

## BRIEF COMMUNICATION

**Effect of day/night temperature difference on chlorophyll content, photosynthesis and fluorescence parameters of tomato at fruit stage**

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In order to investigate the effect of day/night temperature difference (DIF) on photosynthetic characteristics of tomato plants (*Solanum lycopersicum*, cv. Jinguan 5) at fruit stage, an experiment was carried out in climate chambers. Five day/night temperature regimes (16/34, 19/31, 25/25, 31/19, and 34/16°C) with respective DIFs of -18, -12, 0, +12, and +18 were used and measured at mean daily temperature of 25°C. The results showed that chlorophyll (Chl) *a*, Chl *b*, net photosynthetic rate ( $P_N$ ), stomatal conductance ( $g_s$ ), maximum quantum yield of PSII photochemistry ( $F_v/F_m$ ), effective quantum yield of PSII photochemistry ( $\Phi_{PSII}$ ), and photochemical quenching ( $q_p$ ) significantly increased under positive DIF, while they decreased with negative DIF. In contrast, the Chl *a/b* ratio and nonphotochemical quenching (NPQ) decreased under positive DIF, while increased with negative DIF. Chl *a*, Chl *b*,  $P_N$ ,  $g_s$ ,  $F_v/F_m$ ,  $\Phi_{PSII}$ , and  $q_p$  were larger under +12 DIF than those at +18 DIF, while Chl *a/b* and NPQ showed an opposite trend.

*Additional key words:* nonstomatal limitation; photosynthetic efficiency.

Day/night temperature difference (DIF) has important influence on growth and morphogenesis of plants (Lepage *et al.* 1984, Erwin *et al.* 1989, Grimstad *et al.* 1993) and it is applied widely in horticulture industry (Berghage 1998). DIF also may exert significant influence on photosynthesis. Bunce (1985) found that positive DIF increased  $P_N$  in *Glycine max*. Berghage *et al.* (1990) and Agrawal *et al.* (1993) also reported similar results. However, Chabot and Lewis (1976) reported a little effect of DIF on photosynthetic characteristics of *Quercus rubra*. Up to date, the effect of DIF on the photosynthesis of tomato has not been reported. The aim of this study was to investigate whether positive DIF would increase  $P_N$  and photosynthesis efficiency of tomato at the fruit stage. In addition, the effect of negative DIF on photosynthesis of tomato was also determined.

Tomato (*Solanum lycopersicum* L., cv. Jinguan 5) seeds were germinated in vermiculite, and transplanted at the stage of three leaves into 25-cm plastic pots containing standard fertilized peat (*Floralux*, Norway). The plants were watered regularly with a nutrient solution containing N, P, and K in concentrations of 14.3, 1.0, and 5.1 mM,

respectively, and with other microelements. When young fruits showed up, uniform tomato plants were selected for the experiment. Five day/night temperature regimes were set as: 16/34, 19/31, 25/25, 31/19, and 34/16°C (12 h day time/12 h night time), with five DIFs of -18, -12, 0, +12, and +18. PPFD of 500  $\mu\text{mol}(\text{photon}) \text{m}^{-2} \text{s}^{-1}$ ,  $\text{CO}_2$  concentration of  $380 \pm 10 \mu\text{mol}(\text{CO}_2) \text{mol}^{-1}$ , and relative humidity of 60–70% were set in five chambers for each DIF treatment at the same time. Every chamber contained 10 plants.  $P_N$ ,  $g_s$ , and intercellular  $\text{CO}_2$  concentration ( $C_i$ ) were measured between 9:00 and 11:00 h using a portable photosynthesis system (*LI-6400*, *LI-COR Bioscience*, Lincoln, NE, USA). At least three leaves from different plants were chosen for measurement after each treatment. For each measurement, the third fully expanded leaf (from the apex) was exposed to irradiance of 1,000  $\mu\text{mol}(\text{photon}) \text{m}^{-2} \text{s}^{-1}$ ,  $\text{CO}_2$  concentration of  $380 \pm 10 \mu\text{mol}(\text{CO}_2) \text{mol}^{-1}$ , and air temperature of 25°C. Chl fluorescence parameters were recorded on the same leaves, where the gas exchange was evaluated, with a portable fluorimeter (*FMS 2*, *Hansatech*, UK). Leaves were acclimated to darkness for 30 min. The minimal fluorescence level ( $F_0$ ) was measured

