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# Chlorophyll pigmentation and photosynthetic parameters in *Ornithogalum longibracteatum* L. as affected by varying temperatures in hydroponics solution

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The effects of different temperature regimes of hydroponic solution on the chlorophyll pigmentation and photosynthesis of *Ornithogalum longibracteatum* L. were determined in the glasshouse for 10 weeks in 2009 and 2010. The plants were irrigated with hydroponic solution, heated to various temperatures (26, 30 and 34°C) via pumps connected to 4 sets of water tanks, each maintained at the experimental temperatures using dolphin aquarium heaters. Unheated water supplied from the fourth tank served as control (temperature ranged between 10 and 15°C (day/night) throughout the experiment period). All plants were supplied with 1 mgL<sup>-1</sup> nutrient solution of (hortical) and the solution was changed at weekly intervals. After 2 to 10 weeks of experimentation, data showed that chlorophyll a, b and total, were significantly increased by elevating hydroponic solution temperature from 26 to 30°C, and started decreasing at 34°C compared with the control in both 2009 and 2010. Photosynthesis rate (A) and the gas exchange parameters; stomata conductance (gs), intercellular CO<sub>2</sub> concentration (Ci) and transpiration (E), were significantly increased by elevating the hydroponic solution temperatures from 26 to 30°C compared with the control, and then decreased significantly at 34°C. The findings from this study suggest that the performance of *O. longibracteatum* can be improved during winter seasons by heating the hydroponic solution up to 30°C. Beyond this, temperature led to impaired chlorophyll formation and reduced photosynthesis.

**Key words:** Intercellular CO<sub>2</sub> concentration, photosynthesis rate, stomata conductance, transpiration.

## INTRODUCTION

*Ornithogalum longibracteatum* is classified as a medicinal bulb used widely in South Africa. The plant which is also commonly known as a pregnant onion is used as a medicinal plant by traditional healers (Kulkarni et al., 2005). From its potential as a medicinal plant, its on-farm and greenhouse cultivation is becoming popular. *O. longibracteatum* can be grown hydroponically in a greenhouse even during adverse climatic conditions (Rosik-Dulewska and Grabda, 2002) provided that the harsh environmental factors are addressed. *O. longibracteatum* grows best at temperatures ranging from 22 to 27°C (Luria et al., 2002) and it does not grow well in cold weather if temperatures are less than 11°C (Halevy et al., 1971).

Temperature is an important factor affecting physiological processes in plants including photosynthetic rate and chlorophyll synthesis (Lambreva et al., 2005 Calatayud et al., 2008). Like other growth processes in plants, temperature changes in the soil and air may have positive or negative impacts on leaf photosynthetic rate and the chlorophyll synthesis (Vu and Yelenosky, 1987; Huang and Gao, 2000; Xu and Huang, 2000, 2001a and b; Huang et al., 2001; Lyons et al., 2007). For example,

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some of the photosynthetic parameters such as stomata conductance (Gs), intercellular CO<sub>2</sub> concentration (Ci) and transpiration (E) are known to be influenced by the changes in temperature (Drake et al., 1970; Pearcy, 1977; Haldimann and Feller, 2004). In plants such as tomato (Lycopersicon esculentum L.), low temperature (1°C) reduced stomata conductance by 25% leading to decreased leaf chloroplast functioning (Martin et al., 1981). However, low temperatures inhibit the rate of photosynthesis by approximately 60% as the result of the impairment of water oxidation mechanisms (Martin et al., 1981). Other studies (Haldimann and Feller, 2004) have shown that increase in leaf temperature up to 45°C in oak (Quercus pubescens L.) plants reduced photosynthesis rate by 90%, and stomata conductance, but increased intercellular CO<sub>2</sub> compared with when it was at 25°C.

