

## **The effect of high carbon dioxide and temperature on the total chlorophyll content in grape varieties**

### **Abstract:**

The study was carried out to investigate the effect of eCO<sub>2</sub> (550ppm), eT (+3°C) and interaction of eCO<sub>2</sub>+ eT on chlorophyll content of three grape varieties (Thompson Seedless, Bangalore Blue, and Dogridge). The cuttings of these three varieties were grown in specially designed open-top chambers, Free Air Carbon dioxide Enrichment (FACE) and Free Air Temperature Enrichment (FATE) facilities at ICAR-CRIDA. The experiment was laid out in two factorial RBD with varieties as one factor and treatments as another factor with six replications. Destructive sampling was done at 50 and 80 DAP and chlorophyll content (µg/g.fr.wt.) was analysed each time. At final sampling, all the three varieties have shown higher chlorophyll content under eCO<sub>2</sub>+ eT followed by eCO<sub>2</sub>. Among three varieties Bangalore blue has shown highest chlorophyll content (3.1±0.37) followed by Thompson Seedless (3.0±0.06) and Dogridge (2.4±0.08) under eCO<sub>2</sub>+ eT conditions. The plants under eT and ambient conditions have shown lower chlorophyll content than other treatments. The results from this experiment revealed that, the increase in atmospheric carbon dioxide concentration (eCO<sub>2</sub>) and temperature associated with future climates are expected to affect positively on chlorophyll content which can stimulate photosynthetic rate and ultimately leads to increase in biomass of crop.

**Keywords:** CO<sub>2</sub> concentration, Chlorophyll content, Green House Gases, global warming and synthesis

### **INTRODUCTION:**

The global climate is changing by increase in the concentration of Green House Gases (GHGs) such as CO<sub>2</sub> and thereby increasing air temperature. Elevated CO<sub>2</sub> itself is expected to contribute to global warming which affects the whole biome. Grape is sensitive to different environmental factors, including temperature, water availability and CO<sub>2</sub>. Further, as grape is a C<sub>3</sub> plant, its photosynthesis is CO<sub>2</sub>-limited (Bindi et al., 1996; Mullins et al., 1992). So, any increase of atmospheric CO<sub>2</sub> concentration could affect the growth and yield (Bowes 1993), and results in a higher accumulation of biomass (Bindi et al., 1996). So, it is essential to study the independent effects of elevated CO<sub>2</sub> (eCO<sub>2</sub>), elevated temperature (eT) and their interaction (eCO<sub>2</sub>+ eT) on agriculture/ horticulture crop production [8-11]. Keeping this in view, an experiment was carried out with an objective to study the effect of elevated carbon dioxide and temperature on Chlorophyll Content of three popular grape cultivars.

### **MATERIALS AND METHODS:**

The hard wood stem cuttings of 25-30 cm length having 5-6 nodes of three grape varieties i.e. Thompson Seedless, Bangalore Blue, Dogridge were collected from the winter pruning of grape plants. These stem cuttings were treated with standard 1500 ppm IBA solution and planted in poly bags. The poly bags are of 23×12×12 cm and filled with potting mixture of Red Soil + FYM in 1:1 proportion and 15 number of cuttings each variety were exposed to ambient control, eCO<sub>2</sub> (550 ppm), eT (+3°C) and eCO<sub>2</sub>+eT (550 ppm + +3°C) conditions. The planted cuttings were kept under specially designed Open Top Chambers (OTC) and Free Air Temperature Enrichment (FATE) facilities in Rabi 2017 at ICAR-CRIDA. The chlorophyll content of each variety was analyzed from all the conditions (ambient control, eCO<sub>2</sub>, eT and eCO<sub>2</sub>+eT) at 50 and 80 days after planting (DAP).