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**Abbreviations:** Chl – chlorophyll;  $C_i$  – intercellular  $\text{CO}_2$  concentration;  $F_v/F_m$  – maximum quantum yield of PSII photochemistry;  $g_s$  – stomatal conductance; NPQ – nonphotochemical quenching;  $P_N$  – net photosynthetic rate;  $q_p$  – photochemical quenching;  $\Phi_{PSII}$  – effective quantum yield of PSII photochemistry.

with the modulated light [ $0.1 \mu\text{mol}(\text{photon}) \text{m}^{-2} \text{s}^{-1}$ ] at the dark-adapted state, and the maximal fluorescence level ( $F_m$ ) was determined after a 0.8-s saturating flashes of  $8,000 \mu\text{mol}(\text{photon}) \text{m}^{-2} \text{s}^{-1}$  in the dark-adapted leaves. The leaves were then continuously illuminated with white actinic light [ $600 \mu\text{mol}(\text{photon}) \text{m}^{-2} \text{s}^{-1}$ ] for 3 min; the steady state value of fluorescence ( $F_s$ ) was recorded. A second saturating pulse of  $8,000 \mu\text{mol}(\text{photon}) \text{m}^{-2} \text{s}^{-1}$  was imposed to determine maximal fluorescence level in the light-adapted state ( $F_m'$ ). Maximal efficiency of PSII ( $F_v/F_m$ ), effective quantum efficiency of PSII ( $\Phi_{\text{PSII}}$ ), photochemical quenching ( $q_p$ ), and nonphotochemical quenching (NPQ) were calculated as described by van Kooten and Snel (1990). Chl pigment was extracted with 80% (v/v) acetone and absorption at 663.2 and 646.8 nm was measured using a spectrophotometer *UV-1800* (Shimadzu, Japan) according to Lichtenthaler (1987).

Differences among different DIF treatments for the Chl content, photosynthetic parameters, and Chl fluorescence parameters were tested by a one-way analysis of variance (ANOVA) using the statistical software *SPSS 16.0* (SPSS Inc., Chicago, IL, USA) and the treatment means were compared by using *Duncan's* multiple range test (DMRT) at  $P \leq 0.05$ . Data were expressed as mean  $\pm$  SE.

Chl *a*, Chl *b*, and  $P_N$  increased under positive DIF, while they decreased under negative DIF, compared with  $0^\circ\text{C}$  DIF (Table 1). However, the Chl *a/b* ratio and  $C_i$  dropped under positive DIF, while they rose under negative DIF (Table 1). The  $g_s$  was elevated slightly under positive DIF, while it declined slightly under negative DIF (Table 1).  $F_v/F_m$ ,  $\Phi_{\text{PSII}}$ , and  $q_p$  increased under positive DIF, while decreased with negative DIF (Table 1). In contrast, NPQ decreased under positive DIF, while increased with negative DIF (Table 1). Chl *a*, Chl *b*,  $P_N$ ,  $g_s$ ,  $F_v/F_m$ ,  $\Phi_{\text{PSII}}$ , and  $q_p$  were larger under  $+12^\circ\text{C}$  DIF than that of  $+18^\circ\text{C}$  DIF, while Chl *a/b* and NPQ were lesser under  $+12^\circ\text{C}$  DIF than that of  $+18^\circ\text{C}$  DIF.

The Chl content increased under positive DIF, while decreased under negative DIF, which was in agreement with Berghage *et al.* (1990) and Vågen *et al.* (2003). The decrease in the Chl content of tomato plants under negative DIF could be related to Chl photooxidation because of excess energy absorbed (Krause 1988), or it is just an adaptive mechanism in order to prevent the absorption of excessive energy (Elvira *et al.* 1998).  $P_N$  significantly increased under positive DIF. The main reason could be that positive DIF increased Rubisco enzyme activity or increased its amount, thus promoted photosynthesis. The efficiency of the photosynthesis process can be evaluated through Chl fluorescence.  $F_v/F_m$ ,  $\Phi_{\text{PSII}}$ , and  $q_p$  increased under positive DIF, indicating that PSII efficiency was promoted under positive DIF. The reason may be that the activity of PSII was stimulated under positive DIF.  $P_N$ ,  $F_v/F_m$ ,  $\Phi_{\text{PSII}}$ , and  $q_p$  under  $+18^\circ\text{C}$  DIF were lesser than those under  $+12^\circ\text{C}$  DIF; it was in agreement with Chabot and Chabot (1977), who found that a  $+10^\circ\text{C}$  DIF increased  $P_N$ , but  $+20^\circ\text{C}$  DIF led to lower  $P_N$  in *Fragaria vesca*. It