According to Drake et al. (1970), high temperatures, ranging from 35 to 40°C, increased transpiration in leaves and low levels such as 5 and 10°C reduced transpiration rate of *Xanthium spp* plants, ultimately affecting stomata conductance activities. In another study, temperatures above 25°C led to closure of stomata, thus reducing the transpiration rate in potatoes (*Solanum tuberosum* L.) a mechanism for adaptation to hot environment (Ku et al., 1977). In a study by Baig and Tranquillini (1980), it was reported that increasing temperature from 15 to 20°C increased transpiration rate in *Pica abies* and *Pinus cembra* to 65.8 and 63.6% respectively. Similarly, when temperature was further increased to 25°C, transpiration was also increased to 146.3 and 196.7% respectively.

Research has revealed that at low temperatures (10°C), the peroxidation activities in the chloroplast membrane were lowered due to inhibition of the metabolic processes in the leaves of coffee seedlings (Goncalves de Oliveinaa et al., 2009). It was also shown that low temperature (8°C) significantly reduced the chlorophyll levels in spinach leaves (Spinacea oleraceae L.) (Lopez-Ayera et al., 1998). The findings of Ilík et al (2000) strongly suggested that, an increase in temperature within the range of 25 to 75°C affect the chloroplast membrane which resulted in the burst of thylakoids and formed condensed structures in barlev leaves. Temperature variations in the rooting zone are an important factor which may influence different metabolic processes in plants (Walker, 1969; Gur et al., 1972; Cooper, 1973; Sattelmacher et al., 1990). For instance, it is suggested that higher temperatures in the rooting zone above the optimum range could result into excessive consumption of carbon through increased respiration and a reduction in carbon assimilation in photosynthesis (Huang and Gao, 2000; Xu and Huang, 2000a, b, 2001; Liu and Huang, 2001). Furthermore, research has shown that the disturbance of carbohydrate metabolism in roots was a major primary factor responsible for growth inhibition at high soil temperature (Du and Tachibana, 1994; Chung et al., 2002). Therefore, it is worth establishing if any stress factor such as variations in temperature of the hydroponic solution will impair the

physiological function of the plant such as those involving chlorophyll formation and photosynthetic processes. The purpose of this study was to determine the effect of changes in hydroponic solution temperature regimes on chlorophyll synthesis and photosynthetic rate so as to establish the optimum temperature for the growth of *O*. *longibracteatum* in cold season.

### MATERIALS AND METHODS

#### Site location and description

The experiment was conducted at the greenhouse of the Cape Peninsula University of Technology, Cape Town, South Africa during the winter season of 2009 and 2010. A steel table (2.5 x 1 m) was used as a flat surface, black plastic container (50 L), leca clay pebbles were supplied by Horticultural Department of Cape Peninsula University of Technology (CPUT), Cape Town, South Africa. Four (4) plastic gutters (2 x 0.6 m), 4 pumps, 20 ml black plastic pipe, cable tie and 3 dolphin aquarium heaters were purchased from Builders Warehouse (Maitland, Cape Town), South Africa . Bulbs of pregnant onion (*O. longibracteatum*) used as planting material were obtained from the CPUT nursery.

## Experimental design

A randomised complete block design, with four replicates, was conducted to study the effects of temperature on chlorophyll pigmentation and photosynthetic rate in O. longibracteatum. Four white plastic gutters (2 x 0.6 m) filled with leca clay pebbles were placed on a 2.5 x 1 m steel table. Water was supplied to the leca pebbles through pumps projecting from 4 sets of black plastic containers (50 I) placed beneath the table. The water in the 3 containers was heated by using Dolphin aquarium heaters to maintain the temperatures at 26, 30 and 34°C respectively. Unheated water supplied from the forth container served as control. Using the thermometer, the temperature ranged between 10 and 15°C (day/night) throughout the experiment period. 0 longibracteatum bulbs were planted in each gutter (10 bulbs per gutter) and supplied with nutrient solution immediately after transplanting. The nutrient solution was prepared according to Ocean HYDROGRO (2009) and Ocean HORTICAL (2009) respectively. Nutrient solution supplied from the pumps was recirculated back to the black plastic container (50 l) through a 20 ml black plastic pipe. The plants were left to grow for the period of 10 weeks. To prevent concentration of nutrients in the clay pebbles due to evaporation, water was drained from the gutters and refreshed after every week.

