

---

# Low Temperature and Weak Light Affect Greenhouse Tomato Growth and Fruit Quality

Fang Xiaoa<sup>1</sup>, Zaiqiang Yang<sup>1,2,\*</sup>, Liyun Zhua<sup>1</sup>

<sup>1</sup>Collaborative Innovation Center on Forecast and Evaluation of Meteorological Disasters, Nanjing University of Information Science & Technology, Nanjing, China

<sup>2</sup>Jiangsu Key Laboratory of Agricultural meteorology, Nanjing University of Information Science & Technology, Nanjing, China

## Email address:

yzq6751@163.com (Zaiqiang Yang)

\*Corresponding author

## To cite this article:

Fang Xiaoa, Zaiqiang Yang, Liyun Zhua. Low Temperature and Weak Light Affect Greenhouse Tomato Growth and Fruit Quality. *Journal of Plant Sciences*. Vol. 6, No. 1, 2018, pp. 16-24. doi: 10.11648/j.jps.20180601.14

**Received:** January 9, 2018; **Accepted:** January 22, 2018; **Published:** March 7, 2018

---

**Abstract:** To investigate the effects of low temperature (LT) and weak light (WL) on tomato (*Solanum lycopersicum* L., cv.) during flowering and fruit-setting periods, a controlled experiment was conducted. Two levels of day/night temperature and PAR were set: S1 (18/8°C, 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ), S2 (12/2°C, 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ), S3 (18/8°C, 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ), and S4 (12/2°C, 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ), taking 28/18°C and 600  $\mu\text{mol m}^{-2} \text{s}^{-1}$  as control (CK). The results showed that during stress stage, the Chlorophyll (Chl) *a*, photosynthetic rate at irradiation saturation (*P*<sub>max</sub>), light saturation point, stomatal conductance, stomatal limitation value, maximal photochemical efficiency of PSII (Fv/Fm), electron transport rate of PSII, and catalase activity of S1, S2, S3, and S4 were lower than that of CK, while the Chl *b*, carotenoid, light compensation point, superoxide dismutase (SOD), and malondialdehyde (MDA) were opposite. Vitamin C, soluble solid, soluble protein, and lycopene were lower than that of CK, while organic acid was opposite. Plant height and stem diameter significantly correlated with Chl and *P*<sub>max</sub>. After 25 d of recovery, the Fv/Fm, SOD, and MDA for S1 and S2 almost could recover to CK level, but the values for S3 and S4 could not recover to CK level.

**Keywords:** Antioxidant Enzyme, Chlorophyll Fluorescence, Lipid Peroxidation, Poor Light, *Solanum lycopersicum*

---

## 1. Introduction

Tomato (*Solanum lycopersicum* L., cv.) is one of the most important protected-cultivation horticultural crops in northern China, and it is widely cultivated in single-slope solar greenhouse during winter and spring [1].

Temperature and light have been proven to be two important environmental factors affecting plant growth [2], photosynthesis [3], and fruit growth and development [4]. Temperature limits plants growth and distribution [5], light intensity affects plants characteristics [6]. The morphology [7], chlorophyll (Chl) content [8], biochemistry [9], and other physiological parameters [10-12] were affected by light. The combination of low temperature (LT) and weak light (WL) affected tomato cultivation severely. LT and WL reduced photosynthetic efficiency, inhibited biomass production, and increased reactive oxygen species (ROS) and membrane lipid

peroxidation [13-14]. LT and WL also resulted in a dramatic drop in net photosynthetic rate (PN) of *Prunus armeniaca*, further caused a damage to PSII [15].

Tomato is sensitive to temperature and light, the optimal day/night temperature for tomato growth is 25/16°C to 28/18°C, and the optimal light intensity is about 600  $\mu\text{mol m}^{-2} \text{s}^{-1}$  [16-17]. However, in winter of northern China, tomato cultivation is frequently subjected to LT [below 20 /10°C] and WL [below 400  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ], due to clouds, rain, fog and haze [1]. Previous studies just focused on the effects of LT and WL on tomato seedlings, but less about the interaction of LT and WL in flowering and fruits-setting stages. Therefore, we examined the effects of combined LT and WL on growth, photosynthesis, Chl fluorescence, and antioxidants enzyme in tomato leaves, as well as vitamin C (Vc), soluble solid, soluble protein, lycopene, and organic acid in tomato fruits in flowering and fruit-setting stages. The objective in this study was to provide information for improving greenhouse tomato production.

## 2. Materials and Methods

### 2.1. Plant Materials and Stress Treatments

The experiment was conducted from 16th November to 7th April in 2015 at Nanjing University of Information Science & Technology, China. Tomato (*Solanum lycopersicum* L., cv. Jinfen 5) seeds were germinated on filter paper containing distilled water in the dark at 29°C for 2 days. Two germinated seedlings were transferred individually to each pot (25 × 20 cm, diameter × height) containing 1:1 sand: peat by volume and grown in a glass greenhouse at 28/18°C D/N, and the relative humidity (RH) ranged from 60% to 70%. The plants were watered as needed, N, Ca, and K fertilization in the form of urea, calcium dihydrogen phosphate monohydrate (Ca (H<sub>2</sub>PO<sub>4</sub>) 2·H<sub>2</sub>O), and potassium chloride (KCl) at the rate equivalent to 0.78, 0.64 and 1.11 g pot<sup>-1</sup> according to Yang [18].

The LT and WL treatments were started when plants height were approximately about 25 cm and flower buds of the first inflorescence appeared. Plants at the first

inflorescence stage were transferred to five identical growth chambers (A1000, Conviron, Winnipeg, Canada) to begin the stress treatments, and each treatment had 3 pots. Temperature and RH inside the chambers were controlled automatically to reach the set points according to real-time monitoring results of sensors. The fluctuations were less than 0.5°C and 3% of reading for air temperature and RH inside the chambers, respectively. The photoperiod was set to 12/12 h D/N (07:00–19:00). Chambers RH maintained at 75%. Two D/N temperature regimes (18/8 and 12/2°C) were combined with two photosynthetic active radiation (PAR) levels (200 and 80 μmol·m<sup>-2</sup>·s<sup>-1</sup>). Control (CK) plants were maintained at 28/18°C D/N, 600 μmol·m<sup>-2</sup>·s<sup>-1</sup> PAR, and 12/12 h D/N photoperiod. Stress treatments were lasted for 10 days and then all plants were recovered for 25 days (Table 1). Plants were watered with tap water manually throughout the experiment to achieve the optimum soil moisture (31–33% soil volumetric moisture content) by soil moisture meter (DSMM500, General, USA).

Table 1. Day/night temperature and light intensity during stress and recovery stage.