Table 1. Effects of day/night temperature difference (DIF) on chlorophyll (Chl) content, photosynthesis and Chl fluorescence parameters of tomato plants. The values are means ( $\pm$  SE) of at least three replicates. Values not followed by the same letter within each row indicate significant differences between treatments at  $P \leq 0.05$ , based on *Duncan's* means tests.  $C_i$  – intercellular  $\text{CO}_2$  concentration;  $F_v/F_m$  – maximum quantum yield of PSII;  $g_s$  – stomatal conductance; NPQ – nonphotochemical quenching;  $P_N$  – net photosynthetic rate;  $\Phi_{\text{PSII}}$  – effective quantum yield of PSII photochemistry;  $q_p$  – photochemical quenching; FES – fruit expanding stage; FMS – fruit maturation stage.

Parameter	+18 DIF		+12 DIF		0 DIF		-12 DIF		-18 DIF	
	FES	FMS	FES	FMS	FES	FMS	FES	FMS	FES	FMS
Chl <i>a</i> [ $\text{mg g}^{-1}\text{FM}$ ]		2.04 $\pm$ 0.09 <sup>c</sup>		2.28 $\pm$ 0.11 <sup>d</sup>		1.85 $\pm$ 0.07 <sup>b</sup>		1.74 $\pm$ 0.05 <sup>b</sup>		1.62 $\pm$ 0.04 <sup>a</sup>
Chl <i>b</i> [ $\text{mg g}^{-1}\text{FM}$ ]		0.70 $\pm$ 0.05 <sup>c</sup>		0.81 $\pm$ 0.07 <sup>d</sup>		0.60 $\pm$ 0.04 <sup>b</sup>		0.53 $\pm$ 0.03 <sup>b</sup>		0.47 $\pm$ 0.02 <sup>a</sup>
Chl <i>a/b</i>		2.91 $\pm$ 0.09 <sup>c</sup>		2.81 $\pm$ 0.08 <sup>c</sup>		3.08 $\pm$ 0.12 <sup>b</sup>		3.28 $\pm$ 0.13 <sup>a</sup>		3.45 $\pm$ 0.16 <sup>a</sup>
$P_N$ [ $\mu\text{mol m}^{-2} \text{s}^{-1}$ ]	10.34 $\pm$ 0.86 <sup>c</sup>	12.16 $\pm$ 0.73 <sup>c</sup>	12.85 $\pm$ 0.79 <sup>d</sup>	14.34 $\pm$ 0.64 <sup>d</sup>	8.11 $\pm$ 0.55 <sup>b</sup>	9.45 $\pm$ 0.62 <sup>b</sup>	6.82 $\pm$ 0.34 <sup>a</sup>	7.32 $\pm$ 0.52 <sup>a</sup>	5.70 $\pm$ 0.16 <sup>a</sup>	6.20 $\pm$ 0.25 <sup>a</sup>
$g_s$ [ $\text{mol m}^{-2} \text{s}^{-1}$ ]	0.20 $\pm$ 0.03 <sup>c</sup>	0.22 $\pm$ 0.03 <sup>c</sup>	0.22 $\pm$ 0.02 <sup>c</sup>	0.24 $\pm$ 0.02 <sup>c</sup>	0.17 $\pm$ 0.02 <sup>b</sup>	0.19 $\pm$ 0.02 <sup>b</sup>	0.15 $\pm$ 0.01 <sup>b</sup>	0.16 $\pm$ 0.02 <sup>b</sup>	0.14 $\pm$ 0.01 <sup>a</sup>	0.14 $\pm$ 0.01 <sup>a</sup>
$C_i$ [ $\mu\text{mol mol}^{-1}$ ]	342 $\pm$ 4.71 <sup>c</sup>	334 $\pm$ 5.43 <sup>c</sup>	321 $\pm$ 4.56 <sup>d</sup>	317 $\pm$ 4.84 <sup>d</sup>	370 $\pm$ 5.41 <sup>b</sup>	361 $\pm$ 6.48 <sup>b</sup>	383 $\pm$ 7.82 <sup>a</sup>	376 $\pm$ 7.43 <sup>a</sup>	395 $\pm$ 8.43 <sup>a</sup>	389 $\pm$ 8.55 <sup>a</sup>
$F_v/F_m$	0.83 $\pm$ 0.06 <sup>b</sup>	0.85 $\pm$ 0.06 <sup>b</sup>	0.86 $\pm$ 0.06 <sup>c</sup>	0.87 $\pm$ 0.07 <sup>c</sup>	0.80 $\pm$ 0.05 <sup>b</sup>	0.82 $\pm$ 0.05 <sup>b</sup>	0.77 $\pm$ 0.05 <sup>a</sup>	0.78 $\pm$ 0.05 <sup>a</sup>	0.75 $\pm$ 0.04 <sup>a</sup>	0.73 $\pm$ 0.04 <sup>a</sup>
$\Phi_{\text{PSII}}$	0.62 $\pm$ 0.04 <sup>b</sup>	0.65 $\pm$ 0.05 <sup>b</sup>	0.67 $\pm$ 0.05 <sup>c</sup>	0.71 $\pm$ 0.05 <sup>c</sup>	0.58 $\pm$ 0.04 <sup>b</sup>	0.61 $\pm$ 0.04 <sup>b</sup>	0.55 $\pm$ 0.03 <sup>a</sup>	0.58 $\pm$ 0.03 <sup>a</sup>	0.52 $\pm$ 0.02 <sup>a</sup>	0.54 $\pm$ 0.03 <sup>a</sup>
$q_p$	0.78 $\pm$ 0.05 <sup>b</sup>	0.82 $\pm$ 0.06 <sup>b</sup>	0.82 $\pm$ 0.05 <sup>c</sup>	0.85 $\pm$ 0.06 <sup>c</sup>	0.74 $\pm$ 0.05 <sup>b</sup>	0.79 $\pm$ 0.05 <sup>b</sup>	0.67 $\pm$ 0.04 <sup>a</sup>	0.70 $\pm$ 0.04 <sup>a</sup>	0.62 $\pm$ 0.04 <sup>a</sup>	0.65 $\pm$ 0.03 <sup>a</sup>
NPQ	0.51 $\pm$ 0.03 <sup>c</sup>	0.44 $\pm$ 0.03 <sup>c</sup>	0.45 $\pm$ 0.03 <sup>c</sup>	0.38 $\pm$ 0.03 <sup>c</sup>	0.62 $\pm$ 0.04 <sup>b</sup>	0.59 $\pm$ 0.04 <sup>b</sup>	0.71 $\pm$ 0.05 <sup>a</sup>	0.69 $\pm$ 0.05 <sup>a</sup>	0.78 $\pm$ 0.06 <sup>a</sup>	0.75 $\pm$ 0.05 <sup>a</sup>

