

## Effects of Light Quality on Growth and Morphogenesis of 'Muscat Bailey A' Grapevine *in vitro*

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### Summary

Two-node microcuttings were used in this study to evaluate the effect of different qualities of LED lights on growth and morphogenesis of 'Muscat Bailey A' grapevines cultured *in vitro*. Among red-blue mix light treatments, plants in the treatments of 9R, 8R1B, 6R3B and 2R7B had higher percentages of shoot regeneration but there was no significant difference in the percentage of rooting among treatments. The fresh weight and dry weight of leaf, leaf number, leaf area and average primary root length in the 8R1B treatment were the highest. Plants in the treatment of 9R had the highest shoot length, internode length and primary root number. The fresh weight and dry weight of root and secondary root number in 6R3B treatment were the highest. Among red-blue ratio with infrared light treatments, the highest percentage of shoot regeneration was 100% for the treatments of 5R3B1IR, 3R5B1IR and 4R3B2IR, and the lowest one was obtained in the treatment of 3R4B2IR which was only 53.3%. The highest percentage of rooting, shoot fresh weight, shoot dry weight, leaf number, node number, shoot length and internode length were obtained in the treatment of 7R1B1IR. In summary, growth and development of grapevine plantlets were better under higher ratio of red to blue light. Therefore, higher ratio of red to blue light should be used during the early stage of *in vitro* microcutting of grapevines.

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## Introduction

Grape is one of the most important fruit crops in the world which is destined for fresh consumption and wine making. The major cultivars of grape have originated from *Vitis vinifera* (Burger *et al.*, 2009). 'Muscat Bailey A', a hybrid of Bailey and Muscat Hamburg grapevines, is a red wine grape that was developed in Japan.

*In vitro* method for mass propagation of grapevines was mainly developed by using different explant sources (Barlass and Skene, 1978). Growth and development of *in vitro* grown plants are regulated by various micro-environmental factors. However, light quality is a one of the most important factors that affects the growth and morphogenesis of *in vitro* plants (Huges, 1981). Fluorescent lamps are the most frequently used light sources for tissue culture plants. Recently, light-emitting diodes (LEDs) have been considered as alternative light sources for *in vitro* propagation of plants. LEDs have many advantages, including small size, long life, specific wavelength, narrow bandwidth, low thermal energy, as well as adjustable light intensity and quality (Schuerger *et al.*, 1997; Tazawa, 1999). LEDs have been used for studies in many areas such as photosynthesis (Tennessee *et al.*, 1994), chlorophyll contents (Tripathy and Brown, 1995; Tanaka *et al.*, 1998), and growth and morphogenesis (Li *et al.*, 2010; Poudel *et al.*, 2008).

Many studies showed that LED lighting system is more suitable for plant growth and development than that of a fluorescent lamp. The aim of this study was to evaluate the effect of mono-wavelength and different red-blue ratios of LED lights on the growth and morphogenesis of 'Muscat Bailey A' grapevines *in vitro*.

## Materials and methods

### Materials

'Muscat Bailey A' grapevines (*Vitis labruscana* × *V. vinifera*) were used as explant sources. Young shoots were collected from healthy plants. Shoot segments (ca. 1.5 cm) were surface sterilized with 70% ethanol, agitated in 1% sodium hypochlorite for 15 minutes and washed three times with sterile water before cultured in a medium containing ½ strength MS (Murashige and Skoog, 1962), 30 g l<sup>-1</sup> sucrose, 8 g l<sup>-1</sup> agar, 2 ppm IBA and 200 ppm activated charcoal. The pH of medium was adjusted to 5.8 prior to sterilization in an autoclave at 121 °C for 15 minutes. Cultures were incubated at 26 ± 1 °C with a 16-h photoperiod. The shoots proliferated were used for further experiments.

