

# Effects of different light quality and GA<sub>3</sub> concentration on tobacco seedling growth, root characteristics, and carbon and nitrogen metabolism

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Abstract

Light quality and gibberellins affect growth, development and morphogenesis of plants. In order to improve the quality of tobacco seedlings under greenhouse conditions, the influence of different light sources and their interaction with foliar-applied gibberellic acid (GA<sub>3</sub>) on key growth and metabolic parameters in tobacco were examined. Light sources included white, red and blue lights while  $GA_3$  was applied in three concentrations, i.e., 0, 50 and 100 mg L<sup>1</sup>. The survival rate of the seedlings was observed after transplanting. Morphological parameters like plant height, stalk diameter, and length, width and number of leaves increased maximally with higher concentration of GA<sub>3</sub> under all three light regimes, however, light sources did not show any visible difference. Further, root vitality, chlorophyll content and the activities amylase and nitrate reductase increased significantly due to GA3 application and achieved highest values with blue light + 100 mg  $L^{-1}$  GA<sub>3</sub>. Supplementation of red and blue light along with foliar application of GA<sub>3</sub> significantly increased the growth parameters including seedling height, stem diameter, root-shoot ratio, leaf length and root vitality. The contents of carbon metabolites including starch, soluble sugars, and reducing sugars improved significantly in plants grown under red and blue lights in combination with foliar application of GA<sub>3</sub>. The results of the present study depict that growing tobacco under blue light and the application GA<sub>3</sub> is beneficial in terms of achieving increased growth, root characteristics, carbon and nitrogen metabolism of tobacco seedlings.

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# Introduction

Light plays a regulatory role in plant growth and development, and different light qualities have different effects on plant growth and morphogenesis (Franklin, 2009; Shafiq et al., 2021). The growth and development of tobacco seedlings should not have only suitable temperature and water content, but also need sufficient light. Light is the source of energy for photosynthesis in plants. Light quality and its intensity directly affect the photosynthetic performance and the type, quantity and metabolic transformation of assimilation products (Ke et al., 2011; Shafiq et al., 2021). Blue light inhibits the vertical growth of plants while it promotes lateral growth, but in contrast red light imparts opposite effects. It has been shown that the plant height decreased due to blue light in rice seedlings while stem diameter and uniformity increased, and the red-light treated seedlings were slender and irregular (Ni, 1980). Blue light

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(445 nm~470 nm) is considered as most effective in inducing negative effects on growth of plant roots (Han et al., 2005; Li et al., 2020). Blue light induces cell division and improves elongation rate, root absorption area as well as root activity (Chen et al., 2015). Chen et al. (2015) have demonstrated that blue light treatment can enhance the root activity of peanut seedlings, while red light (610 nm ~ 660 nm) treatment decreased root vigor. The roots of blue light-treated peanut seedlings have a small distribution range, but the root activity was higher (Yan et al., 2014). It has been shown that blue light is beneficial to the synthesis of chlorophyll a, which is beneficial to plant growth (Mao et al., 1991; Shafiq et al., 2021). Light-filling treatment improves the quality of seedlings to a certain extent, and shortens the growth time of floating seedlings, but the difference between the emergence rate of light-filled tobacco seedlings and conventional seedlings is not significant (Huang et al., 2014).

Plant hormones regulate growth and development of plants.  $GA_3$  is actively involved in many physiological processes of plant growth and development. Its most significant physiological effect is promotion of plant growth and development through cell proliferation and cell growth (Li et al., 2004; Martins et al., 2022). GA<sub>3</sub> can also effectively activate genes related to IAA synthesis in plants, increase IAA content in plants, induce amylase synthesis and secretion, promote related physiological and biochemical reactions, increase metabolic rate, and also initiate seed germination and promote seedling rooting (Zhou et al., 2004; Nguyen et al., 2019). Gibberellin promotes increase in fresh weight, volume and stem segment of tobacco by increasing tobacco tissue cells and altering metabolic intensity. Research studies have shown that GA<sub>3</sub> treatment significantly promoted the growth and development of amaranth seedlings (Zhang et al., 2009). GA<sub>3</sub> promotes plant growth and development by increasing the activity of hydrolase (Gu et al., 2003). Certain concentrations of GA<sub>3</sub> can increase the germination rate of wild jasmine seeds and root growth of seedlings, but it has little effect on seedling diameter and seedling height (Wang et al., 2013). In addition,  $GA_3$  application has been reported to increase plant height, leaf size and root-shoot ratio of *Platycodon grandiflorum* significantly (Long et al., 2010). GA<sub>3</sub> significantly increased the root length, seedling height and biomass of tobacco seedlings under drought and low temperature stress (Kang et al., 2014).