### **Evaluation of Chlorophyll Content (µg/g.fr.wt.):**

Fresh leaf samples from different treatments were cut and 50 mg of it was weighed accurately with an analytical balance and chlorophyll was extracted by a non macerated method by equilibrating it with 10 ml DMSO (Dimethyl Sulphoxide) in a capped vial and keeping it in dark for 24 hrs. After 24 hrs the decanted solution was used to estimate the absorbance at 645 and 663 nm wavelength using Spectronic-20 Spectrophotometer. Chlorophyll was expressed in leaf weight basis (µg of Chl / gm of leaf fresh weight).

$$\text{Chlorophyll a} = (12.7A_{663} - 2.69A_{645}) (V/1000 \times W)$$

$$\text{Chlorophyll b} = (22.9A_{645} - 4.68A_{663}) (V/1000 \times W)$$

$$\text{Total Chlorophyll} = \text{chlorophyll a} + \text{chlorophyll b}$$

Where, V is the final volume (ml) of extract, W is the weight fresh leaf sample and A<sub>645</sub> and A<sub>663</sub> are the absorbance at 645 and 663 nm, respectively.

## **RESULTS AND DISCUSSION**

### **1) Chlorophyll a (µg/g fr. wt):**

The data on chlorophyll a of grape varieties recorded under different climatic conditions (eCO<sub>2</sub>, eT, eCO<sub>2</sub>+eT) is presented in Table 1 and analysis of variance in Table 2. At 50 DAP, treatments and varieties had significant ( $P \leq 0.01$ ) difference in chlorophyll a. While at 80 DAP, varieties and treatments had significant difference, whereas interaction of varieties and treatments has shown non-significant.

In Thompson Seedless the chlorophyll a content was highest under eCO<sub>2</sub>+eT at 50 (2.1±0.06) and 80 DAP (2.4±0.05) and lowest under eT (0.8±0.01, 1.21±0.23). While Bangalore Blue has shown maximum chlorophyll content under eCO<sub>2</sub> (3.2±0.02) at 50 DAP and under eCO<sub>2</sub>+eT (2.6±0.37) at 80 DAP. Dogridge has shown highest chlorophyll a content under eCO<sub>2</sub>+eT (4.2±0.08) followed by eCO<sub>2</sub> (3.7±0.06) at 50 DAP, however at 80 DAP the content of chlorophyll a was similar (1.9) under eCO<sub>2</sub>+eT and eCO<sub>2</sub>. Chlorophyll a was decreased from 50 to 80 DAP in Bangalore Blue and Dogridge. Thompson Seedless has shown an increase in

chlorophyll a from 50 to 80 DAP. Among all the three varieties, maximum chlorophyll a content was found in Dogridge followed by Bangalore Blue.

At 50 DAP there was a little increase of chlorophyll a content of about 1% in Thompson Seedless under eCO<sub>2</sub> whereas Bangalore Blue and Dogridge has shown an increase of 48 and 327% over control. Under eT Bangalore Blue and Dogridge has shown an increase of 14 and 275% respectively in chlorophyll a over control, while, Thompson Seedless has shown a decrease of 43%. Under eCO<sub>2</sub>+eT there was an increase of 61, 38 and 377% over control in Thompson Seedless, Bangalore Blue and Dogridge respectively (Fig. 1).

At 80 DAP, Thompson Seedless, Bangalore Blue and Dogridge shown a decrease in chlorophyll a content of 43, 13 and 13.6% over control under eT. While under eCO<sub>2</sub> there was an increase of 4.7, 23 and 66% in Thompson Seedless, Bangalore Blue and Dogridge respectively. Under eCO<sub>2</sub>+ eT there was an increase of 56, 43 and 69% in Thompson Seedless, Bangalore Blue and Dogridge respectively over control (Fig. 1).

Under eT the chlorophyll a content decreased in all the varieties by 80 DAP, though in Dogridge at 50 DAP, eT showed an increase in chlorophyll content, however by 80 DAP the content was decreased. This shows that under eT alone there would be decrease in chlorophyll a content, however when eT in combination with eCO<sub>2</sub> has shown an increase. Thus the negative impact of eT on chlorophyll content can be overcome with eCO<sub>2</sub>.

