

The Effects of Day and Night Temperature on the Dry Matter Accumulation of Oriental Hybrid Lily ‘Siberia’ as they Relate to the Photosynthetic and Respiratory Characteristics

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Abstract

The net photosynthetic rate on a per-plant basis of the Oriental hybrid lily ‘Siberia’ was low at the leafing stage but increased rapidly at the visible flower bud and flowering stages. The range of 20 to 25°C was the optimum temperature for photosynthesis at the three plant developmental stages under PPFD of 700 to 850 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. The respiration rate increased with the increasing temperature quadratically (in aerial plant parts) or exponentially (in underground parts). The ‘Siberia’ plants were grown under three day temperature (DT) conditions of 28°C, 24°C or 20°C, with a night temperature (NT) of 15°C or three NTs of 25°C, 20°C or 15°C, with DT of 25°C in the growth chambers. At the flowering stage, the plants grown under low DT of 20°C had a high dry weight (DW) of the total plant, flowers, bulb, and roots, and a high DW/fresh weight (FW) ratio of the leaves, stem, and bulb. The plants grown under low NT of 15°C had a high DW of the total plant and stem, and a high DW/FW ratio of the leaves and bulb. The plants that were grown under a low NT had a long stem length by the positive DIF effect, which was supported by sufficient dry matter accumulation. The plants grown under low DT of 20°C showed a high relative growth rate (RGR) and net assimilation rate (NAR) from planting to flowering, whereas the NT condition did not affect the RGR or NAR.

Discipline: Horticulture

Additional key words: cut flower quality, growth analysis, positive DIF effect

Introduction

Cut Oriental hybrid lilies (*Lilium* hybrids) are very popular in Japanese markets for their colorful and gorgeous appearance. Only a few studies have focused on the temperature and light conditions for Oriental hybrid lily production and quality (Blom & Kerec 2003, Sorrentino et al. 1997, Yamaguchi & Kamijo 1991, Zhang et al. 2011a, Zhang et al. 2011b). The quality of horticultural products is based largely on the accumulation of dry matter (DM), which is expressed based on the dry weight (DW) that is mostly assimilated via photosynthesis. We investigated the

effects of light intensity on the photosynthetic characteristics on a per-whole-plant basis as well as on DM accumulation, focusing on three developmental stages from leafing (when young leaves unfold) to flowering in ‘Siberia’—a typical Oriental hybrid lily (Inamoto et al. 2015). These experiments indicated that leaf photosynthesis was effective for DM accumulation after the visible flower bud stage during ‘Siberia’ lily growth. Moreover, we previously grew ‘Siberia’ lily plants under different combinations of day temperatures (DTs) and night temperatures (NTs) that were changed simultaneously, and found that combinations of low DT and NT promoted DM accumulation and improved

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the quality of cut flowers (Inamoto et al. 2013). It is conceivable that DT and NT significantly affect photosynthesis and dark respiration, respectively.

The present study examined the effects of temperature on photosynthesis and respiration of the Oriental hybrid lily 'Siberia' during different stages of plant development. Standard measurements of photosynthesis in lily have been made on a per-leaf area (LA) basis (Sorrentino et al. 1997, Yamaguchi & Kamijo 1991, Wang 1990, Zhang et al. 2011a). However, Righetti et al. (2007) indicated that the expression of CO₂ assimilation data on a per-LA basis makes it difficult to determine how the efficiency of photosynthesis changes as plants mature. Therefore, we measured the net photosynthetic rate (PR) and dark respiration rate (RR) of potted intact plants on a per-plant basis using an open gas-exchange system. In addition to evaluating the rate of photosynthesis, we also cultured the plants under different DTs and NTs that were changed independently to evaluate the consistency of these results with the characteristics of photosynthesis and dark respiration.

Materials and Methods

1. General methods

All experiments were conducted at the NARO Tohoku Agricultural Research Center in Morioka, Japan. Bulbs of 'Siberia' (18–20 cm in circumference) produced in the Netherlands and stored at sub-zero temperatures were obtained from an importer on January 26, 2010 (Experiments 1 and 2) or February 2, 2009 (Experiment 3). We stored the bulbs at 2°C by planting (Experiment 1) or at -2°C by pre-rooting treatment (Abe et al. 1998) (Experiments 2 and 3). In all three experiments, the plants were grown under a natural day length.

2. Relationship between temperature and PR/RR at different plant developmental stages (Experiment 1)

The same series of photosynthesis measurements were repeated three times in 2010, all yielding consistent results. The typical results are presented here, obtained from the same series of experiments used to assess light-photosynthesis relationships as reported by Inamoto et al. (2015).

The bulbs were planted in pumice in plastic pots (25 cm in diameter; 30 cm in depth) on March 24, 2010. The planting depth was approximately 10 cm to ensure the adequate growth of stem roots (Beattie 1993). A slow-release fertilizer (35 g per pot; Micro Long Total 201-100; Jcam Agri. Co., Tokyo, Japan) with 12:10:11:2:0.1:0.06 (w/w) N:P₂O₅:K:Mg:Mn:B was applied to the surface of each pot. The plants were cultured in a glasshouse with no shading from planting until May 19, 2010. On May 19, the plants were transferred to a plastic house covered by a sheet of black cheesecloth with a shading rate of 40% to

prevent an excessive rise in temperature inside the house. The houses were heated at 15°C and ventilated at 25°C throughout the experiment.

Photosynthesis was measured on April 20, 2010 (27 days after planting; the leafing stage), May 11 (48 days after planting; the visible flower bud stage), and June 24 (92 days after planting; the flowering stage). At the flowering stage, the flower buds swelled rapidly and began to color.