might be caused by the fact that the day temperature of large positive DIF was so high that it become unfavourable for photosynthesis.

Negative DIF reduced substantially  $P_N$ , but only slightly  $g_s$ , while  $C_i$  increased, indicating that the decrease of  $P_N$  could result from nonstomatal factors. Negative DIF also lowered  $F_v/F_m$ ,  $\Phi_{PSII}$ , and  $q_p$  of tomato plants. Many previous studies used a sustained decrease in  $F_v/F_m$  and  $\Phi_{PSII}$  as reliable indicators of photoinhibition of plants in response to stresses (Wagner and Dreyer 1997). Therefore negative DIF inhibited the efficiency of photosynthesis, which might be the main reason for the decrease of  $P_N$  under negative DIF. What is more, the loss of the Chl content under negative DIF may be another factor for the decrease of  $P_N$ . Another important finding was that NPQ of plants under negative DIF increased, so the plants might

be able to counteract the adverse effect of negative DIF at a chloroplast level through dissipating excessive excitation energy, in order to prevent photoinhibitory damage. The Chl *a/b* ratio also increased under negative DIF because Chl *b* was reduced more than Chl *a*, in accordance with Vågen *et al.* (2003). It might be attributed to adaptation mechanisms of tomato plants under negative DIF in order to prevent a capture of excessive light energy, as Chl *b* is the main component of light-harvesting complex protein (Reynolds *et al.* 2000).  $P_N$ ,  $g_s$ ,  $F_v/F_m$ ,  $\Phi_{PSII}$ , and  $q_p$  increased at the fruit maturation stage compared to fruit expanding stage for all DIF treatments; the reason might be that tomato plants possess more powerful photosynthesis at the fruit maturation stage than that at the fruit expanding stage.

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