Treatment	Stress		Recovery	
	day/night temperature (°C)	light (μmol m <sup>-2</sup> s <sup>-1</sup> )	day/night temperature (°C)	light (μmol m <sup>-2</sup> s <sup>-1</sup> )
CK	28/18	600	28/18	600
S1	18/8	200	28/18	600
S2	12/2	200	28/18	600
S3	18/8	80	28/18	600
S4	12/2	80	28/18	600

### 2.2. Plant Growth Measurements

Plant height and stem diameter were measured using a ruler and a Vernier caliper, respectively, and leaf area was measured using digital camera [19]. Three repetitions were measured for each treatment once every two and four days during the stress period.

### 2.3. Chlorophyll (Chl) Measurements

The 5th–8th leaves from the top of plants were placed in a glass tube with 4.5:4.5:1 acetone: ethanol: water by volume for 48 hours. Absorbance was measured using spectrophotometry (Cary 50 conc UV-VIS, Varian, Victoria, Australia) at 663 nm, 646 nm, and 470 nm. The Chl *a*, Chl *b*, and Car content were calculated according to Wellburn [20]. Three repetitions were measured for each treatment once every two and four days during the stress period.

### 2.4. Photosynthetic Parameters Measurements

The PN, stomatal conductance (gs), intercellular CO<sub>2</sub> concentration (Ci), and ambient CO<sub>2</sub> concentration (Ca) were measured between 09:00 and 11:00 am using a portable photosynthesis measurement system (LI-6400, LI-COR Bioscience, Lincoln, NE, USA). Three repetitions were measured for each treatment once every two and four days

during the stress and every five days during the recovery period. Stomatal limitation value (*Ls*) was calculated:  $Ls = 1 - Ci/Ca$  [21]. Light response curves were determined using a photosynthetic active radiation gradient: 2000, 1800, 1500, 1200, 1000, 800, 500, 200, 100, 50, 20, and 0 μmol m<sup>-2</sup> s<sup>-1</sup>. The leaf chamber temperature was 25°C, and the CO<sub>2</sub> concentration was 390 μmol (CO<sub>2</sub>) mol<sup>-1</sup>. The photosynthetic rate was measured automatically at each irradiation level after 3–5 min light exposure. PN/PPFD curves were modeled by fitting nonrectangular hyperbola to data [22]

Where  $\alpha$  is the initial slope or apparent photosynthetic quantum yield (PN/PPFD at low PPFD), PPFD is the photosynthetic photon flux density [μmol (photon) m<sup>-2</sup> s<sup>-1</sup>], *P*<sub>max</sub> is the photosynthetic rate at irradiation saturation [μmol (CO<sub>2</sub>) m<sup>-2</sup> s<sup>-1</sup>], *k* is the curve convexity (dimensionless), and *RD* is the dark respiration rate [μmol (photon) m<sup>-2</sup> s<sup>-1</sup>].

### 2.5. Chlorophyll Fluorescence Measurements

The second fully expanded leaf from the bottom of each plant was used to measure chlorophyll fluorescence parameters by portable fluorimeter (FMS-2, Hansatech, Norfolk, U.K.) once every two and four days during the stress and every five days during the recovery periods. The irradiation adaptation fluorescence parameters, *Fm'*, *Fo'*, and

F<sub>s</sub> were determined under actinic irradiation of 600 μmol (photon) m<sup>-2</sup> s<sup>-1</sup>. And then, the dark-adaptation fluorescence parameters, F<sub>m</sub> and F<sub>o</sub> were determined after the leaves were dark-adapted for 30 min, measurements were repeated three times for each treatment. The F<sub>v</sub>/F<sub>m</sub> and electron transport rate (ETR) were calculated according to Zhang and Gao [23].

### 2.6. Antioxidant Enzyme Measurements

The same leaves as those used in photosynthesis measurements were sampled and immediately frozen in liquid nitrogen and stored at -40°C for further enzyme analyses [24]. Superoxide dismutase (SOD, EC 1.15.1.1) activity was measured using extracts obtained from 300 mg of fresh leaf. The fresh leaf was homogenized in an extraction buffer containing 50 mM phosphate buffer (pH 7.8), 0.1 % (w/v) ascorbate, and 0.05 % (w/v) β-mercaptoethanol. The 3 mL assay mixture contained 50 mM phosphate buffer (pH 7.8), 9.9 mM methionine, 0.025% (w/v) nitroblue tetrazolium chloride (NBT), and 0.0044 % (w/v) riboflavin. Catalase (CAT, EC 1.11.1.6) activity was determined as the decrease in absorbance at 240 nm and the reaction mixture containing 1.5 mL 50 mM phosphate buffer (pH 7.8), 0.3 mL 100 mM H<sub>2</sub>O<sub>2</sub>, and 0.2 mL enzyme extract [25]. One unit of the CAT activity was defined as the reduction in absorbance at 240 nm per min. The extinction coefficient was 36 mm<sup>-1</sup> cm<sup>-1</sup>. SOD and CAT activities were expressed as unit g<sup>-1</sup> (FM). Three repetitions were measured for each treatment once every two and four days during the stress and every five days during the recovery period.

### 2.7. Lipid Peroxidation Measurement

Lipid peroxidation was estimated in terms of malondialdehyde (MDA) content. The MDA content was determined according to Zhao [26]. Fresh leaves (1.0 g) were ground in 10% trichloroacetic acid and then centrifuged at 3000 g for 10 min. Two mL of the supernatant was mixed with 2 mL of 0.6% thiobarbituric acid (TBA) and incubated for 30 min at 100°C to form a MDA-TBA<sub>2</sub> adduct. The mixture was cooled rapidly in an ice bath. After centrifugation at 5000 g for 10 min, the absorbance was measured at 450 nm, 532 nm, and 600 nm. Lipid peroxidation was expressed as μmol g<sup>-1</sup> (FM) using the following formula: MDA [μmol g<sup>-1</sup> (FM)] = 6.45 (A<sub>532</sub> - A<sub>600</sub>) - 0.56A<sub>450</sub>, where A<sub>532</sub>, A<sub>600</sub>, and A<sub>450</sub> are absorbance measured at 532 nm, 600 nm, and 450 nm. Three repetitions were measured for each treatment once every two and four days during the stress and every five days during the recovery period.

### 2.8. Fruits Quality Determination

All marketable tomato fruits from each treatment were cut into small slices after 25 d of recovery, and mixed together. Samples were pressed through cheese cloth to extract the juice, which was analyzed for fruit quality.