## Methods

Middle part of two-node shoots were excised from the proliferated shoots as described above and cultured in the same medium. The cultures were kept under different LED light sources (Nano Bio Light Company) and incubated at  $26 \pm 1$  °C with a 16-h photoperiod and intensity of  $56 \mu\text{mole/m}^2/\text{sec}$  for 30 days. Five two-node shoots in one bottle was one replicate and there were three replicates for each treatment. The following experiments were conducted:

### 1. Effects of red-blue mix light

9R – Red 100%

8R1B – Red 91%, Blue 9%

6R3B – Red 72%, Blue 28%

3R6B – Red 25%, Blue 75%

2R7B – Red 12%, Blue 88%

1R8B – Red 5%, Blue 95%

9B – Blue 100%

CW – Cool white (5500 K)

### 2. Effects of red-blue ratio with concurrent infrared (1 IR or 2 IR as the base)

7R1B1IR – Red 87%, Blue 9%, Infrared 4 %

6R2B1IR – Red 76%, Green 1%, Blue 19%, Infrared 4%

5R3B1IR – Red 58%, Green 1%, Blue 38%, Infrared 3%

3R5B1IR – Red 34%, Green 1%, Blue 62%, Infrared 3%

2R6B1IR – Red 21%, Green 1%, Blue 75%, Infrared 3%

1R7B1IR – Red 10%, Green 2%, Blue 85%, Infrared 3%

4R3B2IR – Red 58%, Green 1%, Blue 31%, Infrared 10%

3R4B2IR – Red 45%, Green 1%, Blue 43%, Infrared 11%

CW – Cool white (5500 K)

## Measurement and analysis

### 1. Plant growth parameters:

1.1) The percentage of shoot regeneration and rooting

1.2) Fresh weight: fresh weight of plants were measured by separating into leaf, shoot and root parts

1.3) Dry weight: leaves, shoots and roots were dried at 60 °C for 72 hour and dry weights were then measured

1.4) The numbers of leaf, node, primary root and secondary root were counted

1.5) Shoot length and primary root length were measured by a ruler

1.6) Leaf color was measured by Leaf Color Meter (model CT-101, SME, FHK) and expressed as relative values

1.7) Leaf area was measured by Leaf Area Meter (AM 300, ADC BioScientific Ltd.)

2. Statistical analysis:

Data were subjected to one-way analysis of variance (ANOVA) using SAS version 9.0 and mean separation was conducted using Least Significant Difference test at  $p \leq 0.05$ .

## Results

### 1. Effects of red-blue mix light

Plants were grown under red-blue mix light for 30 days. There was a significant difference in the percentage of shoot regeneration but no difference in the percentage of rooting (Table 1). Plants in the treatments of 9R, 8R1B, 6R3B and 2R7B had the highest percentage of shoot regeneration (100%), while the lowest (73.33%) was obtained in the treatment of CW (Table 1). Plants in the 8R1B and 6R3B treatments had a higher fresh weight of whole plant than those in other treatments. The fresh weight and dry weight of leaf and fresh weight of root in the 8R1B treatment were the highest. The highest dry weight of whole plant and dry weight of root were 30.41 mg and 10.15 mg for the treatment of 6R3B and the lowest ones were obtained in the treatments of 9B and CW. Plants in treatments of 9R and 8R1B had a higher shoot fresh weight and shoot dry weight (Table 2).

There were significant differences in morphogenesis among treatments (Table 3). Plants in the treatments of 8R1B and 3R6B had higher leaf numbers than those in other treatments. Moreover, the highest leaf area was 1,147 mm<sup>2</sup> for the treatment of 8R1B and lower leaf areas were obtained in the treatments of 9B and CW. There was a slight difference in leaf color. However, plants in the 9R treatment had the lowest leaf color (0.84). Among different light sources, plants in the treatments of 9R, 8R1B and 3R6B had higher node numbers than those in other treatments. The highest shoot length and internode length were 10.15 cm and 2.23 cm for the treatment of 9R. However, plants in treatments of 2R7B, 1R8B and 9B had lower shoot lengths and internode lengths (Table 3).

Table 1. Effects of red-blue mix light on the percentage of shoot regeneration and rooting of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Shoot regeneration (%)				Rooting (%)		
9R	100.00	±	0.00 <sup>z</sup>	a <sup>y</sup>	100.00	±	0.00 a
8R1B	100.00	±	0.00	a	93.33	±	11.55 a
6R3B	100.00	±	0.00	a	100.00	±	0.00 a
3R6B	100.00	±	0.00	a	100.00	±	0.00 a
2R7B	100.00	±	0.00	a	80.00	±	20.00 a
1R8B	86.67	±	11.55	b	86.67	±	11.55 a
9B	93.33	±	11.55	ab	93.33	±	11.55 a
CW	73.33	±	11.55	c	80.00	±	20.00 a

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

There was a slight difference in the primary root number. The highest primary root number was 4.47 for the treatment of 9R. Plants in treatments of 8R1B, 6R3B and 3R6B had higher secondary root numbers than in other treatments. Plants in 8R1B and 3R6B treatments also had higher primary root lengths and a lower secondary root number and primary root length were obtained in the treatment of CW (Table 3).