An open-chamber system for the photosynthesis measurements was constructed using transparent polycarbonate (120 cm long × 50 cm wide × 90 cm high). The chamber was placed in a programmable temperature room (PTR; LP-1PP; Nippon Medical and Chemical Instrument Co., Osaka, Japan). Light was supplied by fluorescent tubes (FPR96EX-D/A; Panasonic Electric Works Co., Osaka, Japan) set above (six tubes) and on both sides (four tubes on each side) of the chamber. Temperature in the chamber was changed by controlling temperature in the PTR in steps from 5°C to 35°C. Photosynthetic photon flux density (PPFD) of 700 to 850 μmol photons m⁻² s⁻¹ was supplied for PR measurements at the plant canopy, and air in the chamber was humidified to > 70% relative humidity (RH) with a supersonic humidifier (HM-303N; TKG Co., Tokyo, Japan). Other equipment, measurement procedures, and calculations for PR estimates are described in our previous report (Inamoto et al. 2015).

Six (at the leafing stage) or four (at the visible flower bud and flowering stages) plants were arranged in the chamber at a sufficient inter-plant distance to avoid mutual shading. For measurements at the flowering stage, the flower buds were removed to avoid any overestimation of the respiration rate because we had previously observed an extremely high rate in flower buds at the flowering stage compared to that of other plant organs (data not shown). The plots of the leaf temperature–PR relationships on a per-plant basis were fitted to a cubic function as follows:

$$P_n = aT_{leaf}^3 + bT_{leaf}^2 + cT_{leaf} + d \quad [1]$$

where, P_n is the net photosynthetic rate (μmol CO₂ plant⁻¹ s⁻¹), T_{leaf} is the leaf temperature (°C), and a , b , c and d are constants. The PRs on a per-LA basis were calculated by dividing the values on a per-plant basis by the total LA.

Following the measurement of PR, the dark respiration rate (RR) of the whole plant was measured without lighting using the same measurement system and method for PR. On the next day or the day after measuring the PR and RR of the whole plant, aerial parts (i.e., leaves, stem aboveground, flower buds) of the plant were excised and the RR of the underground parts (i.e., stem underground, bulb, bulblets, roots) was measured. The plots of the temperature–RR relationships of the underground parts were fitted to an exponential function.

$$R_u = ae^{T_{med}} \quad [2]$$

where, R_u is the RR of underground parts ($\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$), T_{med} is the temperature of the medium ($^{\circ}\text{C}$), e is the base of natural logarithms, and a is the constant.

The RR of the aerial parts of the plant was calculated by subtracting the RR value of the underground parts as estimated by Formula [2] from the measured RR value of the whole plant as follows:

$$R_a = R_w - R_u \quad [3]$$

where, R_a is the RR of the aerial parts ($\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$), R_w is the RR of the whole plant ($\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$), and R_u is the RR of the underground parts ($\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$).

Plots of the temperature–RR relationships of the aerial parts were fitted to a quadric function:

$$R_a = aT_{leaf}^2 + bT_{leaf} + c \quad [4]$$

where, R_a is the RR of the aerial parts ($\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$), T_{leaf} is the leaf temperature ($^{\circ}\text{C}$), and a , b , and c are constants.

3. Effect of DT on DM accumulation (Experiment 2)

The pre-rooting treatment of bulbs from 6 to 10 $^{\circ}\text{C}$ in coir (coconut husk) in the dark to encourage root development was initiated on July 29, 2010. The bulbs were planted in pots on August 30, 2010.

One bulb was planted in each 21-cm-diameter plastic pot filled with a growing medium (Premier PRO-MIX BX Mycorise; Premier Tech Ltd., Rivière-du-Loup, QC, Canada) containing 10 g of the same slow-release fertilizer as in Experiment 1. Starting on the day of planting, the plants were grown in three natural-light growth chambers (GCs; Koitotron S153A; Koito Industry Co., Yokohama, Japan) in which DT/NT was set to 20/15 $^{\circ}\text{C}$ to avoid the inhibited incipient growth of roots caused by high temperature. Seventeen plants were arranged in each of the three chambers.

From September 27 (28 days after planting; the visible flower bud stage), the DT (06:00-18:00) in each GC was set to 28 $^{\circ}\text{C}$, 24 $^{\circ}\text{C}$, or 20 $^{\circ}\text{C}$; the NT (18:00-06:00) was set to 15 $^{\circ}\text{C}$ in all three GCs. At the flowering stage of each experimental plot, 15 plants from each chamber were sampled. The relative humidity (RH) in the GCs was maintained at > 70%.

The sampled plants were separated into their various organs, and the size, fresh weight (FW), and dry weight (DW) of each organ and the LA were recorded. From the DW and LA values, the leaf area ratio (LAR: LA per unit of total plant DW in $\text{cm}^2 \text{ g}^{-1}$), the specific LA (SLA: LA per

unit of leaf DW in $\text{cm}^2 \text{ g}^{-1}$), the relative growth rate (RGR: rate of increase in total plant DW per unit of total plant DW in $\text{mg g}^{-1} \text{ day}^{-1}$), and the net assimilation rate (NAR: rate of increase in total plant DW per unit of LA in $\text{g m}^{-2} \text{ day}^{-1}$) were calculated as follows (Inamoto et al. 2010, Ushio et al. 2008):

$$LAR = \frac{A_{leaf}}{W_{total}} \quad [5]$$

$$SLA = \frac{A_{leaf}}{W_{leaf}} \quad [6]$$

$$RGR = \frac{\ln W_{total2} - \ln W_{total1}}{t_2 - t_1} \quad [7]$$

$$NAR = RGR \times \frac{W_{total2} + W_{total1}}{A_{leaf2} + A_{leaf1}} \quad [8]$$

where, A_{leaf} is the LA (cm^2 or m^2), W_{total} is the total plant DW (g or mg), W_{leaf} is the leaf DW (g), t is the time (day), and the subscript numerals represent the time (1 = initial, 2 = final).