#### Determination of chlorophyll contents in plant leaves

Chlorophyll concentration was extracted from the third leaf from the growing tip of each plant growing in the gutters using dimethyl sulphoxide (DMSO), as described by Hiscox and Israelstam (1979). The strap-like leaves were cut into small pieces, and a 100 mg of the middle portion of the leaf tissue was placed in a 15 ml vial containing 7 ml DMSO and incubated at 4°C for 72 h. After the incubation, the extract was diluted to 10 ml with DMSO, and 3 ml of extract was used to read the absorbance at 645 and 663 nm on a spectrophotometer (UV/Visible Spectrophotometer, Pharmacia LKB. Ultrospec II E) against DMSO blank. Chlorophyll levels were calculated using the equations used by Arnon (1949) with a unit of mgL<sup>-1</sup> and is given thus:

Treatments	Chlorophyll a (mgL <sup>-1</sup> )	Chlorophyll b (mgL <sup>-1</sup> )	Chlorophyll total (mgL <sup>-1</sup> )
2009			
<sup>†</sup> Control	2.25±0.15 <sup>d</sup>	$0.51 \pm 0.03^{d}$	2.77±0.14 <sup>d</sup>
26°C	4.68±0.17 <sup>b</sup>	1.89±0.07 <sup>b</sup>	6.57±0.22 <sup>b</sup>
30°C	6.77±0.25 <sup>a</sup>	3.65±0.05 <sup>a</sup>	10.42±0.24 <sup>a</sup>
34°C	3.39±0.16 <sup>c</sup>	1.42±0.05 <sup>c</sup>	4.81±0.17 <sup>c</sup>
One - way ANOVA (F-statistic)	109.63**	596.70**	271.07**
2010			
<sup>†</sup> Control	2.13±0.20 <sup>d</sup>	0.56±0.04 <sup>d</sup>	2.69±0.22 <sup>d</sup>
26°C	5.83±0.25 <sup>b</sup>	2.95±0.14 <sup>b</sup>	8.78±0.28 <sup>b</sup>
30°C	8.45±0.37 <sup>a</sup>	5.02±0.38 <sup>a</sup>	13.46±0.69 <sup>a</sup>
34°C	2.99±0.17 <sup>c</sup>	1.99±0.21 <sup>°</sup>	4.98±0.31 <sup>°</sup>
One - way ANOVA (F-statistic)	123.38**	67.94***	126.33**

Table 1. Effect of temperature on chlorophyll content in leaves of O. Longibracteatum L. during 2009 and 2010.

Values presented are means  $\pm$  SE, n = 10. \*\*; \*\*\* = significant at P≤0.01, P≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at P = 0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

Chl  $a = 12.7D_{663} - 2.69D_{645}$ Chl  $b = 22.9D_{645} - 4.68D_{663}$ Total Chl =  $20.2D_{645} + 8.02D_{663}$ 

#### Measurement of photosynthesis in plant leaves

At 2, 4, 6, 8 and 10 weeks after planting, photosynthesis, stomata conductance, intercellular  $CO_2$  and evapotranspiration were measured in four young leaves (flag leaves) per gutter using a portable infrared red gas analyzer (LCpro+ 1.0 ADC, Bioscientific Ltd., Hoddesdon, Hertfordshire, UK). Measurements were made from 8 to 11 a.m and from 2 to 4 p.m for each replicate gutter per day. Leaves were allowed at least 5 min to acclimate to the light environment in the chamber. Under normal conditions, each measurement took approximately 2 min, which was the minimum time allowed for the readings to stabilize before they were recorded. During measurements, the conditions in the leaf chamber were: photosynthetic photon flux density (PPFD) = 1100 µmol (quantum) m<sup>-2</sup>s<sup>-1</sup>, relative humidity = 44%, leaf vapor pressure deficit = 1.83 kPa, flow rate = 400 µmols<sup>-1</sup>, reference CO<sub>2</sub> = 400 ppm, and leaf temperature = 25°C.