Vitamin C (ascorbic acid + dehydroascorbic acid) was determined by HPLC [27]. Forty mL of tomato juice were homogenized with 25 mL of extraction solution (30 g L<sup>-1</sup> metaphosphoric acid and 80 g L<sup>-1</sup> acetic acid). The resulting mixture was centrifuged at 10000 g for 10 min at 4°C, and the supernatant was filtered and adjusted to 75 mL with distilled water. Samples were filtered through a 0.45 μm membrane filter and duplicate aliquots of 20 μL for each extract were analyzed by HPLC. Results were expressed as milligrams of ascorbic acid per liter of tomato juice.

Fruit soluble solids were measured with a portable Sper Scientific 300003 refractometer (Sper Scientific Ltd., Scottsdale, AZ, USA) standardized with distilled water [28].

Soluble protein was assayed by dye-binding [29]. The homogenates were centrifuged at 15000 g for 15 min. All steps were performed at 4°C. The protein concentration was determined by a UV/VIS spectrophotometer (*Hitachi U-2000*, Tokyo, Japan) at 595 nm.

Lycopene content was measured according to the improved national standard method. Added 0.1–0.2g samples into 100μL methanol, stirred thoroughly with a glass rod to extract the yellow pigments, and then centrifuged to the colorless filtrate. The lycopene was extracted with toluene repeatedly, filtered to filtrate colorless in 50 mL brown volumetric flask. The absorbance for lycopene was determined by a visible spectrophotometer (Model 722, Shanghai, China) at 485 nm.

Organic acid was estimated by enzymatic procedures, employing malate dehydrogenase (EC 1.1.1.37) and glutamic-oxaloacetic transaminase (EC 2.6.1.1) with NAD reduction for malate, and citrate lyase (EC 4.1.3.6), malate dehydrogenase, and lactate dehydrogenase (EC 1.1.1.27) with NADH oxidation for citrate [30].

*Statistical analysis.* Statistical analyses were performed using *SPSS 16.0* statistical software (*SPSS Inc.*, Chicago, IL, USA). Significant differences among different plant treatments reported at P < 0.05, if not indicated otherwise. The reported data were the mean ± standard deviation (SD) of three biological replications.

## 3. Results

### 3.1. Plant Growth

The plant height and stem diameter increased gradually with an increase of stress days, but plant height and stem diameter declined with a decrease of temperature and light (Table 2). Plant height at 10 d after treatment (dat) under CK, S1, S2, S3, and S4 was significant difference with that at 0 d. Compared with CK, the stem diameter for S1, S2, S3, and S4 decreased by 3.3%, 5.4%, 11.6%, and 21.7% at 10 dat, but there was no significant difference. However, alterations in leaf area were contrary to plant height and stem diameter, the leaf area under S1, S2, S3, and S4 was larger than that under CK.

**Table 2.** Effects of low temperature and weak light on tomato growth.

	Treatment	Days under stress			
		0	2	6	10
Plant height (cm)	CK	35.11 ± 1.34c	39.75 ± 1.86bc	44.39 ± 2.69ab	46.07 ± 3.14a
	S1	33.36 ± 2.76c	37.33 ± 1.79bc	40.86 ± 2.99ab	43.71 ± 01.40a
	S2	32.11 ± 2.47b	34.18 ± 3.01ab	36.63 ± 3.41ab	40.84 ± 3.07a
	S3	30.50 ± 2.38b	33.62 ± 1.53b	35.18 ± 1.73ab	38.63 ± 2.31a
	S4	30.20 ± 1.63c	30.49 ± 2.06bc	34.35 ± 2.57ab	36.47 ± 2.53a
Stem diameter (mm)	CK	6.42 ± 0.31a	7.01 ± 0.33a	7.06 ± 0.45a	7.19 ± 0.43a
	S1	6.45 ± 0.18a	6.69 ± 0.30a	6.75 ± 0.29a	6.96 ± 0.12a
	S2	6.38 ± 0.30a	6.57 ± 0.28a	6.65 ± 0.33a	6.82 ± 0.35a
	S3	5.95 ± 0.27a	6.12 ± 0.20a	6.38 ± 0.23a	6.44 ± 0.23a
	S4	5.62 ± 0.24a	5.76 ± 0.34a	5.84 ± 0.32a	5.91 ± 0.47a
Leaf area (cm <sup>2</sup> )	CK	5.06 ± 0.41a	5.15 ± 0.38a	5.25 ± 0.41a	5.30 ± 0.43a
	S1	5.38 ± 0.25b	5.54 ± 0.34ab	6.09 ± 0.30ab	6.27 ± 0.33a
	S2	5.26 ± 0.33a	5.35 ± 0.35a	5.50 ± 0.29a	5.64 ± 0.30a
	S3	6.02 ± 0.26a	6.12 ± 0.27a	6.43 ± 0.31a	6.65 ± 0.37a
	S4	5.85 ± 0.29a	6.03 ± 0.39a	6.19 ± 0.37a	6.49 ± 0.39a

Note: Small letters indicate significance of  $P < 0.05$  by Duncan's test within each row. Results are presented as mean  $\pm$  SD ( $n = 3$ ). CK, control; S1, 18/8°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S2, 12/2°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S3, 18/8°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S4, 12/2°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR.

### 3.2. Chl Content

The Chl *a* content for S1, S2, S3, and S4 declined significantly from 0 to 10 dat (Table 3). The Chl *a* content at 10 dat significantly decreased by 54.3%, 25.8%, 20.6%, and 18.1% under S1, S2, S3, and S4, compared with CK. The Chl *b* content for S1, S2, S3, and S4 decreased, compared to that of

control. Meanwhile, there was significant difference between CK and S1, S2, S3, and S4, except for that at 10 dat. With an increase of treatment duration (6 and 10 dat), the Car content from S1 to S4 increased gradually. The Car content for S1, S2, S3, and S4 was significantly different from that of CK at 10 dat.