Table 2. Effects of red-blue mix light on fresh weight and dry weight of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Whole plant				Leaf							
	Fresh weight (mg)		Dry weight (mg)		Fresh weight (mg)	Dry weight (mg)						
9R	257.67±	40.46 <sup>z</sup>	ab <sup>y</sup>	17.66±	11.05	abc	67.11±	13.46	bc	8.25±	2.19	bc
8R1B	359.96±	147.77	a	29.04±	11.74	ab	121.19±	45.25	a	15.77±	6.80	a
6R3B	314.24±	122.06	a	30.41±	13.16	a	110.97±	40.61	ab	14.88±	6.15	ab
3R6B	268.60±	63.91	ab	26.95±	6.32	ab	115.17±	24.22	ab	14.97±	3.21	ab
2R7B	150.10±	45.15	bc	15.01±	4.59	bc	68.05±	24.65	bc	8.74±	2.77	bc
1R8B	242.64±	120.30	abc	17.66±	2.36	abc	85.97±	15.38	abc	10.38±	1.67	abc
9B	117.52±	67.92	bc	11.38±	6.05	c	52.07±	31.53	c	6.38±	3.65	c
CW	93.83±	44.25	c	9.59±	3.99	c	40.39±	18.60	c	5.02±	2.23	c

Table 2 (continued). Effects of red-blue mix light on fresh weight and dry weight of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Shoot		Root	
	Fresh weight (mg)	Dry weight (mg)	Fresh weight (mg)	Dry weight (mg)
9R	123.99±16.61 <sup>z</sup> a <sup>y</sup>	7.79±2.19 a	66.58± 11.46 abc	5.13±0.62 bcd
8R1B	116.15±39.24 a	7.17±4.92 a	122.62± 64.47 a	9.51±5.55 ab
6R3B	74.00±34.03 b	5.39±2.81 ab	129.27± 47.45 a	10.15±4.23 a
3R6B	65.30±20.34 bc	4.58±1.40 ab	88.13± 20.18 abc	7.40±1.78 abc
2R7B	39.02±13.22 bc	2.68±0.68 b	54.59± 5.19 abc	4.45±0.40 cd
1R8B	41.54± 7.25 bc	2.73±0.30 b	115.13±102.67 ab	4.55±0.45 cd
9B	30.55±14.97 c	2.00±0.70 b	34.90± 21.61 bc	3.01±1.73 cd
CW	32.46±16.94 c	2.09±0.97 b	26.93± 11.74 c	2.48±0.84 c

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

Table 3. Effects of red-blue mix light on morphology of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Leaf no.	Total leaf area (mm <sup>2</sup> )	Leaf color (relative values)
9R	4.67 ± 0.42 <sup>z</sup> ab <sup>y</sup>	685.30 ± 164.54 abc	0.84 ± 0.08 b
8R1B	4.85 ± 0.88 a	1147.00 ± 380.05 a	0.90 ± 0.07 ab
6R3B	4.60 ± 1.20 ab	1048.60 ± 424.78 ab	0.95 ± 0.03 a
3R6B	5.40 ± 0.72 a	1086.30 ± 195.90 ab	0.97 ± 0.02 a
2R7B	3.40 ± 0.53 bc	637.93 ± 245.12 bc	0.91 ± 0.06 ab
1R8B	4.27 ± 0.68 abc	805.58 ± 180.16 abc	0.93 ± 0.03 ab
9B	2.97 ± 0.57 c	470.98 ± 255.17 c	0.94 ± 0.01 a
CW	3.28 ± 0.82 c	435.73 ± 190.76 c	0.94 ± 0.05 a