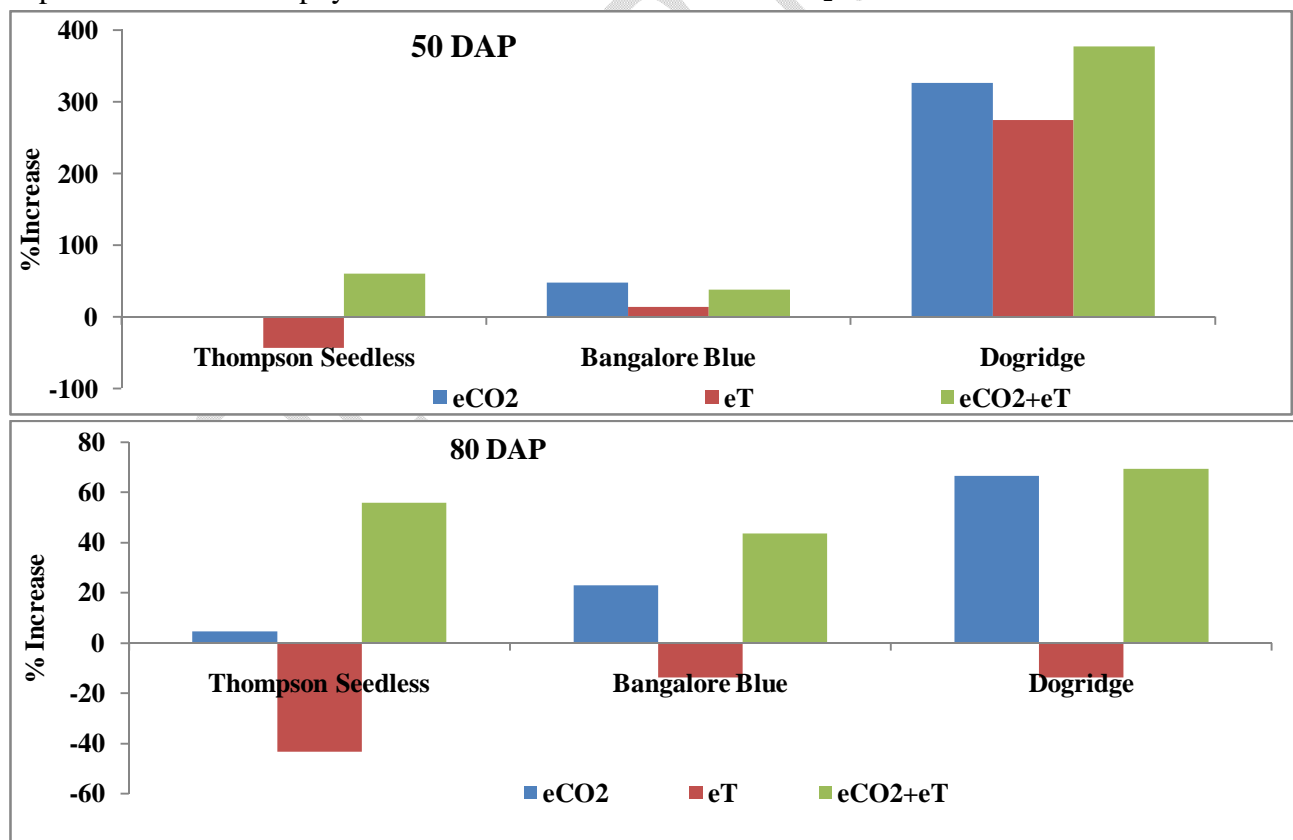


Fig 1: Percentage increase in chlorophyll a content of grape varieties over control at 50 and 80 DAP

## 2. Chlorophyll b ( $\mu\text{g/g fr. wt}$ ):

The data on chlorophyll b content of grape varieties recorded under different climatic conditions (eCO<sub>2</sub>, eT, eCO<sub>2</sub>+eT) is presented in Table 1 and analysis of variance was given in Table 2. At 50 and 80 days after planting, treatments and varieties had significant ( $P \leq 0.01, 0.05$ ) difference on chlorophyll b.

The chlorophyll b content did not differ between treatments. Thompson Seedless has shown higher amount of chlorophyll b under eCO<sub>2</sub>+eT at both 50 ( $0.6 \pm 0.02$ ) and 80 DAP ( $0.6 \pm 0.04$ ). While Bangalore Blue under eCO<sub>2</sub> at 50 ( $0.9 \pm 0.01$ ) and 80 DAP ( $0.7 \pm 0.10$ ) and Dogridge under eCO<sub>2</sub>+eT at 50 ( $0.5 \pm 0.02$ ) and 80 DAP ( $0.5 \pm 0.04$ ).