4. Effect of NT on DM accumulation (Experiment 3)

The pre-rooting treatment of bulbs at 5 $^{\circ}\text{C}$ was initiated on May 27, 2009. The bulbs were planted in plastic pots on June 16, 2009, by using the same method as in Experiment 2. On the day of planting, the plants were arranged in the three GCs. From planting, the NT (18:00-06:00) in each GC was set to 25 $^{\circ}\text{C}$, 20 $^{\circ}\text{C}$, or 15 $^{\circ}\text{C}$; the DT (06:00-18:00) was set to 25 $^{\circ}\text{C}$ in all three GCs. At the flowering stage of each experimental plot, 15 plants from each chamber were sampled. The other methods used in this experiment were the same as in Experiment 2.

5. Temperature and light intensity data

Temperatures and RH during the experiments conducted in the greenhouse and growth chambers were monitored using a digital thermometer (RS-10; ESPEC MIC Co., Aichi, Japan). The Meteorological Observatory of the NARO Tohoku Agricultural Research Center in Morioka measured the ambient light intensity. Because these data were based on the radiant flux density (W m^{-1}), the values were converted into PPFD ($\mu\text{mol photons m}^{-2} \text{ s}^{-1}$) following the protocol described by Thimijan and Heins (1983). For Experiments 2 and 3, the daily light integral (DLI) supplied to the plants was calculated as the product of the natural light intensity and the transmittance of the chamber glass.

Results

1. Relationship between temperature and PR/RR at different plant developmental stages (Experiment 1)

The greenhouse daily temperatures were maintained at a minimum of 15°C and often exceeded 30°C in the day-time. The average temperature throughout the experiment was 19.3°C.

The stem lengths, numbers of leaves, total LA, LA per leaf, and FWs of the whole plant, aerial plant organs, and roots (Table 1) increased with the developmental stage. Only the FW of the bulb decreased. The total LA increased 2.6- and 1.6-fold from the leafing stage to the visible flower bud stage, and from the visible flower bud stage to the flowering stage, respectively. The temperature of the medium at measurement of the PR ranged from 10 to 20°C.

Fig. 1 shows the leaf temperature–PR relationships on a per-plant (A) and per-LA (B) basis at different developmental stages. The temperature–PR curves on a per-plant basis showed a general pattern of plant photosynthesis (Fig. 1A). As the leaf temperature increased, the PR increased, peaked, and then decreased. Simultaneously, the PR on a per-plant basis increased as the plants developed (Fig. 1A). From the cubic functions of the temperature–PR curves on a per-plant basis ($R^2 > 0.9$), the PR values/temperatures at the maximum PR were calculated as 0.32 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}/19.9^\circ\text{C}$ at the leafing stage, 1.58 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}/23.9^\circ\text{C}$ at the visible flower buds, and 1.97 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}/23.4^\circ\text{C}$ at the flowering stage (Fig. 1A). However, the PR on a per-LA basis at the leafing stage (8.67 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ at maximum) was not very low compared to that at the visible flower bud and flowering stages (Fig. 1B). The PR on a per-LA basis was highest at the visible flower bud stage (16.9 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ at maximum), and reduced slightly at the flowering stage (12.8 $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ at maximum) (Fig. 1B).

Fig. 2 shows the temperature–RR relationships on a per-plant basis in the aerial (A) and underground (B) parts

at different developmental stages, which were approximated well by quadratic and exponential curves, respectively ($R^2 > 0.9$). The RR of the aerial parts on a per-plant basis at 25°C was estimated at 0.11 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$ at the leafing stage, 0.20 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$ at the visible flower bud stage, and 0.19 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$ at the flowering stage. The RR of the underground parts on a per-plant basis at 25°C was estimated at 0.11 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$ at the leafing stage, 0.15 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$ at the visible flower bud stage, and 0.20 $\mu\text{mol CO}_2 \text{ plant}^{-1} \text{ s}^{-1}$ at the flowering stage.

3. Effect of DT on DM accumulation (Experiment 2)

The total FW at planting was 96.2 g (bulb: 68.2 g; shoot: 26.3 g; bulb roots: 1.7 g). The total DW at planting was 21.0 g (bulb: 18.3 g; shoot: 2.6 g; bulb roots: 0.1 g). Fig. 3A shows the PPF and DLI that were supplied to the plants during the experiment.

The date of sampling at the flowering stage was earlier for plants that were grown under high DT conditions (Table 2). At the flowering stage, the plants grown under high DTs of 28° and 24°C showed long stem length and low bulb FW (Table 2). DT did not affect the FWs of the whole plant, stem, flowers, and stem roots. The total plant DW increased between planting and flowering under all DT conditions, and was particularly large under low DT of 20°C (Fig. 4A). The flowers, stem roots, bulb roots, and bulbs of plants grown under low DT of 20°C also showed a high DW (Fig. 4A). The leaves, stems, and bulbs of plants grown under DTs of 24° and 20°C showed high DW/FW ratios (Table 2).

The plants that were grown under low DT of 20°C showed the lowest LAR and SLA (Table 3). The plants grown at 20°C also showed a high RGR and NAR from planting to flowering (Table 3).