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

Table 3 (continued). Effects of red-blue mix light on morphology of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Node no.	Shoot length (cm)	Average internode length (cm)
9R	4.53 ± 0.31 <sup>z</sup> a <sup>y</sup>	10.15 ± 0.95 a	2.23 ± 0.12 a
8R1B	4.85 ± 0.88 a	8.26 ± 1.41 b	1.71 ± 0.03 b
6R3B	4.40 ± 1.11 ab	4.38 ± 1.75 c	0.96 ± 0.19 c
3R6B	5.33 ± 0.42 a	3.68 ± 0.60 cd	0.71 ± 0.11 de
2R7B	3.27 ± 0.64 bc	1.95 ± 0.50 e	0.56 ± 0.06 e
1R8B	4.13 ± 0.55 ab	2.33 ± 0.29 e	0.57 ± 0.01 e
9B	2.90 ± 0.61 c	1.92 ± 0.45 e	0.68 ± 0.04 de
CW	3.19 ± 0.76 bc	2.55 ± 0.94 de	0.78 ± 0.16 cd

  

Light source	Primary root no.	Secondary root no.	Average primary root length (cm)
9R	4.47 ± 1.70 <sup>z</sup> a <sup>y</sup>	33.53 ± 6.89 ab	3.78 ± 0.27 abc
8R1B	3.02 ± 0.78 ab	39.32 ± 13.89 a	4.65 ± 0.73 00a
6R3B	4.07 ± 2.01 ab	45.80 ± 10.35 a	3.89 ± 0.23 abc
3R6B	2.87 ± 1.15 ab	35.87 ± 6.87 a	4.42 ± 0.30 00a
2R7B	2.16 ± 0.36 b	33.33 ± 7.67 ab	3.43 ± 0.68 bcd
1R8B	2.22 ± 1.20 b	31.87 ± 4.15 abc	4.09 ± 0.14 0ab
9B	2.53 ± 0.76 ab	19.82 ± 11.37 bc	3.07 ± 0.98 0cd
CW	2.11 ± 0.84 b	17.67 ± 6.81 c	2.62 ± 0.43 00d

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

## 2. Effects of red-blue ratio with concurrent infrared (1 IR or 2 IR as the base)

There were significant differences in the percentages of shoot regeneration and rooting among different LEDs (Table 4). The highest percentage of shoot regeneration was 100% for the treatments of 5R3B1IR, 3R5B1IR and 4R3B2IR, and the lowest one (53.33%) was obtained in the treatment of 3R4B2IR. The percentage of rooting in the 7R1B1IR treatment was the highest. There were significant differences in whole plant fresh weight and whole plant dry weight (Table 5). The highest whole plant fresh weight and whole plant dry weight were 190.95 mg and 18.49 mg for the treatment of 6R2B1IR and the lowest ones (53.46 mg and 7.05 mg) were

obtained in the treatment of 3R4B2IR. Plants in the treatments of 7R1B1IR, 6R2B1IR and 2R6B1IR had higher leaf fresh weights than those in other treatments and the lowest one was obtained in the treatment of 3R4B2IR. There was no significant difference in the leaf dry weight.

Table 4. Effects of red-blue ratio with concurrent infrared (1 IR or 2 IR as the base) on the percentage of shoot regeneration and rooting of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Shoot regeneration (%)	Rooting (%)
7R1B1IR	86.67 ± 23.09 <sup>z</sup> ab <sup>y</sup>	100.00 ± 0.00 a
6R2B1IR	93.33 ± 11.55 ab	86.67 ± 23.09 ab
5R3B1IR	100.00 ± 0.00 a	66.67 ± 41.63 ab
3R5B1IR	100.00 ± 0.00 a	80.00 ± 20.00 ab
2R6B1IR	73.33 ± 23.09 bc	66.67 ± 11.55 ab
1R7B1IR	73.33 ± 11.55 bc	60.00 ± 34.64 ab
4R3B2IR	100.00 ± 0.00 a	86.67 ± 11.55 ab
3R4B2IR	53.33 ± 23.09 c	53.33 ± 30.55 b
CW	73.33 ± 11.55 bc	80.00 ± 20.00 ab

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

The fresh weight of shoot and dry weight of shoot in the 7R1B1IR treatment were the highest. Plants in the treatments of 1R7B1IR and 3R4B2IR had lower shoot fresh weights and shoot dry weights (Table 5). There were significant differences in root fresh weight and root dry weight. The highest root fresh weight was 87.10 mg for the treatment of 6R2B1IR and the lowest one (19.54 mg) was obtained in the treatment of 3R4B2IR. The dry weight of root in the treatment of 5R3B1IR was the highest (Table 5).