**Table 3.** Effects of low temperature and weak light on chlorophyll content in tomato leaves.

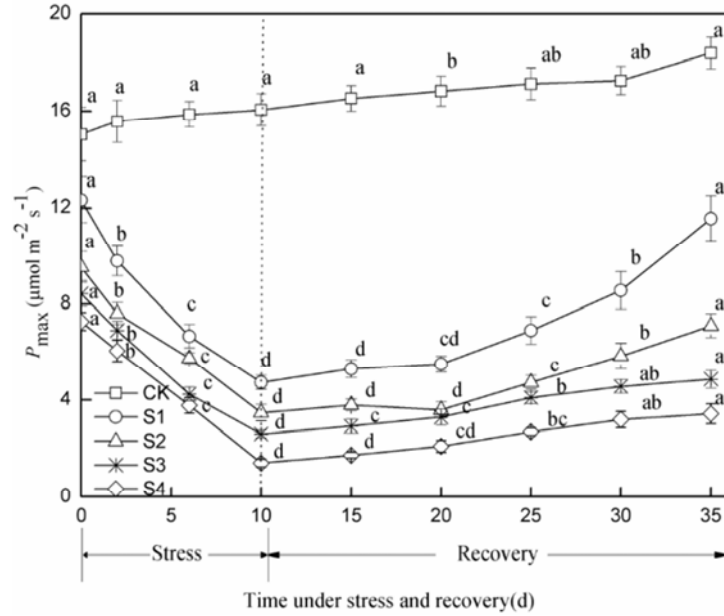
	Treatment	Days under stress			
		0	2	6	10
Chl <i>a</i> content [ $\text{mg g}^{-1}$ (FM)]	CK	2.46 ± 0.11c	2.62 ± 0.10bc	2.87 ± 0.05b	3.26 ± 0.19a
	S1	2.44 ± 0.28a	1.99 ± 0.19b	1.71 ± 0.15bc	1.49 ± 0.10c
	S2	2.38 ± 0.15a	1.33 ± 0.12b	1.16 ± 0.11b	0.84 ± 0.10c
	S3	2.17 ± 0.11a	1.21 ± 0.06b	0.73 ± 0.04c	0.67 ± 0.05c
	S4	2.08 ± 0.19a	1.15 ± 0.15b	0.67 ± 0.07c	0.59 ± 0.05c
Chl <i>b</i> content [ $\text{mg g}^{-1}$ (FM)]	CK	6.42 ± 0.31a	7.01 ± 0.33a	7.06 ± 0.45a	7.19 ± 0.43a
	S1	0.15 ± 0.02b	0.19 ± 0.03b	0.20 ± 0.04b	0.70 ± 0.03a
	S2	0.22 ± 0.04b	0.29 ± 0.03b	0.39 ± 0.04a	0.43 ± 0.03a
	S3	0.16 ± 0.02b	0.22 ± 0.03b	0.23 ± 0.03b	0.52 ± 0.05a
	S4	0.22 ± 0.03c	0.23 ± 0.04c	0.37 ± 0.03b	0.51 ± 0.05a
Car content [ $\text{mg g}^{-1}$ (FM)]	CK	0.23 ± 0.02a	0.22 ± 0.01a	0.19 ± 0.03ab	0.17 ± 0.01b
	S1	0.26 ± 0.03a	0.25 ± 0.02a	0.22 ± 0.02a	0.20 ± 0.02a
	S2	0.14 ± 0.02b	0.21 ± 0.02a	0.24 ± 0.02a	0.28 ± 0.04a
	S3	0.16 ± 0.02c	0.19 ± 0.2c	0.25 ± 0.02b	0.34 ± 0.02a
	S4	0.15 ± 0.02c	0.21 ± 0.02bc	0.28 ± 0.04ab	0.42 ± 0.04a

Note: Small letters indicate significance of  $P < 0.05$  by Duncan's test within each row. Results are presented as mean  $\pm$  SD ( $n = 3$ ). Car, carotenoids; Chl, chlorophyll. CK, control; S1, 18/8°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S2, 12/2°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S3, 18/8°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S4, 12/2°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR.

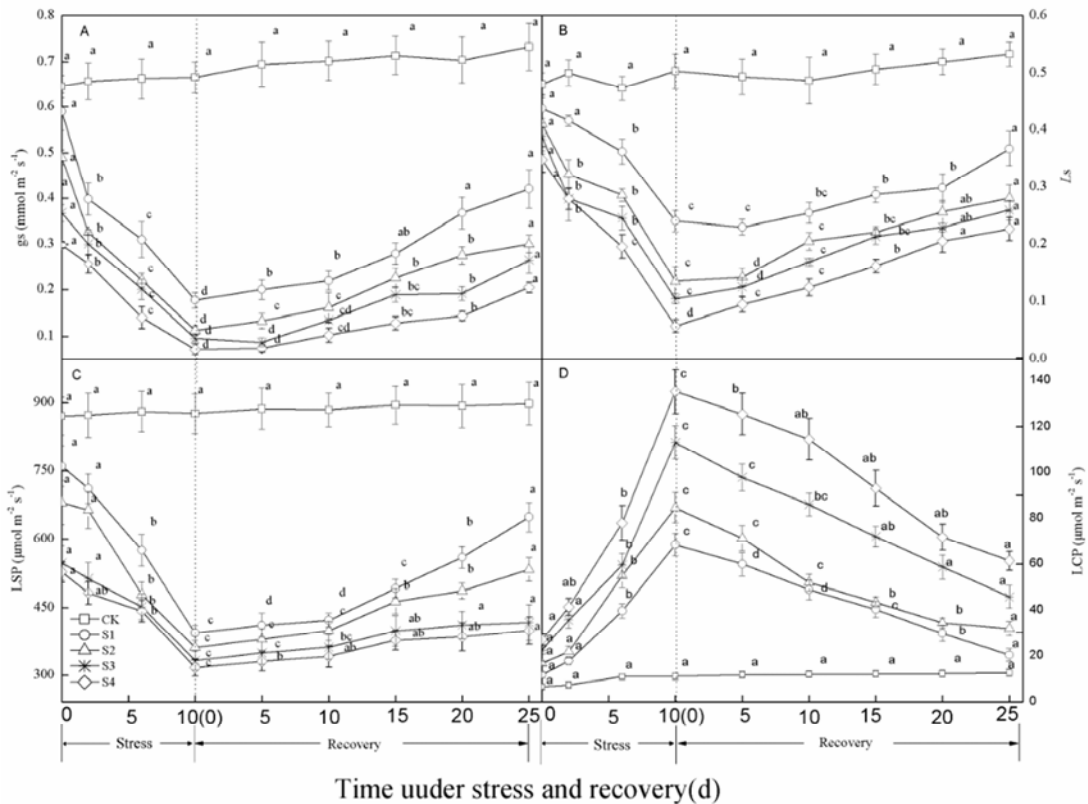
### 3.3. Photosynthetic Parameters

Tomato  $P_{\text{max}}$  declined during stress treatments, while it increased after plants were recovered for 25 d (Figure 1). The  $P_{\text{max}}$  decreased slightly in the first 2 dat and significantly at 6 and 10 dat. When plants were treated for 10 d, the  $P_{\text{max}}$  for S1,

S2, S3, and S4 significantly decreased compared to that of control. After 25 d of recovery, The  $P_{\text{max}}$  for S1 almost reached to that at 0 dat, the S2 was 74.5% of that at 0 dat, S3 and S4 were 57.8% and 47.2% of that at 0 dat. There was a significant difference between 25 and 0 d of recovery.