4. Effect of NT on DM accumulation (Experiment 3)

The total FW at planting was 89.7 g (bulb: 64.9 g; shoot: 21.7 g; bulb roots: 3.1 g). The total DW at planting

Table 1. Stem lengths, number of leaves, leaf areas and fresh weights of ‘Siberia’ lily plants used to measure photosynthetic rate in Experiment 1

Developmental Stage	Stem length ¹⁾ (cm)	Number of leaves		Unfolded leaf area (cm ²)		Whole plant	Fresh weight (g)									
		Folded	Unfolded	Total	Per leaf		Aerial organs			Underground organs						
							Total	Leaves	Stem	Flower buds	Total	Stem	Bulb	Bulblets	Stem roots	Basal roots
Leafing	23.7±2.4 ²⁾	14.5±1.9	35.5±3.0	378±20	10.7±1.3	159±13	30.4±2.3	13.4±1.0	17.0±2.6	–	129±12	32.1±4.1	70.5±6.0	–	21.0±8.4	5.3±2.7
Visible flower buds	56.3±2.6	–	55.3±1.2	933±49	16.9±0.8	227±12	72.7±1.5	37.8±1.9	33.5±1.7	1.4±0.3	155±12	28.6±6.8	58.3±7.6	–	63.0±13.8	4.6±3.8
Flowering	78.8±1.7	–	61.3±4.6	1543±161	25.2±1.7	375±39	124.5±12.7	69.3±8.0	55.2±5.3	(49.1±3.9) ³⁾	250±42	27.9±2.8	30.4±2.1	2.6±1.2	178.2±45.5	11.3±9.1

The same plants as described in the previous report by Inamoto et al. (2015) were used.

¹⁾ Length between basal plate and peduncle of the top flower bud.

²⁾ Values are means (leafing stage n=6; the visible flower bud stage and flowering stage, n=4) ± SD.

³⁾ Flower buds were excised at photosynthesis measurement.

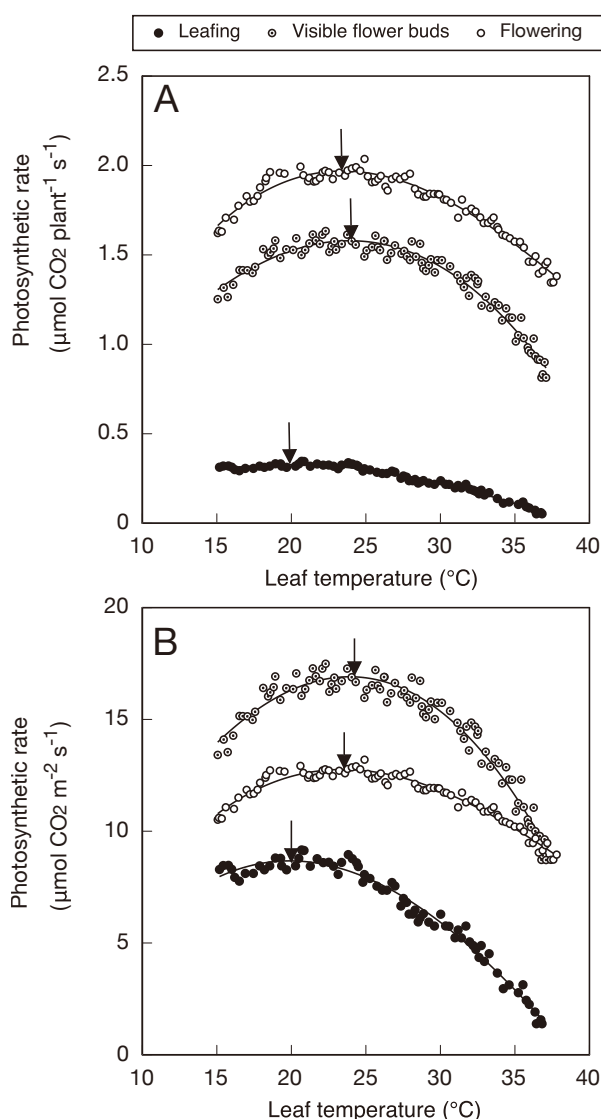


Fig. 1. Relationships between leaf temperature and net photosynthetic rate (PR) of Oriental hybrid lily ‘Siberia’ at three developmental stages (Experiment 1)

A: PR on a per-plant basis. B: PR on a per-leaf area basis.

Arrows indicate the maximum PR points of each curve.

was 21.5 g (bulb: 18.9 g; shoot: 2.3 g; bulb roots: 0.4 g). Fig. 3 B shows the PPFD and DLI that were supplied to the plants during the experiment.

The date of sampling at the flowering stage was earlier for the plants that were grown under high NT conditions (Table 4). At the flowering stage, the plants grown under low NT of 15°C showed the longest stem length and a high FW of the whole plant and stem (Table 4). NT did not affect the FWs of the flowers and stem roots. The total plant DW was particularly high under low NT of 15°C