#### Statistical analysis

The experimental data collected were analysed by using a one-way analysis of variance (ANOVA). The analysis was performed using STASTICA software programme 2010 (StatSoft Inc., Tulsa OK, USA). Where F-value was found to be significant, Fisher's least significant difference (LSD) was used to compare the means at P < 0.05 level of significance (Steel and Torrie, 1980).

## RESULTS

## Effect of temperature on chlorophyll content of leaves of *O. longibracteatum*

Table 1 shows the effect of four different temperature treatments on chlorophyll content in the leaves of *O*.

*longibracteatum.* Results showed that, relative to the control treatment, increasing the water temperature to 26, 30 and 34°C significantly increased the levels of chlorophyll a, b, and total chlorophyll in 2009 and 2010 (Table 1). For instance, at 30°C, the level of chlorophyll a, b and total chlorophyll were significantly higher compared with all the other treatments (Table 1). However, as the temperature was increased to 34°C, leaf chlorophyll content was significantly reduced compared with the other treatments in both 2009 and 2010. Although plants grown during 2010 season contained more chlorophyll than their counterparts grown in 2009, the influence of temperature showed similar trend across seasons.

## Effect of temperature on photosynthesis and gasexchange parameters of leaves of *O. longibracteatum*

There was significant difference in the photosynthesis rate (A) and the gas exchange parameters (E, Ci and Gs) at different temperature treatments during 2009 and 2010 (Tables 2, 3, 4, 5, 6 and 7). Generally, the photosynthesis rate and the gas exchange parameters were significantly increased by elevating the hydroponic solution temperatures to 26, 30, and 34°C compared with the control (10/15°C). For example, results indicated that in week 2, 4, 6, 8 and 10 the photosynthesis rate (A) values were consistently increased by modifying the temperature to 26 and 30°C respectively and then decreased significantly at 34°C. Data from this study showed that raising the temperature beyond 30°C, the photosynthesis rate (A) started experiencing significantly negative effects and the values were significantly maximized at 30°C in both years.

The values for transpiration (E) recorded in weeks 2, 4,

		WEEK 2				WEEK 4			
Treatments	Α	Е	Ci	gs	Α	E	Ci	gs	
	µmol CO₂ m <sup>-2</sup> s <sup>-1</sup>	mmol.m <sup>-2</sup> s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>	µmol CO₂ m⁻² s⁻¹	mmol m <sup>-2</sup> .s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>	
<sup>†</sup> Control	1.14±0.01 <sup>d</sup>	0.38±0.01 <sup>d</sup>	216.00±0.77 <sup>d</sup>	0.02±0.00 <sup>d</sup>	1.29±0.01d	0.52±0.03d	223.00±2.69d	0.03±0.00d	
26°C	2.48±0.10 <sup>b</sup>	0.70±0.01 <sup>b</sup>	261.30±5.19 <sup>b</sup>	0.04±0.00 <sup>b</sup>	2.63±0.05b	0.77±0.02b	274.60±5.01b	0.05±0.00b	
30°C	2.78±0.04 <sup>a</sup>	0.89±0.02 <sup>a</sup>	336.10±8.55 <sup>a</sup>	0.06±0.00 <sup>a</sup>	3.81±0.04a	1.30±0.07a	375.20±4.33a	0.07±0.00a	
34°C	1.57±0.02 <sup>c</sup>	0.52±0.02 <sup>c</sup>	240.70±7.51 <sup>°</sup>	0.03±0.00 <sup>c</sup>	2.29±0.02c	0.66±0.01c	248.10±4.68c	0.04±0.00c	
One - way ANOVA	194.00**	156.89**	68.34***	74.73***	936.14**	75.16***	244.11**	49.47***	
(F-statistic)									

Table 2. Effect of temperature on photosynthesis and gas-exchange parameters of leaves of O. Longibracteatum L. as measured from weeks 2 and 4 during 2009.