**Figure 1.** Photosynthetic rate at irradiation saturation ( $P_{max}$ ) in tomato leaves during the stress and recovery stage. Small letters indicate significance of  $P < 0.05$  by Duncan's test. Error bars represent SD,  $n = 3$ . CK, control; S1, 18/8°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S2, 12/2°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S3, 18/8°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S4, 12/2°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR.



**Figure 2.** Stomatal conductance ( $G_s$ ) (A), stomatal limitation value ( $L_s$ ) (B), light saturation point (LSP) (C), and light compensation point (LCP) (D) in tomato leaves during the stress and recovery stage. Small letters indicate significance of  $P < 0.05$  by Duncan's test. Error bars represent SD,  $n = 3$ . CK, control; S1, 18/8°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S2, 12/2°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S3, 18/8°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S4, 12/2°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR.

As shown in Figure 2, tomato  $g_s$ ,  $L_s$ , and light saturation point (LSP) reduced during the stress stage, and increased at 25 d after recovery (dar); while the changes of light compensation point (LCP) were opposite. The  $g_s$  for S1, S2, S3, and S4 significantly decreased compared with that of control at 10 dat;

the  $g_s$  for S1 could recover more than half of control, while the S2, S3, and S4 recovered 41.1%, 35.6%, and 27.4% of control at 25 dar (Figure 2A). The  $L_s$  declined slightly at 2 and 6 dat, and reduced significantly at 10 dat. During the treatments, the  $L_s$  for S4 was only 11.4% of control at 10 dat, but it recovered to

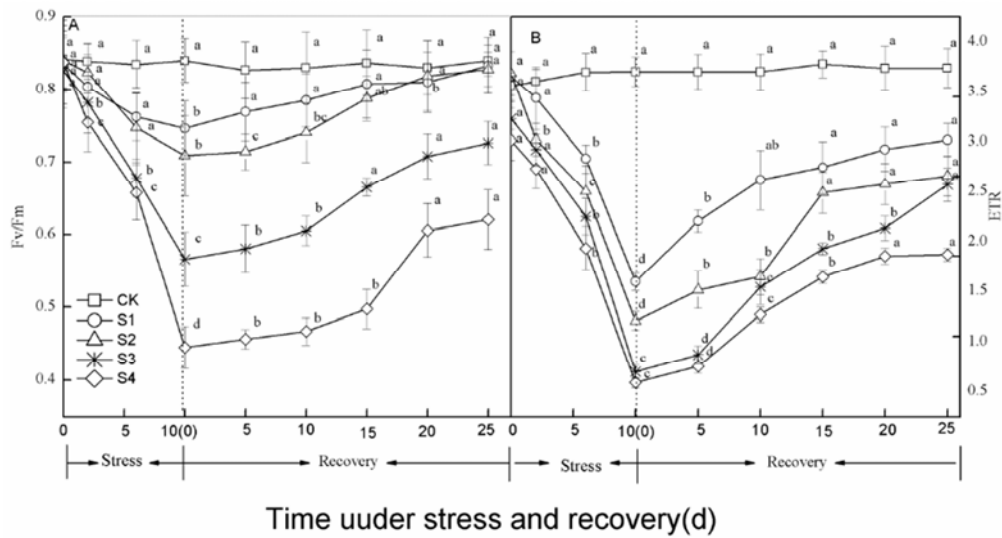
43.4% of control at 25 dar (Figure 2B). The LSP of all treatments was lower than that of control; the S1, S2, S3, and S4 significantly reduced at 10 dar, S1 and S2 could recover more than half of control at 25 dar (Figure 2C). It was contrast to LSP, the LCP of all treatments was higher than that of control. The LCP increased slightly at 2 dar and significantly at 6 and 10 dar. With an increase of recovery time, the LCP of all treatments declined; the value for S1, S2, S3, and S4 at 25 dar was significantly different from that at 0 dar (Figure 2D).

### 3.4. Chl Fluorescence Parameters

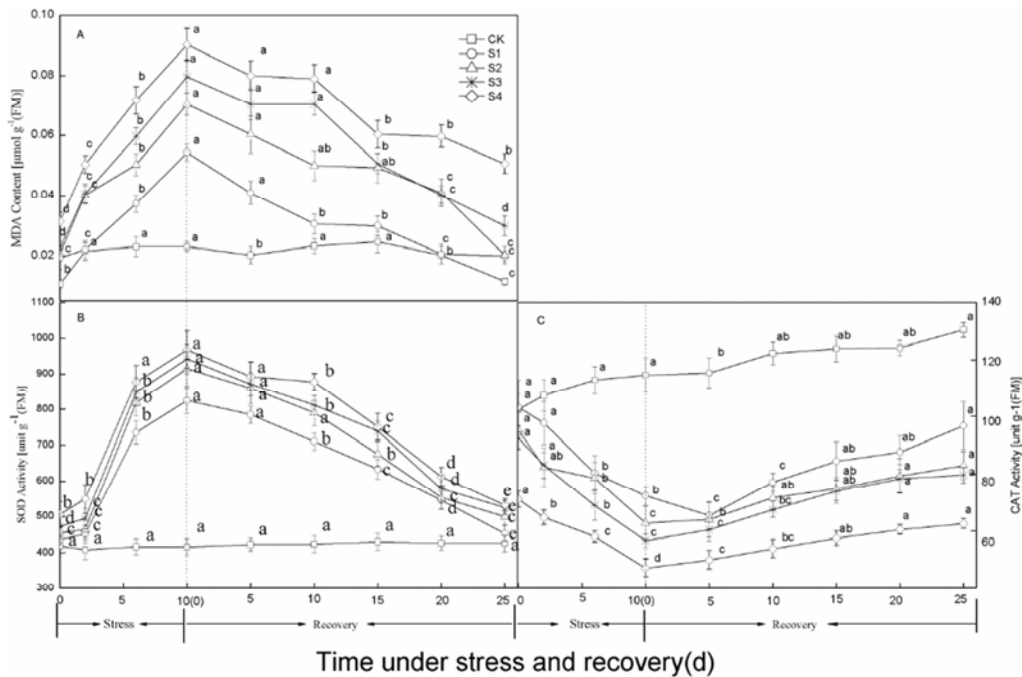
Both Fv/Fm and ETR decreased during stress stage, and

increased during recovery stage (Figure 3). Fv/Fm of tomato leaves under different LT and WL treatments was lower than that of control, and there was significant difference between 0 and 10 dar. Fv/Fm of S1 and S2 almost recovered to CK level after 20–25 d of recovery treatment, while that of S3 and S4 decreased to 84.9% and 72.1%, respectively, compared with control at 25 dar (Figure 3A).