Thompson Seedless has shown an increase of 8.5 and 24% in chlorophyll b under eCO<sub>2</sub> at 50 and 80 DAP and increase of 30% and 73% under eCO<sub>2</sub>+eT at 50 and 80 DAP over control, whereas under eT Thompson Seedless had shown a decrease of 52 and 38% at 50 and 80 DAP respectively. Bangalore Blue has shown an increase of 235, 19 and 56% under eCO<sub>2</sub>, eT and eCO<sub>2</sub>+eT at 50 DAP and an increase of 84, 9 and 41% under eCO<sub>2</sub>, eT and eCO<sub>2</sub>+eT at 80 DAP over control. Dogridge has also recorded an increase of 162, 157 and 211% under eCO<sub>2</sub>, eT and eCO<sub>2</sub>+eT at 50 DAP and an increase of 66, 8 and 80% under eCO<sub>2</sub>, eT and eCO<sub>2</sub>+eT at 80 DAP over control (Fig 2).

Similar to chlorophyll a content, the chlorophyll b content was also decreased under eT in all the treatments, thus it is clear that higher temperatures there would be decrease in leaf chlorophyll content in grape varieties.

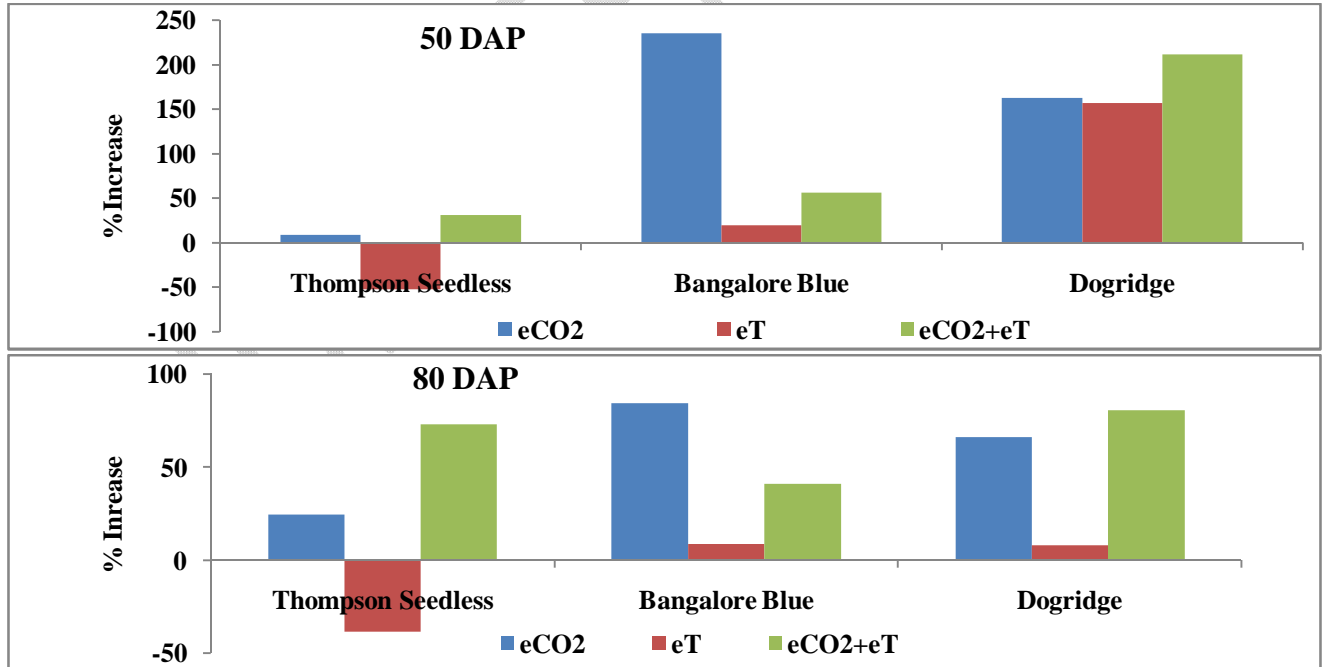


Fig 2: Percentage increase in chlorophyll b content of grape varieties over control at 50 and 80 DAP

## 3. Total chlorophyll ( $\mu\text{g/g fr. wt}$ ):

The data on Total chlorophyll of grape varieties recorded under different climatic conditions (eCO<sub>2</sub>, eT, eCO<sub>2</sub>+eT) is presented in Table 1 and analysis of variance in Table 2. At 50 days after planting, treatments and varieties had significant ( $P \leq 0.01$ ) difference on total chlorophyll. While at 80 days after planting, varieties and treatments has shown significant, whereas interaction of varieties and treatments had shown non-significant.

