature and daylength are main factors influencing flower initiation, and SD and/or lower temperature treatments are commonly used to promote flowering and fruiting for octoploid strawberries (Durner, 2015, 2016; Heide and Sønsteby, 2007; Heide et al., 2013; Ruan et al., 2011; Serge and

Hancock, 2005; Sønsteby et al., 2009). In Central Yunnan, which is characterized by perpetual spring-like weather (Wu et al., 2017), Ruan et al. (2015) reported that 4 weeks of a 10-h SD treatment could make octoploid strawberries such as 'Akihime' and 'Seolhyang' initiate early flowering and realize a fruit harvest in the middle of October. Because of the cool summer temperatures in Central Yunnan (Wu et al., 2017), just the SD treatment, in natural temperatures, promotes flowering and fruiting of 'Tokun', which is more convenient and effective compared with the reported methods of Noguchi and Yamada (2015) in Japan.

In terms of duration of SD treatment, 3 to 4 weeks of SD treatment was enough for flower induction of octoploid strawberries such as 'Korona', 'Akihime', and 'Seolhyang' (Ruan et al., 2011, 2015; Verheul et al., 2006). For the decaploid strawberry 'Tokun', which starts to flower in late December in Central Yunnan, which is 2 months later than 'Akihime', we demonstrated that 2 to 3 weeks more of the SD treatment is needed for flower induction, compared with the SD duration needed for 'Akihime' in Central Yunnan.

Plant nutrient status, especially N status, is another factor influencing flower bud induction (Guttridge 1985). In general, less N can promote flower bud induction (Seo et al., 2014), especially for vigorous cultivars. But, SD treatments and the timing of N fertilization have strong interactive effects on flower bud induction, and the right timing of N fertilization can promote flower bud initiation for some octoploid strawberries such as 'Korona' (Sønsteby et al., 2009, 2013). In our study, an interaction between SD treatment and timing of N fertilization on flowering was not found, which could be attributed to

Fig. 1. (A) Days from planting to flowering, (B) number of flowers on the primary inflorescence, (C) number of runners, and (D) number of lateral branches of strawberry 'Tokun' plants conditioned by different short-day (SD) treatment durations and different timing of nitrogen fertilization in the 2016, 2017, and 2018 seasons. Bars denote standard deviation.

sufficient N nutrition indicated by leaf chlorophyll concentrations (SPAD values; data not shown). In other studies, plants suffered N nutrient deficiency to some extent (Sønsteby et al., 2009, 2013).

Number of runners and lateral branches. SD treatments can increase the number of lateral branches and decrease the number of runners for octoploid strawberries such as 'Korona' and 'Elsanta', and the effects were enhanced by longer duration SD treatments (Konsin et al., 2001; Sønsteby and Nes,

1998). In our study, SD also had similar effects, reducing runners and increasing lateral branches for the decaploid strawberry 'Tokun'; a longer duration of SD enhanced these effects. As described by Konsin et al. (2001), a reduced number of runners could save labor input to remove them, and an increased number of lateral branches could increase yield potential by providing more sites to initiate flower budding. These changes of reducing the number of runners and increasing the number of lateral branches

of 'Tokun' plants conditioned by SD are positive for farmers.

During the 3 years of the experiment, a consistent trend was noted: 6 to 7 weeks of SD treatment promoted the precocity of plants, indicated by days to inflorescence, days to flowering, days to harvest; reduced the number of flowers in primary inflorescence and the number of runners; and increased the number of lateral branches. The varying data for each parameter among years might be attributed to the varying sunshine,

Table 3. The marketable fruit yield of strawberry 'Tokun' plants conditioned by different short-day durations (SD) and different timing of nitrogen fertilization (T).

Treatment groups	Monthly yield per plant				Total yield/plant
	Nov. 2018	Dec. 2018	Jan. 2019	Feb. 2019	Nov. 2018–Feb. 2019
SD					
5 weeks	36.0 b	49.9 b	81.8 b	323.8 a	491.3 a
6 weeks	44.8 a	84.7 a	78.8 b	303.9 a	511.8 a
7 weeks	44.9 a	81.8 a	113.0 a	264.4 b	503.8 a
T					
T1	40.0 cd	78.1 a	100.9 a	314.3 b	532.8 a
T2	65.3 a	80.1 a	93.2 a	270.2 b	508.1 a
T3	60.5 ab	91.1 a	88.5 a	282.7 b	522.8 a
T4	50.7 bc	86.6 a	94.3 a	267.5 b	498.7 a
T5	35.0 d	84.0 a	93.8 a	297.3 b	641.3 a
T6	41.3 cd	84.8 a	89.9 a	264.8 b	480.7 a
T7	0 e	0.3 b	77.7 a	384.8 a	462.9 b
Analysis of variance					
Factors	<i>P</i> value				
D	***	***	**	*	NS
T	***	***	NS	*	*
D × T	NS	NS	NS	NS	NS

Different letters in the same column mean a significant difference at the level of $P \leq 0.05$ based on Duncan's multiple range test.

NS, *, **, ***Nonsignificant or significant at $P < 0.05$, 0.01, or 0.001, respectively

temperature, and other factors in different years.

Conclusion

In Central Yunnan, 6 to 7 weeks of SD treatment in natural temperatures is required to advance flowering and fruiting of the decaploid strawberry 'Tokun' and to realize a fruit yield of ≈ 200 g/plant in November and December. Six to 7 weeks of SD treatment also reduced significantly the number of runners and increased the number of lateral branches of the strawberry 'Tokun'.

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