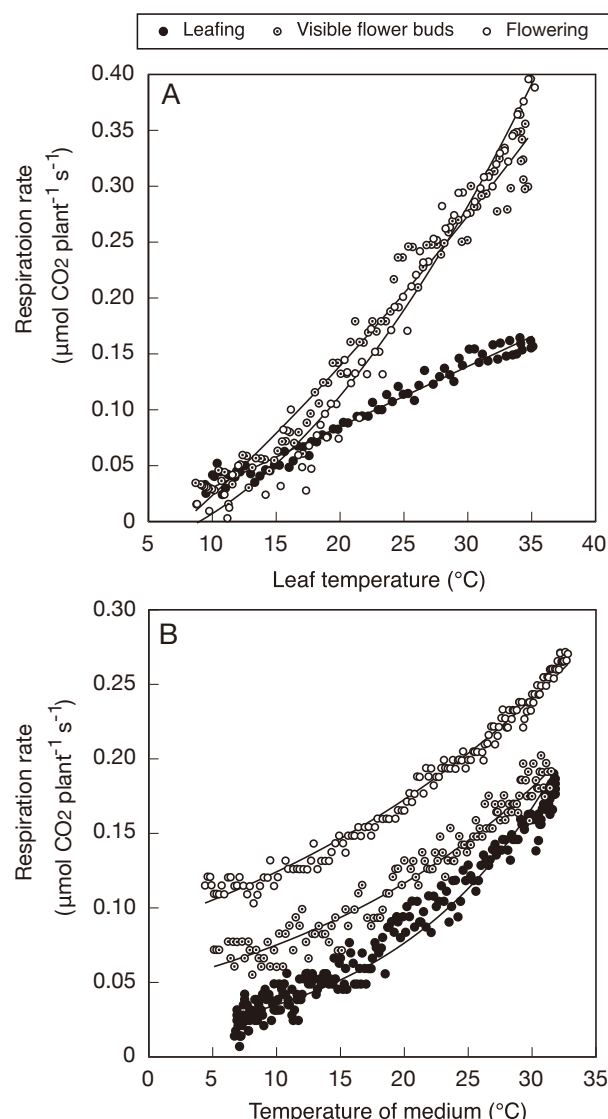


Fig. 2. Relationships between temperature and respiration rate (RR) in the aerial (A) and underground (B) parts of Oriental hybrid lily ‘Siberia’ at three developmental stages (Experiment 1)

(Fig. 4B). The stem of plants grown under low NT of 15°C also showed a high DW, and NT did not affect the DWs of flowers, leaves, stem roots, and bulb (Fig. 4B). The bulbs of plants grown under low NT of 15°C also showed high DW/FW ratios, and NT did not affect the DW/FW ratio of the stem (Table 4).

While the NT conditions did not affect the total LA at the flowering stage, the plants that were grown under low NT of 15°C showed a high LA per leaf (Table 5). The plants grown under low NT also showed a low LAR and SLA

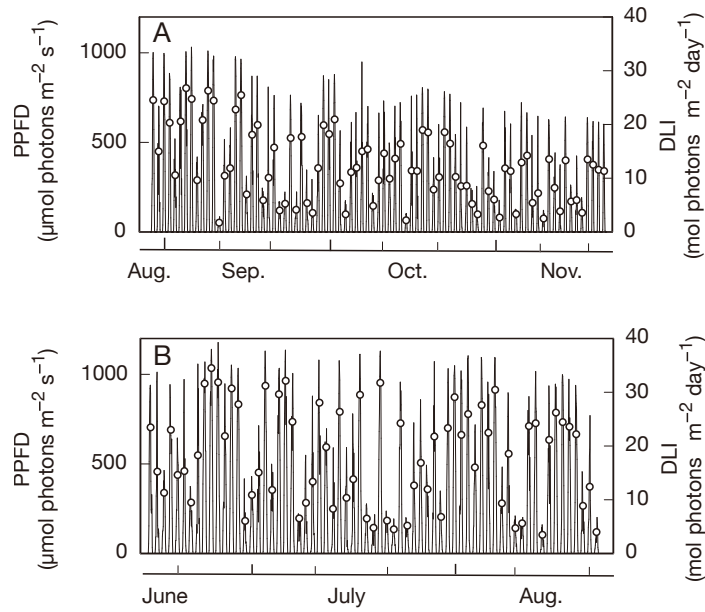


Fig. 3. PPFD and DLI supplied to the plants during Experiments 2 (A) and 3 (B)
 The solid lines and open circles indicate the PPFD and DLI, respectively. The data were multiplied by the transmittance of the glass of the growth chambers (70%).

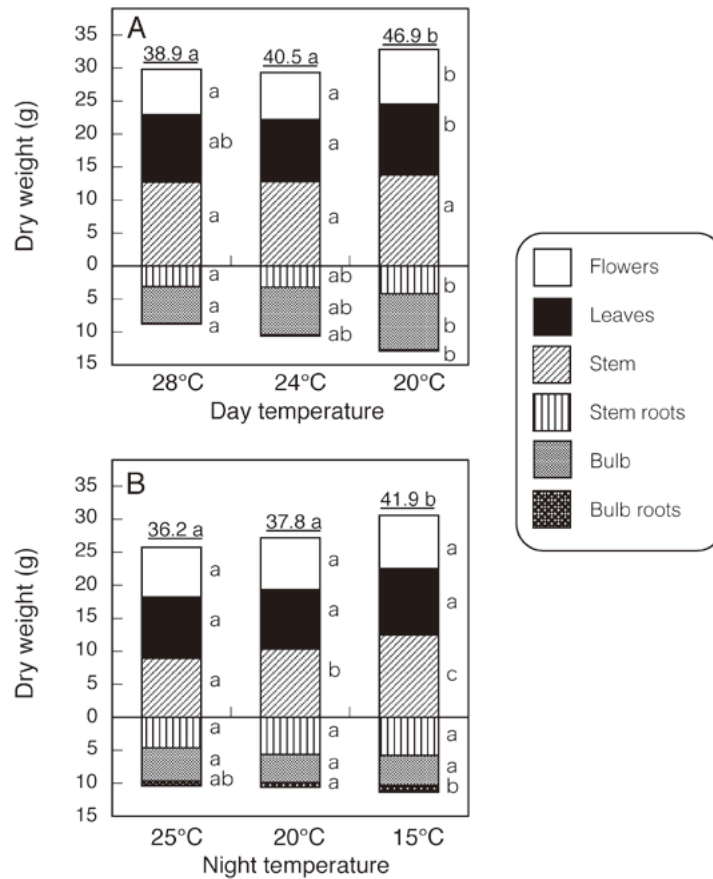


Fig. 4. Dry matter distribution of Oriental hybrid lily 'Siberia' grown under different day (A: Experiment 2) or night (B: Experiment 3) temperature conditions
 The values above each bar indicate the total dry weights. The mean separation within organs and total dry weight by Tukey's multiple range test at the 5% level.