There was a slight difference in leaf number. Plants in the treatments of 7R1B1IR and 6R2B1IR had higher leaf numbers than those in other treatments. Nevertheless, there were no significant differences in leaf area and leaf color (Table 6). Plants grown under different red-blue ratio with infrared LED lights had significant differences in node number, shoot length and internode length (Table 6). The highest node number and shoot length were 4.15 and 4.44



cm for the treatment of 7R1B1IR and the lowest ones (2.28 an and 1.17 cm) were obtained in the treatment of 1R7B1IR. The internode length in the 7R1B1IR treatment also was the highest (1.06 cm). There was no significant difference in the primary root number (Table 6). The number of secondary root in the 2R6B1IR treatment and the primary root length in the 3R5B1IR treatment were the highest, respectively. Plants in the treatment of 3R4B2IR had a lower secondary root number and primary root length than those in other treatments.

Table 5. Effects of red-blue ratio with concurrent infrared (1 IR or 2 IR as the base) on fresh weight and dry weight of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Whole plant		Leaf	
	Fresh weight (mg)	Dry weight (mg)	Fresh weight (mg)	Dry weight (mg)
7R1B1IR	161.54± 61.93 <sup>z</sup> ab <sup>y</sup>	15.89± 4.62 abc	71.73±26.69 a	10.25±3.55 a
6R2B1IR	190.95± 41.09 a	18.49± 2.14 a	70.72±14.55 a	10.27±1.43 a
5R3B1IR	157.17±122.76 ab	16.35±10.99 ab	65.36±51.46 ab	9.85±6.30 a
3R5B1IR	137.50± 25.63 abc	13.97± 3.16 abc	59.07±15.70 abc	8.54±1.82 a
2R6B1IR	89.47± 54.81 bc	15.84± 7.35 abc	82.29±50.90 a	11.50±7.70 a
1R7B1IR	86.45± 21.76 bc	8.53± 1.90 bc	37.30± 6.55 bc	5.68±1.18 a
4R3B2IR	132.81± 37.67 abc	15.13± 4.83 abc	58.49± 9.19 abc	9.63±3.00 a
3R4B2IR	53.46± 21.52 c	7.05± 2.67 c	31.72±10.26 c	5.27±1.43 a
CW	93.83± 44.25 bc	9.59± 3.99 abc	40.39±18.60 bc	5.02±2.23 a

  

Light source	Shoot		Root	
	Fresh weight (mg)	Dry weight (mg)	Fresh weight (mg)	Dry weight (mg)
7R1B1IR	54.04±19.72 <sup>z</sup> a <sup>y</sup>	4.08±1.48 a	57.87±32.50 abc	4.12±1.85 abc
6R2B1IR	46.36± 8.83 ab	3.79±0.67 ab	87.10±62.26 a	5.12±2.73 ab
5R3B1IR	35.52±26.97 abc	2.74±2.08 ab	74.05±25.52 ab	5.51±0.75 a
3R5B1IR	26.47± 6.85 bc	2.35±0.31 ab	51.96±11.67 abc	3.79±0.42 abc
2R6B1IR	40.00±20.30 abc	3.05±1.88 ab	50.52±19.58 abc	4.13±2.04 abc
1R7B1IR	18.59± 3.26 c	1.73±0.37 b	30.56±17.06 bc	2.56±1.38 bc
4R3B2IR	35.74±15.12 abc	2.87±0.90 ab	43.55±26.86 abc	2.94±1.48 abc
3R4B2IR	15.21± 7.12 c	1.58±0.55 b	19.54± 8.47 c	1.89±0.99 c
CW	32.46±16.94 abc	2.09±0.97 ab	26.93±11.74 bc	2.48±0.84 bc