Values presented are means  $\pm$  SE, n = 10. \*\*; \*\*\* = significant at P≤0.01, P≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at P = 0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

Table 3. Effect of temperature on photosynthesis and gas-exchange parameters of leaves of O. Longibracteatum L. as measured from weeks 6 and 8 during 2009.

	WEEK 6				WEEK 8			
Treatments	Α	E	Ci	gs	Α	E	Ci	gs
	µmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup>	mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO <sub>2</sub> mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>	µmol CO₂ m <sup>-2</sup> s <sup>-1</sup>	Mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>
<sup>†</sup> Control	1.43±0.09d	0.62±0.00d	226.90±2.38d	0.03±0.00d	1.66±0.07d	0.74±0.01d	248.30±5.30d	0.04±0.00d
26°C	3.38±0.07b	0.81±0.02b	292.70±1.51b	0.05±0.00b	3.29±0.05b	0.89±0.02b	333.60±6.12b	0.06±0.00b
30°C	4.61±0.05a	1.59±0.05a	365.00±4.79a	0.07±0.00a	6.84±0.19a	1.91±0.02a	372.80±6.10a	0.08±0.00a
34°C	2.33±0.07c	0.71±0.02c	266.30±7.70c	0.04±0.00c	2.59±0.08c	0.81±0.01c	273.30±8.31c	0.05±0.00c
One - way ANOVA (F-statistic)	367.21**	219.00**	150.20**	58.64***	412.46**	1022.03**	74.58***	35.69***

Values presented are means  $\pm$  SE, n = 10.\*\*; \*\*\* = significant at P≤0.01, P≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at P=0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

6, 8 and 10 during 2009 and 2010 indicated that there was significant increase in this parameter when temperatures were raised to 26, 30 and  $34^{\circ}$ C compared with the control (10/15°C). Relative to the control treatment, increasing temperature to 26°C in weeks 2, 4, 6, 8 and 10 significantly increased E values in the average range of 21 to 159% in 2009 and 30 to 136% in 2010 respectively (Tables 2, 3, 4, 5, 6 and 7). Further, compared with the control, increasing temperature to 30°C in weeks 2, 4, 6, 8 and 10 significantly increased E values in the average range of 134 to 206% in 2009 and 114 to 339% in 2010 respectively (Tables 2, 3, 4, 5, 6 and 7). However, the E values showed a decreasing trend by raising temperatures to 34°C. Generally, there

were significant reduction in E values when the hydroponic solution temperature was raised to  $34^{\circ}$ C compared with the  $30^{\circ}$ C treatment in both 2009 and 2010 (Tables 2, 3, 4, 5, 6 and 7). The data showed that the best result for E was recorded in the  $30^{\circ}$ C treatment.

In this study, the intercellular  $CO_2$  concentration (Ci) values were significantly increased by elevating

	Week 10						
Treatments	Α	E	Ci	gs			
	µmol CO₂ m⁻²s⁻¹	mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO <sub>2</sub> mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>			
<sup>†</sup> Control	3.50±0.12 <sup>d</sup>	0.69±0.08 <sup>d</sup>	231.70±6.10 <sup>d</sup>	0.04±0.00 <sup>d</sup>			
26°C	5.39±0.09 <sup>b</sup>	1.79±0.03 <sup>b</sup>	335.60±2.25 <sup>b</sup>	$0.06 \pm 0.00^{b}$			
30°C	8.23±0.10 <sup>a</sup>	2.12±0.02 <sup>a</sup>	384.00±4.06 <sup>a</sup>	0.08±0.00 <sup>a</sup>			
34°C	4.59±0.12 <sup>c</sup>	0.95±0.01 <sup>°</sup>	284.90±3.39 <sup>c</sup>	0.05±0.00 <sup>c</sup>			
One - way ANOVA (F-statistic)	334.78**	228.89**	244.90**	88.22***			

Table 4. Effect of temperature on photosynthesis and gas-exchange parameters of leaves of *O. longibracteatum* L. as measured at week 10 during 2009.