Total chlorophyll content of Thompson Seedless was higher under eCO<sub>2</sub>+eT at 50 (2.8±0.05) and 80 DAP (3.0±0.06). In Bangalore Blue the chlorophyll content was higher under eCO<sub>2</sub> at 50DAP (4.1±0.02) and under eCO<sub>2</sub>+eT (3.1±0.37) at 80DAP. Dogridge has shown maximum chlorophyll content under eCO<sub>2</sub>+eT at both 50 (4.7±0.09) and 80 DAP (2.4±0.08). The chlorophyll content tends to be decreased from 50 to 80DAP under all treatments. High chlorophyll content was found either under eCO<sub>2</sub> or eCO<sub>2</sub> when combined with elevated temperature (eCO<sub>2</sub>+eT). The plants under eT and ambient has shown lower chlorophyll content than other two treatments.

Thompson Seedless, Bangalore Blue and Dogridge has shown increase in total chlorophyll than control under eCO<sub>2</sub> and eCO<sub>2</sub>+eT at 50 and 80 DAP, whereas there was an increase of 15 and 257% under eT in Bangalore Blue and Dogridge at 50 DAP over control, however Thompson Seedless shown a decrease of 46%. At 80 DAP there was a decrease of 42, 9 and 13.6% in chlorophyll content in Thompson Seedless, Bangalore Blue and Dogridge respectively under eT over control. From these results it is clear that under eT the chlorophyll content was lowered in all the three grape varieties with different magnitude (Fig 3).

One of the most consistent effects of elevated atmospheric CO<sub>2</sub> on plants is an increase in the rate of photosynthetic carbon fixation by leaves. Across a range of FACE experiments, with a variety of plant species, growth of plants at elevated CO<sub>2</sub> concentrations of 475–600 ppm increases leaf photosynthetic rates by an average of 40% (Ainsworth & Rogers 2007). Since photosynthesis and stomatal behavior are central to plant carbon and water metabolism, growth of plants under elevated CO<sub>2</sub> leads to a large variety of secondary effects on plant physiology. The availability of additional photosynthates enables most plants to grow faster under elevated CO<sub>2</sub>, with dry matter production in FACE experiments being increased on average by 17% for the aboveground, and more than 30% for the belowground portions of plants (Ainsworth & Long 2005; de Graaff *et al.*, 2006). Wang *et al.* (2004) found that elevated CO<sub>2</sub> increased the number of chloroplasts per unit cell area by 71% in *Nicotiana sylvestris* leaves.