The changes of ETR were consistent with that of Fv/Fm, the value of S1, S2, S3, and S4 decreased which compared with CK at 10 dar. After 25 d of recovery, the ETR of S1, S2, S3, and S4 was 80.7%, 73.0%, 70.3%, and 51.4% of control, respectively (Figure 3B).



**Figure 3.** Maximal quantum yield of PSII photochemistry (Fv/Fm) (A) and electron transport rate (ETR) (B) in tomato leaves during the stress and recovery stage. Small letters indicate significance of  $P < 0.05$  by Duncan's test. Error bars represent SD,  $n = 3$ . CK, control; S1, 18/8°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S2, 12/2°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S3, 18/8°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S4, 12/2°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR.



**Figure 4.** Malondialdehyde (MDA) content (A), Superoxide dismutase (SOD) activity (B), and catalase (CAT) activity (C) in tomato leaves during the stress and recovery phase. Small letters indicate significance of  $P < 0.05$  by Duncan's test. Error bars represent SD,  $n = 3$ . CK, control; S1, 18/8°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S2, 12/2°C D/N and 200  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S3, 18/8°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR; S4, 12/2°C D/N and 80  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR.

### 3.5. Antioxidant Enzyme and MDA

The MDA content and SOD activity of tomato leaves increased under stress treatments and decreased under recovery treatments, while the CAT activity was opposite to the SOD and MDA (Figure 4).

The MDA under different LT and WL significantly increased during the 10 d stress treatments. After 25 d of recovery, the MDA of S1 and S2 almost recovered to CK level, while S3 and S4 were 2.5 and 4.3 times of CK, and there was a significant difference between 25 dar and 0 dar (Figure 4A). The SOD for all LT and WL was higher than that of control under stress treatments. The SOD of S1, S2, S3, and S4 could obviously recover to control level at 25 dar (Figure 4B). The

CAT decreased under stress treatments, and it could not reach control level at 25 dar, but there was significant difference between 25 dar and 0 dar. (Figure 4C).

### 3.6. Fruit Quality

After 25 d of recovery, the Vc, soluble solid, soluble protein, and lycopene of tomato fruits less than that of CK, while the organic acid was opposite (Table 4). With a decline of temperature and light, the Vc, soluble solid, soluble protein, and lycopene of S1, S2, S3, and S4 significantly decreased compared with CK, except lycopene under S1. However, the organic acid for S1, S2, S3, and S4 increased significantly compared to that of CK.

Table 4. Effects of low temperature and weak light on tomato fruit quality.

Treatment	Vc [mg g <sup>-1</sup> (FM)]	Organic acid [mg g <sup>-1</sup> (FM)]	Soluble solid (%)	Soluble protein [mg g <sup>-1</sup> (FM)]	Lycopene [μg g <sup>-1</sup> (FM)]
CK	22.40 ± 2.23a	0.19 ± 0.02d	8.10 ± 0.57a	46.19 ± 3.15a	2.33 ± 0.16a
S1	14.90 ± 1.31b	0.27 ± 0.03c	7.10 ± 0.41b	42.01 ± 2.71a	2.21 ± 0.14a
S2	12.40 ± 1.15bc	0.34 ± 0.03b	6.93 ± 0.49b	41.00 ± 2.01a	1.74 ± 0.16b
S3	11.37 ± 1.07c	0.37 ± 0.03b	5.87 ± 0.41c	34.42 ± 2.35b	1.62 ± 0.14b
S4	10.03 ± 1.02c	0.44 ± 0.04a	5.40 ± 0.29c	32.76 ± 2.04b	1.56 ± 0.13b

Note: Small letters indicate significance of  $P < 0.05$  by Duncan's test within each column. Results are presented as mean ± SD (n = 3). Vc, Vitamin C. CK, control; S1, 18/8°C D/N and 200 μmol m<sup>-2</sup> s<sup>-1</sup> PAR; S2, 12/2°C D/N and 200 μmol m<sup>-2</sup> s<sup>-1</sup> PAR; S3, 18/8°C D/N and 80 μmol m<sup>-2</sup> s<sup>-1</sup> PAR; S4, 12/2°C D/N and 80 μmol m<sup>-2</sup> s<sup>-1</sup> PAR.

### 3.7. Correlation Analysis

We undertook a correlated analysis between morphological and physiological and quality indexes for tomato under different LT and WL (Table 5). The results showed that there was a highly significant positive correlation between plant height and Chl, soluble solid, and lycopene ( $P < 0.01$ ). There was an extremely significant negative correlation between

plant height and MDA, organic acid ( $P < 0.01$ ). Stem diameter and MDA, organic acid had a highly significant negative correlation ( $P < 0.01$ ), while stem diameter and soluble solid, soluble protein had an extremely significant positive correlation ( $P < 0.01$ ). There was no significant correlation between leaf area and other indexes.

Table 5. Correlation coefficient between morphological and physiological and quality indexes.

	Chl	$P_{max}$	SOD	CAT	MDA	Vc	Organic acid	Soluble solid	Soluble protein	Lycopene
Plant height	0.971**	0.919*	-0.929*	0.953*	-0.994**	0.938*	-0.998**	0.968**	0.956*	0.970**
Stem diameter	0.843	0.789	-0.805	0.870	-0.970**	0.811	-0.941**	0.966**	0.965**	0.860
Leaf area	-0.236	-0.456	0.434	-0.326	0.003	-0.415	0.104	-0.061	0.001	-0.064

Note: \* Mean significantly correlated at 0.05 level; \*\* mean significantly correlated at 0.01 level. Chl, chlorophyll;  $P_{max}$ , photosynthetic rate at irradiation saturation; SOD, superoxide dismutase; CAT, catalase; MDA, malondialdehyde; Vc, vitamin C.

## 4. Discussion

LT and WL limit tomato production in most single-slope solar greenhouses in northern China. The Chl contents are basic materials of photosynthesis, and photosynthesis is an important physiological process in plants, which can synthesize organic matter and generate energy [31-33]. In the present study, the Chl *a* significantly decreased under LT and WL, Chl *b* and Car fluctuated at early stage, but showed an increasing trend in later stage under LT and WL. The  $P_{max}$  decreased with an increase of stress duration under LT and WL (Figure 1). Meanwhile, the plant height and stem diameter changed consistently with that of  $P_{max}$ , while leaf area was opposite with that of  $P_{max}$  (Table 2). The reason was that photosynthesis was inhibited and the respiration increased after 10 d of LT and WL stress treatments, which resulting in

lower growth rate and larger leaves, these results had been confirmed [34-35]. The decrease of  $P_{max}$  may be determined by changes in *gs* [36]. *Ls* decreased with reduce of  $P_{max}$ , which indicated that the decrease of photosynthetic rate was not caused by stomatal conductance, but by non-stomatal factors. LSP also decreased, while LCP increased during the stress treatments. During the growth, all species should make photosynthetic and respiratory adjustments to LT and WL [37].