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

Table 6. Effects of red-blue ratio with concurrent infrared (1 IR or 2 IR as the base) on morphology of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Leaf no.	Total leaf area (mm <sup>2</sup> )	Leaf color (relative values)
7R1B1IR	4.04 ± 0.72 <sup>z</sup> a <sup>y</sup>	749.68 ± 254.79 a	1.00 ± 0.10 a
6R2B1IR	3.98 ± 0.36 a	693.59 ± 136.75 a	0.95 ± 0.02 a
5R3B1IR	3.73 ± 1.14 ab	633.73 ± 546.74 a	0.96 ± 0.11 a
3R5B1IR	3.33 ± 0.50 abc	538.80 ± 136.06 a	1.05 ± 0.02 a
2R6B1IR	2.89 ± 0.77 bc	773.15 ± 450.00 a	0.98 ± 0.08 a
1R7B1IR	2.36 ± 0.13 c	360.02 ± 59.66 a	1.01 ± 0.06 a
4R3B2IR	3.48 ± 0.20 ab	726.57 ± 244.38 a	0.97 ± 0.02 a
3R4B2IR	2.67 ± 0.29 bc	326.67 ± 106.14 a	0.96 ± 0.08 a
CW	3.28 ± 0.82 ab	435.73 ± 190.76 a	0.94 ± 0.05 a

  

Light source	Node no.	Shoot length (cm)	Average internode length (cm)
7R1B1IR	4.15 ± 0.88 <sup>z</sup> a <sup>y</sup>	4.44 ± 1.03 a	1.06 ± 0.04 a
6R2B1IR	3.98 ± 0.36 ab	3.48 ± 0.35 ab	0.83 ± 0.05 b
5R3B1IR	3.47 ± 1.42 abc	2.47 ± 1.64 bcd	0.64 ± 0.18 cd
3R5B1IR	3.27 ± 0.42 abcd	1.71 ± 0.40 cd	0.48 ± 0.07 de
2R6B1IR	2.89 ± 0.77 bcd	1.66 ± 0.61 cd	0.49 ± 0.09 de
1R7B1IR	2.28 ± 0.05 d	1.17 ± 0.13 d	0.47 ± 0.08 de
4R3B2IR	3.42 ± 0.18 abcd	2.19 ± 0.36 bcd	0.60 ± 0.05 de
3R4B2IR	2.58 ± 0.14 cd	1.22 ± 0.33 cd	0.46 ± 0.08 0e
CW	3.19 ± 0.76 abcd	2.55 ± 0.94 bc	0.78 ± 0.16 bc

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

Table 6 (continued). Effects of red-blue ratio with concurrent infrared (1 IR or 2 IR as the base) on morphology of 'Muscat Bailey A' grapevines after 30 days of *in vitro* microcutting.

Light source	Primary root no.	Secondary root no.	Average primary root length (cm)
7R1B1IR	2.82 ± 0.50 <sup>z</sup> a <sup>y</sup>	33.26± 6.73 ab	3.09 ± 0.81 ab
6R2B1IR	2.84 ± 1.37 a	33.67± 7.96 ab	3.18 ± 1.48 ab
5R3B1IR	2.63 ± 1.10 a	31.27± 7.55 abc	3.44 ± 1.64 ab
3R5B1IR	1.97 ± 0.91 a	28.36± 8.88 abcd	3.67 ± 0.27 a
2R6B1IR	2.50 ± 1.32 a	38.39±14.07 a	2.79 ± 0.32 ab
1R7B1IR	1.57 ± 0.40 a	19.33± 3.75 bcd	3.03 ± 0.61 ab
4R3B2IR	2.40 ± 0.77 a	28.81±12.81 abcd	2.87 ± 0.49 ab
3R4B2IR	2.33 ± 0.58 a	14.11± 4.73 d	1.99 ± 0.95 b
CW	2.11 ± 0.84 a	17.67± 6.81 cd	2.62 ± 0.43 ab

<sup>z</sup>Mean ± standard deviation.

<sup>y</sup>Means separation within each column by the Least Significant Difference test at  $p \leq 0.05$ .

## Discussions

Red light promoted axillary shoot elongation in many woody plants (Norton *et al.*, 1988 and Noè *et al.*, 1998) while blue light inhibited it (Baraldi *et al.*, 1992). It has been found that red light is a considerable light source for shoot and stem elongation (Schuerger *et al.*, 1997). In contrast, blue light is important for photosynthesis, stomatal opening and chlorophyll biosynthesis (Tibbitts *et al.*, 1983). In this study, it was found that under 9R, 8R1B or 7R1B1IR light treatments, grapevine explants had higher shoot fresh weight, shoot dry weight, shoot length and internode length (Table 2; Table 3; Table 5; Table 6). On the other hand, the treatment of 9B had the lowest shoot fresh weight, shoot dry weight, node number and shoot length (Table 2; Table 3). The shoot length and internode length were decreased when the RB ratios were decreased (Table 3). Previous experiments found that shoot elongation and internode length were the greatest when red light was used (Heo *et al.*, 2006; Kim *et al.*, 2004; Poudel *et al.*, 2008). However, Li *et al.* (2010) found that stem length of upland cotton plantlets was higher when plantlets were cultured under B : R = 1 : 1 LED light. In addition, Heo *et al.* (2002) reported that stem length in salvia was obviously decreased under red light as compared to other treatments and stem length in marigold was higher in blue light treatment. The difference with respect to the synergistic interaction between blue and red light receptors and phytochrome can