Tobacco is an important industrial crop in China. It is grown on a large area in China and is an important economic crop that increases farmers' income, meets people's needs, enhances the national economy and earns foreign exchange through exports. Successful tobacco seedling cultivation is a key link in tobacco planting. In order to increase land utilization rate, increase tobacco multi-cropping index, and effectively prevent and control soil-borne diseases and insect pests, the current national tobacco-planting area has realized intensive production methods of concentrated seedling and field transplanting (Zou et al., 2014). Cultivating high-quality tobacco seedlings is the prerequisite and guarantees for producing high-quality tobacco leaves. In recent years, scientists have conducted extensive research on seedling substrate, seedling tray, fertilization amount, fertilization method, nutrient solution, temperature and humidity control in the greenhouse and seedling control technology (Lencucha et al., 2022).

In this study, the effects of light quality and  $GA_3$  on agronomic traits, root growth, root activity, key enzymes and products of carbon and nitrogen metabolism were studied by supplementing different light quality and spraying different concentrations of  $GA_3$ , in order to provide a practical basis for cultivating high quality tobacco seedlings.

# **Material and Methods**

### **Experimental design**

Seeds of tobacco variety Yunyan 99 were provided by the Yunnan Academy of Agricultural Sciences. The experiment was conducted at the Tobacco Leaf Nursery Workshop of Yanwan Village, Yungaisi Town, Zhen'an County, Shangluo City, Shaanxi Province-China. The experimental set-up comprised three different light quality treatments and three GA<sub>3</sub> concentrations under each light quality condition. A total of 12 trays for each treatment, and 108 trays for 9 treatments were arranged according to the experimental layout. Except for the shearing and smelting of seedlings, other management technical measures were implemented in accordance with the local floating nursery management regulations. The specific test design is shown in **Table 1**.

For each light quality treatment, lamps were used and the surrounding of the seedling pool was shielded by a black light-shielding material. The light period was 12 h d<sup>-1</sup>. After sowing, the seedling trays were placed in the pool to start different light quality treatments. As the tobacco seedlings gradually grew taller and the position of the tubes was adjusted so that the height from the seedlings was controlled between 28 cm and 30 cm.  $GA_3$  solution was sprayed onto the leaves after every 10 days making a total of three sprays.

Treatment	Light quality	$GA_3$ concentration (mg·L <sup>-1</sup> )
CK	White LED light source	Spray 0 mg·L <sup><math>1</math></sup> GA <sub>3</sub>
T1	(36 W, 660 nm, light intensity at 0.3 m under the lamp of 180	Spray 50 mg·L <sup>-1</sup> GA <sub>3</sub>
Т2	µmol·m <sup>-2</sup> ·s <sup>-1</sup> )	Spray 100 mg·L <sup>-1</sup> GA <sub>3</sub>
Т3	Red LED light source	Spray 0 mg·L <sup>-1</sup> GA <sub>3</sub>
T4	(36 W, 660 nm, light intensity at 0.3 m under the lamp of 180	Spray 50 mg·L <sup>-1</sup> GA <sub>3</sub>
Т5	µmol·m <sup>-2</sup> ·s <sup>-1</sup> )	Spray 100 mg·L <sup>-1</sup> GA <sub>3</sub>
Т6	Blue LED light source	Spray 0 mg·L <sup>-1</sup> GA <sub>3</sub>
Τ7	(36 W, 450 nm, light intensity at 0.3 m under the lamp of 180	Spray 50 mg $L^1$ GA <sub>3</sub>
Т8	μmol·m <sup>-2</sup> ·s <sup>-1</sup> )	Spray 100 mg·L <sup>-1</sup> GA <sub>3</sub>

# Table 1. Treatment details

# Determination of the growth status of tobacco seedlings

At 63 days after sowing, 4 seedlings were randomly selected from each treatment and growth parameters like plant height, maximum leaf length, maximum leaf width, stem diameter, and number of leaves were measured. The whole plant was washed and divided into roots, stems and leaves. The main root length, lateral root length and lateral root number of each seedling were measured. The root volume was determined by the drainage method. After the fresh tobacco seedlings were dried at 100 °C for 30 min, they were consequently dried to constant weight at 60 °C, and the dry weight was taken to calculate the root-shoot ratio (Li, 2000).

### **Determination of root activity**

Root activity was determined by the TTC method at 42, 49, 56 and 63 days after sowing (Gao, 2006).

# **SPAD value determination**

At 42, 49, 56 and 63 days after sowing, 4 tobacco seedlings with the same growth and no obvious lesions were selected from each treatment. At 10:00~11:00 in the morning, fully expanded blades at the top of the tobacco seedlings were selected from top to bottom and were assayed for SPAD values using SPAD-502.

# Determination of key enzyme activities of carbon and nitrogen metabolism

Amylase activity was measured by the dinitrosalicylic acid method and nitrate reductase activity was determined by the sulfonamide colorimetric method (*in vivo* method) (Zou, 1995).