Chl fluorescence parameters are important indicators of plant photosystem activity [38]. The Fv/Fm and ETR declined under LT and WL treatments, which consistent with findings of Li *et al.* [32], who suggested that LT and WL reduced Fv/Fm and ETR of cut-flower chrysanthemum. Decrease of ETR showed that photosynthetic electron transport was inhibited.

Antioxidant system can prevent plants from negative effects of ROS. The antioxidant enzymes SOD and CAT play an important role in scavenging destructive oxidant species. High antioxidant enzyme levels have been found in response to heat, cold, and oxidative stress [39-40]. This study showed that LT and WL significantly enhanced the SOD activity and MDA content, and reduced CAT activity. The SOD increased significantly during the whole stress stage, mainly because the plants started self-protection mechanisms to adapt to external environment through regulation of antioxidant enzyme activities [41]. The reason might be that metabolic disorders caused by LT and WL resulted in an increase of ROS [1]. The CAT declined with the prolonged duration of LT and WL treatments, suggesting that the enzyme activity reduced dramatically. The reason maybe that severe stress treatments could cause the loss of CAT activity, and activated the Mehler reaction enzymes [30].

Lipid peroxidation has often been used to monitor ROS damage, it reflects a basic cell membrane reactive damage under abiotic stress [39, 42-43]. In this study, the MDA content increased greatly during long term LT and WL stress treatments (more than 6 d), which implied that the cell membrane system of plants suffered more serious cell damage.

Fruit quality of tomato includes the Vc, organic acid, soluble solid, soluble protein, and lycopene [44]. In this study, the Vc, soluble solid, soluble protein, and lycopene decreased with a decline of temperature and light, while the organic acid increased with a decrease of temperature and light. At the same light intensity conditions, the changes in Vc, organic acid, soluble solid, soluble protein, and lycopene were consistent with the results of Zhao et al. [26].

After 25 d of recovery, the  $P_{max}$ ,  $g_s$ ,  $L_s$ , LSP, and LCP for S1, S2, S3, and S4 could not recover to CK level (except the LCP for S1), but they could recover to 0 d stress level. The reason may be that photosynthetic parameters of higher plants are sensitive to temperature and light, and it is difficult to recover to normal level once the photosynthetic machinery is damaged. The Fv/Fm for S1 and S2 could be equal to that of CK, but ETR for all treatments could not reach the CK level, which indicated that LT and WL caused an irreversible damage to PSII reaction centers [37]. SOD activity and MDA content were higher than those of CK, while CAT activity was lower than that of CK after 25 d of recovery, which in agreement with Yang et al. [18].

## 5. Conclusion

In all, the results suggested that temperature and light play an important role in tomato growth and development. The Fv/Fm, as a key fluorescence parameter, will not be affected by species and environmental factors under normal growth conditions. In this study, the Fv/Fm for S1 and S2 almost recovered to normal level after 25 d of recovery, that is, we should adopt measures to increase temperature and light once light intensity is  $80 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  and D/N temperature is lower than  $18/8^\circ\text{C}$ . The aim was to reduce economic losses caused by LT and WL.

## Acknowledgements

Funding for this study was provided by the National Natural Science Foundation of China (41475107 and 31300420), and National Science & Technology Support Program during the Twelfth Five Year Plan Period (2014BAD10B07).

## References

- [1] Song, L. L., and Zhang, Q. (2007) Reactive oxygen gene network of plants and its regulation. Chinese Bulletin of Life Science, 19, 346-352. [In Chinese].
- [2] Allen, D. J., and Ort, D. R. (2001) Impacts of chilling temperatures on photosynthesis in warm-climate plants. Trends in Plant Science, 6, 36-42.
- [3] Marion, M. B. (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72:248-254.
- [4] Adams, S., Cockshull, K., and Cave, C. (2001) Effect of temperature on the growth and development of tomato fruits. Annals of Botany, 88, 869-877.
- [5] Keleş, Y., and Ncel, I. (2002) Response of antioxidative defence system to temperature and water stress combinations in wheat seedlings. Plant Science, 163, 783-790.
- [6] Boardman, N. (1977) Comparative photosynthesis of sun and shade plants. Annual Review of Plant Physiology, 28, 355-377.
- [7] Papageorgiou, G. (1975) Chlorophyll fluorescence: an intrinsic probe of photosynthesis. Bioenergetics of photosynthesis, pp 319-371.
- [8] Caesar, J. (1989b) Effect of simulated shade radiation quality on the chlorophyll contents of long and short early leaves of Birch (*Betula-pendula* roth). Inst experimental botany, acad sci czech republic NA Karlovce 1A, Prague 6, czech republic cs-160 00.
- [9] Ali, M. B., Hahn, E. J., and Paek, K. Y. (2005) Effects of light intensities on antioxidant enzymes and malondialdehyde content during short-term acclimatization on micropropagated Phalaenopsis plantlet. Environmental and Experimental Botany, 54, 109-120.
- [10] Feng, Y. L., K. F. Cao and J. L. Zhang. (2004) Photosynthetic characteristics, dark respiration, and leaf mass per unit area in seedlings of four tropical tree species grown under three irradiances. Photosynthetica 42:431-437.
- [11] Shu, S., Tang, Y., and Yuan, Y. (2016) The role of 24-epibrassinolide in the regulation of photosynthetic characteristics and nitrogen metabolism of tomato seedlings under a combined low temperature and weak light stress. Plant Physiology and Biochemistry, 107, 344-353.
- [12] Thompson, W., Huang, L., and Kriedemann, P. (1992) Photosynthetic response to light and nutrients in sun-tolerant and shade-tolerant rainforest trees. II. Leaf gas exchange and component processes of photosynthesis. Functional Plant Biology, 19, 19-42.
- [13] Höglind, M., Hanslin, H. M., and Mortensen, L. M. (2011) Photosynthesis of *Lolium perenne* L. at low temperatures under low irradiances. Environmental and Experimental Botany, 70, 297-304.