Values presented are means  $\pm$  SE, n = 10. \*\*; \*\*\* = significant at *P*≤0.01, *P*≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at P = 0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

Table 5. Effect of temperature on photosynthesis and gas-exchange parameters of leaves of O. Longibracteatum L. as measured during weeks 2 and 4 in 2010.

	WEEK 2				WEEK 4			
Treatments	Α	E	Ci	gs	Α	E	Ci	gs
	µmol CO₂ m <sup>-2</sup> s <sup>-1</sup>	mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>	µmol CO₂ m <sup>-2</sup> s <sup>-1</sup>	mmol m <sup>-2</sup> .s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>
<sup>†</sup> Control	1.15±0.01 <sup>d</sup>	0.26±0.01 <sup>d</sup>	154.90±7.17 <sup>d</sup>	0.01±0.00 <sup>d</sup>	1.19±0.02 <sup>d</sup>	0.35±0.01 <sup>d</sup>	227.70±4.74 <sup>d</sup>	0.02±0.00 <sup>d</sup>
26°C	1.87±0.03 <sup>b</sup>	0.42±0.02 <sup>b</sup>	267.40±4.46 <sup>b</sup>	0.03±0.00 <sup>b</sup>	1.99±0.07 <sup>b</sup>	0.82±0.07 <sup>b</sup>	281.90±4.25 <sup>b</sup>	$0.06 \pm 0.00^{b}$
30°C	2.79±0.03 <sup>a</sup>	0.69±0.01 <sup>a</sup>	359.50±7.68 <sup>ª</sup>	0.06±0.00 <sup>a</sup>	3.30±0.06 <sup>a</sup>	1.52±0.10 <sup>a</sup>	351.30±11.49 <sup>a</sup>	0.13±0.01 <sup>ª</sup>
34°C	1.46±0.03 <sup>c</sup>	0.35±0.02 <sup>c</sup>	242.90±1.92 <sup>c</sup>	0.03±0.00 <sup>c</sup>	1.74±0.03 <sup>c</sup>	0.57±0.02 <sup>c</sup>	257.80±6.79 <sup>°</sup>	0.04±0.00 <sup>c</sup>
One - way ANOVA	743.57**	113.70**	211.35**	71.02***	329.39**	61.94***	50.69***	66.87***
(F-statistic)								

Values presented are means  $\pm$  SE, n = 10. \*\*; \*\*\* = significant at *P*≤0.01, *P*≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at *P*=0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

the hydroponic solution temperatures to 26, 30 and 34°C compared with the control (10/15°C). Data collected in weeks 2, 4, 6, 8 and 10 showed that Ci values increased significantly when temperatures were raised from 26 to 30°C compared with the control during 2009 and 2010 (Tables 2, 3, 4, 5, 6 and 7) and started decreasing at 34°C. For example, at 26°C, Ci values increased

significantly between 21 to 45% in 2009, and 24 to 73% in 2010 (Tables 2, 3, 4, 5, 6 and 7). Raising temperature to 34°C, however, resulted into significant decrease in Ci values compared with 30°C treatments in both years. The overall results obtained during weeks 2, 4, 6, 8 and 10 showed that at 30°C, Ci was significantly the highest with the average values ranging from 50 to 68% in

## 2009 and 54% to 132% in 2010 respectively.

The value of stomata conductance (gs) increased significantly in weeks 2, 4, 6, 8 and 10 when temperature was increased to 26, 30 and 34°C compared with the control (10/15°C) in 2009 and 2010 (Tables 2, 3, 4, 5, 6 and 7). For example, at 26°C, results observed during weeks 2, 4, 6, 8 and 10 showed significantly (P = 0.05)