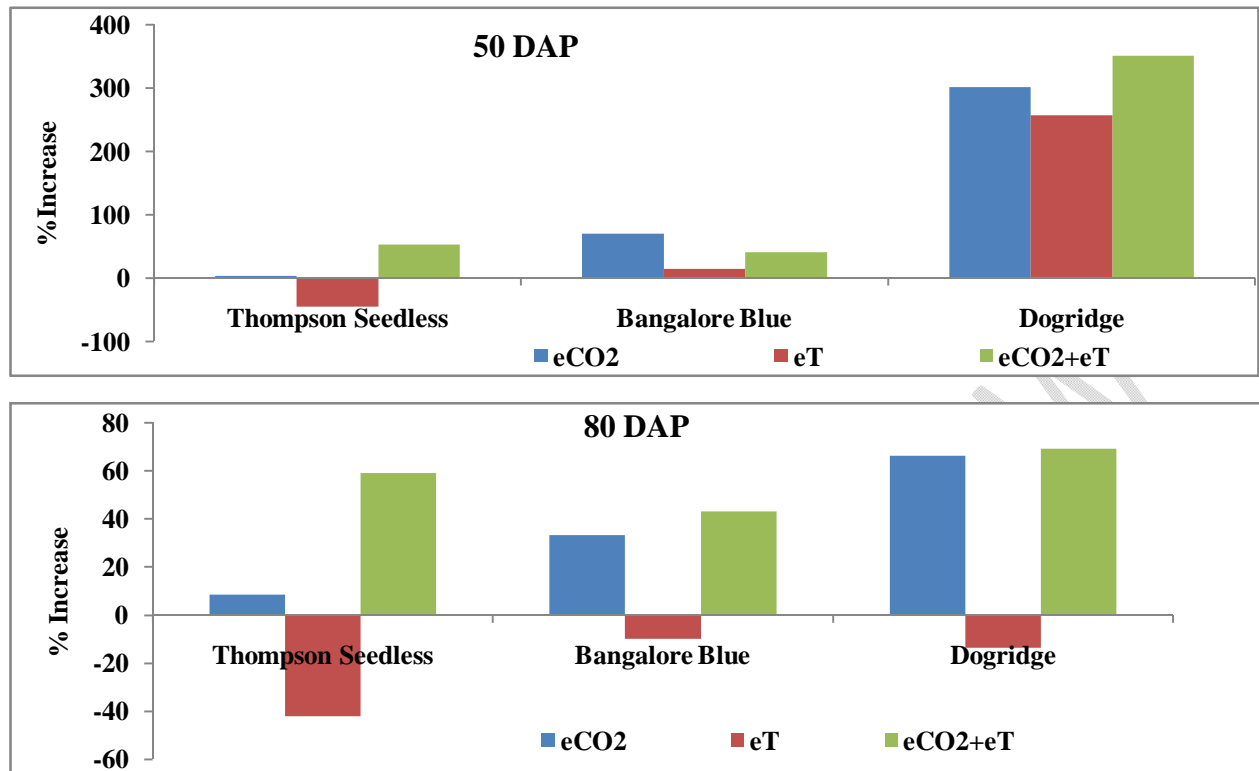


Fig 3: Percentage increase in total chlorophyll content of grape varieties over control at 50 and 80 DAP

**Table 1: Mean ( $\pm$ SE) performance for Chlorophyll content of grape varieties under different treatments;**

Treatments	Varieties	Chlorophyll a ( $\mu\text{g/g fr. wt.}$ )		Chlorophyll b ( $\mu\text{g/g fr. wt.}$ )		Total chlorophyll ( $\mu\text{g/g fr. wt.}$ )	
		50 DAP	80DAP	50DAP	80DAP	50DAP	80DAP
eCO <sub>2</sub>	TS	<b>1.3<math>\pm</math>0.05</b>	<b>1.6<math>\pm</math>0.05</b>	<b>0.5<math>\pm</math>0.01</b>	<b>0.5<math>\pm</math>0.01</b>	<b>1.9<math>\pm</math>0.05</b>	<b>2.1<math>\pm</math>0.05</b>
	BB	<b>3.2<math>\pm</math>0.02</b>	<b>2.2<math>\pm</math>0.27</b>	<b>0.9<math>\pm</math>0.01</b>	<b>0.7<math>\pm</math>0.10</b>	<b>4.1<math>\pm</math>0.02</b>	<b>2.9<math>\pm</math>0.35</b>
	DR	<b>3.7<math>\pm</math>0.06</b>	<b>1.9<math>\pm</math>0.21</b>	<b>0.4<math>\pm</math>0.02</b>	<b>0.5<math>\pm</math>0.07</b>	<b>4.2<math>\pm</math>0.07</b>	<b>2.4<math>\pm</math>0.26</b>
e T	TS	<b>0.8<math>\pm</math>0.01</b>	<b>1.21<math>\pm</math>0.23</b>	<b>0.2<math>\pm</math>0.0</b>	<b>0.2<math>\pm</math>0.04</b>	<b>1.0<math>\pm</math>0.01</b>	<b>1.1<math>\pm</math>0.24</b>
	BB	<b>2.4<math>\pm</math>0.05</b>	<b>1.6<math>\pm</math>0.10</b>	<b>0.3<math>\pm</math>0.01</b>	<b>0.4<math>\pm</math>0.04</b>	<b>2.8<math>\pm</math>0.05</b>	<b>1.9<math>\pm</math>0.13</b>
	DR	<b>3.3<math>\pm</math>0.06</b>	<b>1.0<math>\pm</math>0.17</b>	<b>0.4<math>\pm</math>0.01</b>	<b>0.3<math>\pm</math>0.03</b>	<b>3.7<math>\pm</math>0.07</b>	<b>1.3<math>\pm</math>0.20</b>
eCO <sub>2</sub> + eT	TS	<b>2.1<math>\pm</math>0.06</b>	<b>2.4<math>\pm</math>0.05</b>	<b>0.6<math>\pm</math>0.02</b>	<b>0.6<math>\pm</math>0.04</b>	<b>2.8<math>\pm</math>0.05</b>	<b>3.0<math>\pm</math>0.06</b>
	BB	<b>3.0<math>\pm</math>0.02</b>	<b>2.6<math>\pm</math>0.37</b>	<b>0.4<math>\pm</math>0.0</b>	<b>0.5<math>\pm</math>0.01</b>	<b>3.4<math>\pm</math>0.01</b>	<b>3.1<math>\pm</math>0.37</b>
	DR	<b>4.2<math>\pm</math>0.08</b>	<b>1.9<math>\pm</math>0.05</b>	<b>0.5<math>\pm</math>0.02</b>	<b>0.5<math>\pm</math>0.04</b>	<b>4.7<math>\pm</math>0.09</b>	<b>2.4<math>\pm</math>0.08</b>
Control	TS	<b>1.3<math>\pm</math>0.03</b>	<b>1.5<math>\pm</math>0.03</b>	<b>0.5<math>\pm</math>0.03</b>	<b>0.4<math>\pm</math>0.01</b>	<b>1.8<math>\pm</math>0.05</b>	<b>1.9<math>\pm</math>0.03</b>
	BB	<b>2.1<math>\pm</math>0.05</b>	<b>1.8<math>\pm</math>0.09</b>	<b>0.3<math>\pm</math>0.0</b>	<b>0.4<math>\pm</math>0.01</b>	<b>2.4<math>\pm</math>0.05</b>	<b>2.2<math>\pm</math>0.08</b>
	DR	<b>0.9<math>\pm</math>0.03</b>	<b>1.1<math>\pm</math>0.10</b>	<b>0.2<math>\pm</math>0.01</b>	<b>0.3<math>\pm</math>0.02</b>	<b>1.0<math>\pm</math>0.02</b>	<b>1.4<math>\pm</math>0.12</b>