promote or inhibit stem elongation according to species (Kim *et al.*, 2004).

Schuerger *et al.* (1997) found that blue light can be an influence on growth inhibition and changes in stem and leaf morphogenesis of pepper plants. In 'Muscat Bailey A' grapevine, leaf fresh weight, leaf dry weight, leaf number and leaf area showed the greatest values when 8R1B light was used (Table 2; Table 3). In *Withania Somnifera*, application of a mixture of red and blue light showed the highest number of leaves (Lee *et al.*, 2007). Shin *et al.* (2008) found that the biomass and leaf growth of *Doritaenopsis* plants were significantly increased under the mixture of red and blue LED treatment as compared with fluorescent and red or blue LED treatments. The growth of leaf area is controlled by red light (Wu *et al.*, 2007). However, Li *et al.* (2010) detected that the upland cotton leaf area under blue light was the largest. In contrast, Mortensen and Stromme (1987) showed that blue light can reduce the leaf area of *Chrysanthemum*. It was assumed that blue light was important for leaf expansion, but it depends on plant species (Li *et al.*, 2010).

Light quality can affected rooting (Moon *et al.*, 2006). Lee *et al.* (2007) reported that root induction is probably dependent on the light quality. Blue light can inhibit root development of birch shoot *in vitro* (Pinker *et al.*, 1989). In contrast, in *Vitis* plants, when long-day applications of red light were used, root number and root length were increased as compared with blue light treated plants (Chee and Pool, 1989). Longer roots were observed under PGF light for 'Hybrid Franc' and 'Kadainou R-1' or red LED light for 'Ryuukyuganebu' grapevine (Poudel *et al.*, 2008). However, Lian *et al.* (2002) found that in *Lilium* oriental hybrid 'Pesaro', the number of root, fresh weight and dry weight of root were higher in the bulblets cultured under red light combined with blue light and fluorescent light. Similarly, it was found that root fresh weight and root length were higher in the plants cultured in the 8R1B treatment in this study (Table 2; Table 3). However, root number was higher in plants cultured under the 9R light (Table 3).

In summary, a mixture of red and blue light will be necessary for normal growth and development of grapevines. Especially, the 8R1B treatment showed better results for *in vitro* culture of 'Muscat Bailey A' grapevines than mono-wavelength and other mixed radiations. Blue light was required for chlorophyll biosynthesis and stomata development. Further research on this aspect would enhance the efficiency of *in vitro* culture techniques combine with the application of LEDs system for grapevines.

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## 光質對離體培養之'貝利 A'葡萄生長及形態之影響

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關鍵字:葡萄、發光二極體、生長及形態

**摘要：**本研究使用雙節微體扦插來評估不同 LED 光源對'Muscat Bailey A'葡萄瓶內生長及形態的影響。結果顯示，以紅藍混合光處理下，9R、8R1B、6R3B 及 2R7B 處理組之萌芽率較高，但發根率則處理間並無顯著差異。8R1B 處理組有較高的葉片鮮乾重、葉片數、葉面積及主根長度。於 9R 處理組有較高的枝條長、節間長與主根數。而於 6R3B 處理組有較高的根鮮乾重及側根數。而紅藍光伴隨遠紅光處理下，於 5R3B1IR、3R5B1IR 及 4R3B2IR 處理組有 100% 的萌芽率，而 3R4B2IR 處理組萌芽率最低，只有 53.3%。7R1B1IR 處理組有較高的發根率、枝條鮮乾重、葉片數、節數、枝條長度與節間長。綜合而論，紅/藍光比率較高時，葡萄培植體生長及發育較佳，因此，於葡萄微體扦插初期發育階段應使用較高比率之紅光。

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