Table 2. Stem lengths, numbers of leaves and flower buds, fresh weights, and dry/fresh weight ratios of ‘Siberia’ lily grown under 3 day temperature levels measured at the flowering stage

Day temperature	Date of sampling at the flowering stage ¹⁾	Days from plating to sampling	Stem length ²⁾ (cm)	Number		Fresh weight (g)						Dry/fresh weight ratio (g g ⁻¹)		
				Intact leaves ³⁾	Flower buds	Whole plant	Leaves	Stem	Flowers	Stem roots	Bulb	Leaves	Stem	Bulb
28°C	Nov. 12	74	108.2 b ⁴⁾	49.6 a	5.0 a	302.9 a	77.7 b	78.9 a	58.9 a	63.5 a	23.4 a	0.131 a	0.161 a	0.238 a
24°C	Nov. 15	77	105.3 b	48.9 a	4.8 a	293.5 a	66.6 a	73.6 a	56.0 a	67.4 a	27.2 ab	0.142 b	0.173 b	0.264 b
20°C	Nov. 20	82	100.2 a	50.3 a	4.9 a	325.4 a	76.6 b	79.4 a	64.1 a	69.0 a	32.5 b	0.140 b	0.174 b	0.259 b

From planting (August 30, 2010) to September 27 (the visible bud stage), all plants were grown under 20/15°C (day/night temperature). From September 27, the day temperature in each treatment was set to 28°C, 24°C, or 20°C; the night temperature was set to 15°C in all the treatments.

¹⁾: In 2010, all plants were sampled when several plants had well-colored flower buds in each plot.

²⁾: Length between basal plate and peduncle of the top flower bud.

³⁾: Intact leaves on the stem and peduncles.

⁴⁾: Values with the same letter are not significantly different by Tukey’s multiple range test at the 5% level.

Table 3. Leaf areas, leaf area ratios, specific leaf areas, relative growth rates, and net assimilation rates of ‘Siberia’ lily grown under 3 day temperature levels estimated at the flowering stage

Day temperature	Leaf area (cm ²)		LAR ¹⁾ (cm ² g ⁻¹)	SLA ²⁾ (cm ² g ⁻¹)	RGR ³⁾ from planting (mg g ⁻¹ day ⁻¹)	NAR ⁴⁾ from planting (g·m ⁻² day ⁻¹)
	Total	Per leaf				
28°C	1,956 b ⁵⁾	39.5 b	50.6 c	193 b	8.2 a	2.53 a
24°C	1,727 a	35.5 a	43.0 b	184 b	8.4 ab	3.02 a
20°C	1,837 ab	36.7 ab	39.4 a	172 a	9.7 b	3.62 b

¹⁾: Leaf area ratio.

²⁾: Specific leaf area.

³⁾: Relative growth rate.

⁴⁾: Net assimilation rate.

⁵⁾: Values with the same letter are not significantly different by Tukey’s multiple range test at the 5% level.

Table 4. Stem lengths, numbers of leaves and flower buds, fresh weights, and dry/fresh weight ratios of ‘Siberia’ lily grown under 3 night temperature levels measured at the flowering stage

Night temperature	Date of sampling at the flowering stage ¹⁾	Days from plating to sampling	Stem length ²⁾ (cm)	Number		Fresh weight (g)						Dry/fresh weight ratio (g g ⁻¹)		
				Intact leaves ³⁾	Flower buds	Whole plant	Leaves	Stem	Flowers	Stem roots	Bulb	Leaves	Stem	Bulb
25°C	Aug. 11	56	79.7 a ⁴⁾	59.7 b	6.2 a	316.3 a	75.3 ab	61.2 a	62.2 a	81.4 a	26.1 b	0.114 a	0.147 a	0.192 a
20°C	Aug. 17	62	91.2 b	53.9 a	5.5 a	323.5 a	69.3 a	67.7 a	64.5 a	90.1 a	21.9 ab	0.130 c	0.153 a	0.194 ab
15°C	Aug. 21	66	99.6 c	55.5 ab	5.8 a	371.2 b	81.5 b	84.0 b	68.8 a	100.4 a	21.0 a	0.123 b	0.149 a	0.214 b

From planting (June 16, 2009), the night temperature in each treatment was set to 25°C, 20°C, or 15°C; the day temperature was set to 25°C in all the treatments.

¹⁾: In 2009. All plants were sampled when several plants had well-colored flower buds in each plot.

²⁾: Length between basal plate and peduncle of the top flower bud.

³⁾: Intact leaves on the stem and peduncles.

⁴⁾: Values with the same letter are not significantly different by Tukey’s multiple range test at the 5% level.

(Table 5). The RGRs and NARs of plants that were grown under the three NT conditions did not differ significantly at the 5% level (Table 5).

Discussion

In Experiment 1, the optimum temperature for the PR of ‘Siberia’ lily was estimated within the range of 20–25°C at the three plant developmental stages under PPFD of 700 to 850 $\mu\text{mol photons m}^{-2}$ (Fig. 1). We previously observed that light intensity of 700 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ PPFD was close to light saturation and sufficient for photosynthesis in ‘Siberia’ (Inamoto et al. 2015). Compared to other major ornamental plants, the optimum temperature of PR in ‘Siberia’ lily was lower than that in rose (Ushio et al. 2008), chrysanthemum (Körner et al. 2009) and eustoma (Imamura et al. 1989) and higher than that in carnation (Enoch & Hurd 1997), as estimated under sufficient light intensity and ambient CO_2 concentrations. The results of the past few studies conducted on photosynthesis in Oriental hybrid lilies do not agree. Yamaguchi and Kamijo (1991) reported an optimum photosynthesis temperature of 15 to 20°C for ‘Star Gazer’, while Sorrentino et al. (1997) reported 31 to 32°C for ‘Casa Blanca’. Considering the differences among their data and our present data, an estimation above 25°C as the optimum temperature for PR should be too high when taking the results of Experiment 2 into account.