Treatments	WEEK 6				WEEK 8			
	Α	E	Ci	gs	Α	Е	Ci	gs
	µmol CO₂ m <sup>-2</sup> s <sup>-1</sup>	mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO <sub>2</sub> mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>	µmol CO₂ m⁻²s⁻¹	mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup>
<sup>†</sup> Control	1.35±0.01 <sup>d</sup>	0.41±0.01 <sup>d</sup>	207.40±15.67 <sup>d</sup>	0.03±0.00 <sup>d</sup>	1.38±0.04 <sup>d</sup>	0.50±0.02 <sup>d</sup>	238.90±2.35 <sup>d</sup>	0.03±0.00 <sup>d</sup>
26°C	2.55±0.09 <sup>b</sup>	0.63±0.03 <sup>b</sup>	334.00±16.31 <sup>b</sup>	0.07±0.01 <sup>b</sup>	3.07±0.09 <sup>b</sup>	0.86±0.04 <sup>b</sup>	308.00±9.61 <sup>b</sup>	0.09±0.02 <sup>b</sup>
30°C	4.16±0.31 <sup>a</sup>	0.88±0.03 <sup>a</sup>	374.40±2.72 <sup>a</sup>	0.15±0.01 <sup>a</sup>	7.01±0.15 <sup>ª</sup>	1.54±0.06 <sup>a</sup>	352.10±3.86 <sup>a</sup>	0.17±0.01 <sup>a</sup>
34°C	1.89±0.02 <sup>c</sup>	0.51±0.03 <sup>c</sup>	258.80±12.23 <sup>c</sup>	$0.05 \pm 0.00^{\circ}$	2.35±0.11 <sup>°</sup>	0.63±0.03 <sup>c</sup>	267.30±1.99 <sup>c</sup>	$0.06 \pm 0.00^{\circ}$
One - way ANOVA	55.43***	69.15***	33.51***	97.51**	538.2**	152.33**	83.27***	42.57***
(F-statistic)								

Table 6. Effect of temperature on photosynthesis and gas-exchange parameters of leaves of *O. longibracteatum* L. as measured during weeks 6 and 8 in 2010.

Values presented are means  $\pm$  SE, n = 10. \*\*; \*\*\* = significant at *P*≤0.01, *P*≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at P = 0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

	Week 10						
Treatments	Α	E	Ci	gs			
	µmol CO₂ m <sup>-2</sup> s <sup>-1</sup>	mmol m <sup>-2</sup> s <sup>-1</sup>	mmol CO₂ mol <sup>-1</sup> air	mmol H₂O m <sup>-2</sup> s <sup>-1</sup>			
<sup>†</sup> Control	3.55±0.03 <sup>d</sup>	1.35±0.03 <sup>d</sup>	241.90±13.20 <sup>d</sup>	0.03±0.00 <sup>d</sup>			
26°C	5.77±0.05 <sup>b</sup>	1.76±0.03 <sup>b</sup>	352.50±5.14 <sup>b</sup>	$0.06 \pm 0.00^{b}$			
30°C	8.33±0.17 <sup>a</sup>	2.59±0.09 <sup>a</sup>	378.50±1.97 <sup>a</sup>	0.18±0.01 <sup>a</sup>			
34°C	4.30±0.03 <sup>c</sup>	1.54±0.05 <sup>c</sup>	320.10±9.94 <sup>c</sup>	0.04±0.00 <sup>c</sup>			
One - way ANOVA (F-statistic)	508.81**	96.43**	46.31***	362.44**			

**Table 7.** Effect of temperature on photosynthesis and gas-exchange parameters of leaves of *Ornithogalum longibracteatum* L. as measured during week 10 in 2010.

Values presented are means  $\pm$  SE, n = 10. \*\*; \*\*\* = significant at P≤0.01, P≤0.001 respectively, ns = not significant, SE = standard error. Means followed by dissimilar letters in a column are significantly different from each other at P=0.05 according to Fischer least significance difference. †=Temperature of control treatment ranged between 10/15°C (day/night) throughout the experiment period.

greater gs values and ranged from 38 to 137% in 2009 and from 97 to 258% in 2010. During the two years of experimentation, best results for stomata conductance were obtained at 30°C but the values increased significantly between 89 and 300%, in 2009, and 369 and 611% in 2010 (Tables 2, 3, 4, 5, 6 and 7). However, a signify-cant decline in the levels of gs when temperature was increased to 34°C was observed.