\* eCO<sub>2</sub>- Elevated CO<sub>2</sub> (550ppm); eT- Elevated Temperature (+3°C); DAP: Days after planting; TS- Thompson Seedless; BB-Bangalore ;DR-Dogridge

**Table 2: ANOVA for chlorophyll content of grape varieties**

Factors	Chlorophyll a		Chlorophyll b		Total chlorophyll	
	50 DAP	80DAP	50 DAP	80DAP	50 DAP	80DAP
V	<b>17.944**</b>	<b>2.115 **</b>	<b>0.074**</b>	<b>0.043*</b>	<b>17.022**</b>	<b>2.602**</b>
T	<b>9.280**</b>	<b>4.581**</b>	<b>0.439**</b>	<b>0.299**</b>	<b>13.169**</b>	<b>7.018**</b>
V*T	<b>3.415**</b>	<b>0.227NS</b>	<b>0.206**</b>	<b>0.039**</b>	<b>4.617**</b>	<b>0.366NS</b>
LSD (V)	<b>0.084</b>	<b>0.315</b>	<b>0.029</b>	<b>0.057</b>	<b>0.087</b>	<b>0.355</b>
LSD (T)	<b>0.097</b>	<b>0.364</b>	<b>0.033</b>	<b>0.088</b>	<b>0.101</b>	<b>0.410</b>
LSD (V*T)	<b>0.168</b>	<b>NS</b>	<b>0.058</b>	<b>0.153</b>	<b>0.175</b>	<b>NS</b>
CV	<b>4.62</b>	<b>24.22</b>	<b>8.39</b>	<b>22.75</b>	<b>4.04</b>	<b>21.55</b>

V- Varieties; T- Treatments; \*\*, \*significance at  $p \leq 0.01$ ,  $0.05$  respectively; NS-Non significant

### **Conclusion**

It is concluded that, the increase in atmospheric carbon dioxide concentration (eCO<sub>2</sub>) and temperature associated with future climates are expected to affect positively on chlorophyll content which can stimulate photosynthetic rate and ultimately leads to increase in biomass of crop.

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