In Experiment 2, in which plants were grown under the same NT of 15°C, the total DW at the flowering stage was greater in the plants grown under low DT of 20°C than in the plants grown at 28°C or 24°C (Fig. 4A). The RGRs and NARs of the plants grown at low DT of 20°C were higher (Table 3) from the planting stage to the flowering stage. The NAR is an indicator of photosynthetic efficiency

per LA throughout the plant growth period. From the results of Experiment 1, DT of 28°C is obviously higher than the optimum temperature for PR. The total DW (Fig. 4A) and NAR (Table 3) of the plants that were grown under DT of 20°C was higher than those of plants grown under DT of 24°C, although both DT values are within the temperature range suitable for photosynthesis (Experiment 1). The reasons for this result may be that the natural light environment fluctuated and that sufficient light intensity for PR was not always supplied to the plants during Experiment 2 (Fig. 3A). As shown in carnation (Enoch & Hurd 1977) and rose (Jiao et al. 1991), the optimum temperature for PR decreases as the light intensity decreases. The stems and bulb roots of plants grown under high DT of 28°C had a significantly low DW (Fig. 4A). Inhibited root development was observed under the combination of high DT and NT in our previous report (Inamoto et al. 2013). Still, the high DT condition of 28°C in Experiment 2 may have caused some development and/or functional disorder of the roots that affected the absorption of water and mineral nutrition. This point demands further investigation.

In Experiment 1, the order and degree of the temperature–PR response curves obtained for each plant developmental stage differed between the per-plant basis and per-LA basis (Fig. 1) as was shown in our previous report regarding the light-PR response curves (Inamoto et al. 2015). The PR on a per-plant basis just after shoot emergence (the leafing stage) was very low, but rapidly increased at the visible flower bud and flowering stages (Fig. 1A). The PR on a per-LA basis peaked at the visible flower bud stage and was diminished at the flowering stage (Fig. 1B). Therefore, the highest PR on a per-plant basis may be caused by a rapid increase in LA due to the plant growth (Table 1) of Oriental hybrid lilies.

Table 5. Leaf areas, leaf area ratios, specific leaf areas, relative growth rates, and net assimilation rates of ‘Siberia’ lily grown under 3 night temperature levels estimated at the flowering stage

Day temperature	Leaf area (cm^2)		LAR ¹⁾ ($\text{cm}^2 \text{g}^{-1}$)	SLA ²⁾ ($\text{cm}^2 \text{g}^{-1}$)	RGR ³⁾ from planting ($\text{mg g}^{-1} \text{day}^{-1}$)	NAR ⁴⁾ from planting ($\text{g} \cdot \text{m}^{-2} \text{day}^{-1}$)
	Total (cm^2)	Per leaf (cm^2)				
25°C	1,829 a ⁵⁾	30.7 a	51.0 b	199 b	9.2 a	2.93 a
20°C	1,714 a	31.8	45.6 a	191 ab	8.9 a	3.11 a
15°C	1,862 a	33.6 b	44.6 a	187 a	10.0 a	3.41 a

¹⁾: Leaf area ratio.

²⁾: Specific leaf area.

³⁾: Relative growth rate.

⁴⁾: Net assimilation rate.

⁵⁾: Values with the same letter are not significantly different by Tukey’s multiple range test at the 5% level.

In Experiment 3, in which the plants were grown under the same DT of 25°C, the total DW at the flowering stage was greater in the plants grown under low NT of 15°C than in the plants grown at 25°C or 20°C (Fig. 4B). This difference may be the result of reduced DM consumption by respiration under the low NT condition, as illustrated by the temperature–RR relationships in Experiment 1 (Fig. 2). However, contrary to expectations, the RGRs and NARs from planting to flowering did not differ significantly among the three NT plants (Table 5). Concerning this point, two possible factors are conceivable by considering Formula [7]. For one thing, NT has a great effect on the flowering date (Tables 2 and 4). Conversely, NT has a small effect on DW (Fig. 4A and B) as compared with that by DT. Hurd and Enoch (1976) showed that NT only had a slight effect on the growth rate of carnation plants. Moreover, these authors reported similar rates of photosynthesis and transpiration at 20°C for carnation plants grown under different NT values.

Compared to the data in the present experiments, in which DT and NT changed independently, ‘Siberia’ lily plants that were grown under three DT/NT combinations of 28/23°C, 24/19°C, and 20/15°C showed great differences in DM accumulation at flowering in our previous experiment (Inamoto et al. 2013). This result suggests that the positive effects on DM accumulation by promoting photosynthesis at low DT and suppressing dark respiration at low NT are additive.

The stem length and stiffness are important characteristics of the cut flower quality. From the perspective of quality, the plants that were grown under low DT of 20°C in Experiment 2 (Table 2) and low NT of 15°C in Experiment 3 (Table 4) produced desirable cut flowers with satisfactory DM accumulation (Fig. 4). However, regarding stem length, high DT (Table 2) and low NT (Table 4) conditions, especially the latter, produced a significantly long stem length in lily plants, which may be a result of the "positive DIF effect" (Blom & Kerec 2003, Erwin et al. 1989). The elongated stems of plants grown under high DT of 28°C might be considered "succulent elongation" because such growth was accompanied by a low DW/FW ratio (Table 2). However, the plants grown under low NT of 15°C maintained the DW/FW ratio (Table 4), indicating that sufficient DM accumulation supported stem elongation (Fig. 4B). The high DW/FW ratios of the leaves of the plants grown under low DT and NT conditions also correspond to stout cut flowers (Tables 2 and 4). Moreover, their low SLA implies thick and massive leaves (Tables 3 and 5).