## DISCUSSION

Optimum temperature in hydroponics plays a crucial role in plant growth and other plant physiological characteristics including chlorophyll content. Exposure of plants to high or low temperatures may damage the chlorophyll membrane structures leading to low chlorophyll content (llík, 2000). Likewise, extreme temperatures may either

stop or denature enzyme activities leading to reduced rate of A, gs, Ci and E (Pearcy, 1977; Camejo et al., 2005). Maintaining optimum hydroponics temperature may be one way to ensure optimum chlorophyll content and increased A, gs, Ci and E in such plants. In this study, the chlorophyll content in the leaves of *O. longibracteatum* showed that by increasing the hydroponics water temperature to 26 and 30°C, significantly increased the levels of chlorophyll a, chlorophyll b and total chlorophyll compared with the control (10/15°C) during 2009 and 2010 (Tables 1). For example, in 2009, compared with the control, chlorophyll a content was 108, 201 and 51% at 26, 30 and 34°C respectively (Table 1). Similar trend was observed in 2010, where chlorophyll a content increased by 174, 297 and 41% by elevating temperatures to 26, 30 and 34°C respectively over the control (Table 1). Greater accumulation of the chlorophyll content in the leaves of O. longibracteatum at 30°C suggests that there was no damage in its physical or chemical properties and its functional organization. However, as the hydroponics water temperature was raised to 34°C, leaf chlorophyll content was significantly decreased. The reduced leaf chlorophyll content suggests that the temperature altered its physical and chemical properties and its functional organization as similarly reported by Taylor and Craig (1971), Ferrini et al. (1995) and llik et al. (2000).

Photosynthesis is considered as one of the most temperature sensitive processes, and may be completely inhibited by high temperatures above the optimum (Camejo et al., 2005). In this study, increasing the temperature from 26 to 30°C increased A, gs, Ci and E in the leaves of O. longibracteatum compared with the control (10/15°C) (Tables 2, 3, 4, 5, 6 and 7). From the results, the peak of these parameters was observed at 30°C. For example, in the second week of 2009, the rate of photosynthesis was 118, 144 and 38% at 26, 30 and 34°C respectively, and in 2010, photosynthesis rate was 63, 143 and 23% at 26, 30 and 34°C respectively over the control (Tables 2, 3, 4, 5, 6 and 7). The increase in the rate of photosynthesis and the related parameters suggests that these temperatures were limiting the A, Ci, gs and E. The data also showed that as the temperature was increased to 34°C, A and the related parameters were significantly reduced (Tables 2, 3, 4, 5, 6 and 7). The diminution in the rate of photosynthesis at 34°C may be ascribed to disruption of structure and the function of chloroplasts, reduction of chlorophyll accumulation (Table 1), enzyme denaturation due to oxidative stress, stomata closing or increased respiration rate (Xu et al., 1995; Dekov et al., 2000). These findings suggest that the rate of photosynthesis in plants growing at 35°C depends not on stomatal opening but on biochemical factors of an enzymatic nature. Previous reports have indicated that increased temperature beyond optimum (30°C) in citrus plant (Ribeiro et al., 2004; Hu et al., 2007) and self-rooted cv. Trebbiano grapevines (Ferrini et al., 1995) was the reason for decreased carboxylation efficiency. In another study, Ku and Edwards (1977) revealed that increasing temperature resulted not only in reduced internal CO<sub>2</sub> concentration in potatoes (Solanum tuberosum L.) but also, the rate of photosynthesis was inhibited by 38%.

In conclusion, increasing hydroponics water temperature to 30°C, leads to a significantly positive increase of chlorophyll content in *O. longibracteatum*, greater photosynthesis rate, stomata conductance, intercellular  $CO_2$  concentration and transpiration. However, increasing the temperature to 34°C resulted in possible structural and functional disruptions of chloroplasts and reduced chlorophyll accumulation leading to decreased rate of photosynthesis and related parameters in *O. longibracteatum.* 

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