- [14] Sun, J. L., Sui, X. L., Huang, H. Y., Wang, S. H., Wei, Y. X., and Zhang, Z. X. (2014) Low light stress down-regulated Rubisco gene expression and photosynthetic capacity during Cucumber (*Cucumis sativus* L.) leaf development. *Journal of Integrative Agriculture*, 13, 997-1007.
- [15] Sun, S., Zhang, L., and Wang, J. (2008) Effects of low temperature and weak light on the functions of photosystem in *Prunus armeniaca* L. leaves in solar greenhouse. *The journal of applied ecology*, 19, 512-516.
- [16] Coleman, W. K., and Greyson, R. I. (1976) The growth and development of the leaf in tomato (*Lycopersicon esculentum*). I. The plastochron index, a suitable basis for description. *Canasian Journal of Plant Science*, 54, 2421-2428.
- [17] Hu, W. H., Zhou, Y. H., and Du, Y. S. (2006) Differential response of photosynthesis in greenhouse-and field-ecotypes of tomato to long-term chilling under low light. *Journal of Plant Physiology*, 163, 1238-1246.
- [18] Yang, Z. Q., Yuan, C. H., Han, W., and Xiao, F. (2016) Effects of low irradiation on photosynthesis and antioxidant enzyme activities in cucumber during ripening stage. *Photosynthetica*, 54, 251-258.
- [19] Gai, J. 2000 *Methods of experimental statistics*. Chinese Agriculture Press, China, Beijing.
- [20] Wellburn, A. R. (1994) The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. *Journal of Plant Physiology*, 144, 307-313.
- [21] Berry, J. A., and Downton, W. J. S. (1982) *Environmental regulation of photosynthesis*. Wiley, New York, pp 263-343.
- [22] Prioul, J., and Chartier, P. (1977) Partitioning of transfer and carboxylation components of intracellular resistance to photosynthetic CO<sub>2</sub> fixation: a critical analysis of the methods used. *Annals of Botany*, 41, 789-800.
- [23] Zhang, S., and Gao, R. (1999) Diurnal changes of gas exchange, chlorophyll fluorescence, and stomatal aperture of hybrid poplar clones subjected to midday light stress. *Photosynthetica*, 37, 559-571.
- [24] Schneider, S., Ziegler, C., and Melzer, A. (2006) Growth towards light as an adaptation to high light conditions in *Chara* branches. *New Phytologist*, 172, 83-91.
- [25] Diaz-Vivancos, P., Clemente-Moreno, M. J., and Rubio, M. (2008) Alteration in the chloroplastic metabolism leads to ROS accumulation in pea plants in response to plum pox virus. *Journal of Experimental Botany*, 59, 2147-2160.
- [26] Zhao, Y. P., Zou, Z. R., Yang, Z. C., Hu, X. H., Bai, P. W., Li, P. F., and R, L. (2010) Effect of temperature and light to tomato photosynthesis and quality in greenhouse. *Journal of northwest A. & F. university*, 38, 125-130. [In Chinese].
- [27] Sánchez-Moreno, C., Plaza, L., de Ancos, B., and Cano, M. P. (2003) Vitamin C, provitamin A carotenoids, and other carotenoids in high-pressurized orange juice during refrigerated storage. *Journal of Agricultural and Food Chemistry*, 51, 647-653.
- [28] Gutiérrez-Miceli, F. A., Santiago-Borraz, J., and Molina, J. A. M. (2007) Vermicompost as a soil supplement to improve growth, yield and fruit quality of tomato (*Lycopersicon esculentum*). *Bioresource Technology*, 98, 2781-2786.
- [29] Thomas, B. (2006) Light signals and flowering. *Journal of Experimental Botany*, 57, 3387-3393.
- [30] Moellering, H. and W. Gruber. 1966. Determination of citrate with citrate lyase. *Analytical Biochemistry*, 17, 369-376.
- [31] Caesar, J. (1989a) Effect of simulated shade radiation quality on the chlorophyll content of long and short shoot early leaves of birch (*Betula pendula* Roth.). *Photosynthetica*, 23, 126-129.
- [32] Li, X. G., Meng, Q. W., and Jiang, G. Q. (2003) The susceptibility of cucumber and sweet pepper to chilling under low irradiance is related to energy dissipation and water-water cycle. *Photosynthetica*, 41, 259-265.
- [33] Richardson, A. D., Duigan, S. P., and Berlyn, G. P. (2002) An evaluation of noninvasive methods to estimate foliar chlorophyll content. *New Phytologist*, 153, 185-194.
- [34] Crafts-Brandner, S. J., and Salvucci, M. E. (2002) Sensitivity of photosynthesis in a C4 plant, maize, to heat stress. *Plant Physiology*, 129, 1773-1780.
- [35] Murchie, E., Hubbart, S., and Peng, S. (2005) Acclimation of photosynthesis to high irradiance in rice: gene expression and interactions with leaf development. *Journal of Experimental Botany*, 56, 449-460.
- [36] Farquhar, G. D., and Sharkey, T. D. (1982) Stomatal conductance and photosynthesis. *Annual Review of Plant Physiology*, 33, 317-345.
- [37] Vieira, S., Calado, R., and Coelho, H. (2009) Effects of light exposure on the retention of kleptoplastic photosynthetic activity in the sacoglossan mollusc *Elysia viridis*. *Marine Biology*, 156, 1007-1020.
- [38] Hernandez, J., Jimenez, A., and Mullineaux, P. (2000) Tolerance of pea (*Pisum sativum* L.) to long-term salt stress is associated with induction of antioxidant defences. *Plant Cell Environment*, 23, 853-862.
- [39] Hernandez, J., Olmos E., and Corpas, F. (1995) Salt-induced oxidative stress in chloroplasts of pea plants. *Plant Science*, 105, 151-167.
- [40] Sabehat, A., Weiss, D., and Lurie, S. (1998) Heat-shock proteins and cross-tolerance in plants. *Physiologia Plantarum*, 103, 437-441.
- [41] Sun, G. C., Zeng, X. P., and Zhao, P. (2003) Photosynthesis and free radical yield of Litchi chinensis leaves under increased CO<sub>2</sub> partial pressure in atmosphere. *Chinese Journal of Apply Ecology*, 14, 331-335.
- [42] Liu, P., Yang, Y., and Xu, G. (2006) Physiological response of rare and endangered seven-son-flower (*Heptacodium miconioides*) to light stress under habitat fragmentation. *Environmental and Experimental Botany*, 57, 32-40.
- [43] Nouairi, I., Ammar, W. B., and Youssef, N. B. (2009) Antioxidant defense system in leaves of Indian mustard (*Brassica juncea*) and rape (*Brassica napus*) under cadmium stress. *Acta Physiologiae Plantarum*, 31, 237-247.
- [44] Kortstee, A., Appeldoorn, N., and Oortwijn, M. (2007) Differences in regulation of carbohydrate metabolism during early fruit development between domesticated tomato and two wild relatives. *Planta*, 226, 929-939.