The present results support the advantages of culturing Oriental lilies under cool DT and/or NT conditions, especially in summer. In growing facilities, a heat pump cooling system is expected to become available for cooling NT (Ninomiya et al. 2012). Evaporative cooling systems,

such as pad-and-fan and fogging systems, are also effective for cooling DT (Arbel et al. 1999). However, it is difficult to reduce the temperature of open fields for lily production. Shading is a simple and passive solution, but one should be careful not to disturb photosynthesis by applying the reference data of the light-PR relationships (Inamoto et al. 2015). Though the soil temperature is stable and difficult to control, organic mulch is a practical and low cost passive method (Chalker-Scott 2007).

References

- Abe, J. et al. (1998) Quality improvement in Oriental lilies using pre-rooting method. *Tohoku Agri. Res.*, **51**, 249-250 [In Japanese].
- Arbel, A. et al. (1999) Performance of a fog system for cooling greenhouses. *J. Agric. Eng. Res.*, **72**, 129-136.
- Beattie, D. J. & White, J. W. (1993) *Lilium* –Hybrids and species. *The Physiology of Flower Bulbs*. Elsevier, Amsterdam, Netherlands, 423-454.
- Blom, T. J. & Kerec, D. R. (2003) Effects of far-red light/temperature DIF and far-red light/temperature pulse combinations on height of lily hybrids. *J. Hort. Sci. & Biotech.*, **78**, 278-282.
- Chalker-Scott, L. (2007) Impact of mulches on landscape plants and the environment –a review. *J. Environ. Hort.*, **25**, 239-249.
- Enoch, H. Z. & Hurd, R. G. (1977) Effect of light intensity, carbon dioxide concentration, and leaf temperature on gas exchange of spray carnation plants. *J. Exp. Bot.*, **28**, 84-95.
- Erwin, J. E. et al. (1989) Thermomorphogenesis in *Lilium longiflorum*. *Amer. J. Bot.*, **76**, 47-52.
- Hurd, R. G. & Enoch, H. Z. (1976) Effect of night temperature on photosynthesis, transpiration, and growth of spray carnations. *J. Exp. Bot.*, **27**, 695-703.
- Imamura, H. et al. (1989) Improvement of cut flower productivity by efficient use of solar radiation. 4. Photosynthetic response of *Eustoma grandiflorum*. *J. Japan. Soc. Hort. Sci.*, **58** (suppl. 2), 458-459 [In Japanese].
- Inamoto, K. et al. (2000) Effects of duration of bulb chilling on dry matter distribution in hydroponically forced tulips. *Sci. Hort.*, **85**, 295-306.
- Inamoto, K. et al. (2013) Influence of growing temperature on dry matter accumulation in plant parts of ‘Siberia’ Oriental hybrid lily. *JARQ.*, **47**, 435-441.
- Inamoto, K. et al. (2015) Influence of light intensity on the rate of photosynthesis and dry matter accumulation in Oriental hybrid lily ‘Siberia’ at different developmental stages. *J. Hort. Sci. & Biotech.*, **90**, 259-266.
- Jiao, J. et al. (1991) Influence of radiation and CO₂ enrichment on whole plant net CO₂ exchange in roses. *Can. J. Plant Sci.*, **71**, 245-252.
- Körner, O. et al. (2009) Quantification of temperature, CO₂, and

- light effects on crop photosynthesis as a basis for model-based greenhouse climate control. *J. Hort. Sci. & Biotech.*, **84**, 233-239.
- Ninomiya, C. et al. (2012) Effect of low night temperature management by heat-pump system on growth and flowering of Oriental hybrid lily 'Siberia' in summer culture. *Hort. Res. (Japan)*, **11**, 537-543 [In Japanese with English summary].
- Righetti, T. et al. (2007) Assessments CO₂ assimilation on a per-leaf-area basis are related to total leaf area. *J. Amer. Soc. Hort. Sci.*, **132**, 230-238.
- Sorrentino, G. et al. (1997) Effect of shading and air temperature on leaf photosynthesis, fluorescence and growth in lily plants. *Sci. Hort.*, **69**, 259-273.
- Thimijan, R. W. & Heins, R. D. (1983) Photometric, radiometric, and quantum light units of measure: a review of procedure for interconversion. *HortSci.*, **18**, 818-822.
- Ushio, A. et al. (2008) Effects of temperature on photosynthesis and plant growth in the assimilation shoots of a rose. *Soil Sci. Plant Nutr.*, **54**, 253-258.
- Yamaguchi, T. & Kamijo, M. (1991) Effect of reflective film mulching on the growth and flowering of Oriental hybrid lily cv. 'Star Gazer'. *Yasai-Chagyo Sikenjyo Kakibu Kenkyunepou* (National Research Institute of Vegetables, Ornamental Plants and Tea), **4**, 41-42 [In Japanese].
- Wang, Y. (1990) Growth and leaf photosynthesis of *Lilium longiflorum* Thunb. 'Nellie White' in response to partial defoliation after anthesis. *Acta Hort.*, **266**, 197-204.
- Zhang, Y. J. et al. (2011a) Effect of water stress on leaf photosynthesis, chlorophyll content, and growth of oriental lily. *Russ. J. Plant Physiol.*, **58**, 844-850.
- Zhang, Y. J. et al. (2011b) Light intensity affects dry matter, photosynthesis and chlorophyll fluorescence of Oriental lily. *Philipp. Agric. Sci.*, **94**, 232-238.