#### **Determination of metabolites**

The soluble sugar content and starch content were determined by the indolinone method. The reducing sugar content was determined by the dinitrosalicylic acid method and the soluble protein content was determined by the Coomassie Brilliant Blue G-250 staining method (Gao, 2006).

# Statistical analysis of data

Office 2013 version of Excel 2013 software was used for data collation and chart production. SPSS-19.0 and DPS software items were employed for the analysis of variance of the data for each parameter using multiple comparisons through Duncan's method.

# Results

# Effects of different light quality and GA<sub>3</sub> treatments on agronomic traits of tobacco seedlings

The results of agronomic traits of tobacco seedlings as affected by light and GA<sub>3</sub> are shown in **Table 2**. The treatments T1, T2, T4, T5, T7, and T8 had a significant effect on plant height of tobacco seedlings, and there was an increase of 28.18%, 62.71%, 29.48%, 55.45%, 24.68%, and 48.96%, respectively, over control (CK). The maximum leaf length of T2, T5 and T8 treated tobacco seedlings was significantly higher than that of CK treatment by 15.63%, 16.71% and 13.72%, respectively (**Table 2**). The maximum leaf width of T1, T2, T5 and T8 treated tobacco seedlings was significantly lower than that of CK treatment exhibiting a reduction of 17.40%, 22.69%, 20.73% and 22.69%, respectively. In T5 treatment, the root - crown ratio increased by 77.78% over that of CK treatment. The effect of T1 and T7 was also consistent on the root/shoot ratio. There was no significant difference in the stem diameter and leaf number of each treated seedlings (**Table 2**).

Table 2. Effects of different light quality and GA3 treatments on agronomic traits of 63-day old tobacco seedlir	igs
(Mean values bearing different letters within each column differ significantly at $P < 0.05$	

	Seedling height (cm)	The biggest leaf		—Stalk diameter	Number of	Root/	
Treatment		Length (cm)	Width (cm)	(cm)	leaves	shoot ratio	
СК	7.70c	14.72b	6.61a	0.613ab	8.13ab	0.18bc	
T1	9.87b	15.43ab	5.46b	0.610ab	7.83ab	0.24ab	
T2	12.53a	17.02a	5.11b	0.610ab	7.57b	0.13c	
Т3	8.27c	14.62b	6.41a	0.603b	8.03ab	0.18bc	
Τ4	9.97b	15.95ab	6.49a	0.650a	8.27ab	0.16bc	
T5	11.97a	17.18a	5.24b	0.587b	7.77ab	0.32a	
Т6	7.93c	14.56b	6.44a	0.601b	8.47ab	0.18bc	
Т7	9.60b	16.21ab	6.51a	0.667a	8.87a	0.24ab	
Т8	11.47a	16.74a	5.11b	0.611ab	7.9ab	0.11c	

# Effects of different light quality and GA<sub>3</sub> treatments on root vigor

Root vitality is the main indicator for judging the vitality of tobacco seedlings, and it is also an important factor in determining the survival rate of tobacco seedling transplanting. The root activity of each treatment increased first and then decreased, and peaked at 56 days after sowing. Compared with CK, the root activity of T7 treatment was significantly improved, increasing by 162.5%, 94.23%, 102.27%, and 107.87% at 42, 49, 56, and 63 days after sowing, followed by T8 treatment, wherein the root activity increased by 92.19%, 85.58%, 94.32%, and 77.17% at 42, 49, 56, and 63 days after sowing, respectively (**Figure 1A**).

# Effects of different light quality and GA<sub>3</sub> treatments on leaf SPAD value

Chlorophyll is the main photosynthetic pigment of higher plants, and its content is important for plant photosynthesis. The SPAD value can objectively reflect the content of chlorophyll. The SPAD values of each treatment increased compared with CK, and the increase of SPAD value of T7 treatment was the most significant, and compared with CK, it continued to rise by 45.13%, 41.23%, 45.45%, and 47.96%, respectively, at 42, 49, 56 and 63 days after sowing (Figure 1B).

# Effects of different light quality and GA3 treatments on enzyme activities of carbon and nitrogen metabolism

Amylase can catalyze the hydrolysis of glycosidic bonds in starch to form reducing sugars such as maltose and glucose. **Figure 1C** clearly indicates that the amylase activity of each treatment increased first and then decreased. The T1 treatment caused a maximal activity of amylase at 49 days after sowing, and the rest of the treatments showed highest amylase activity at 56 days after sowing, so the treatments in terms of amylase activity can be categorized as T4 > T5 > T7 > T8 > T3 > T2 > T6 > CK. Compared with CK, the activity of amylase in T4, T5 and T7 treatments was significantly increased by 99.29%, 45.41% and 62.59%, respectively, at 42 days after sowing, and increased by 77.26%, 62.33% and 52.33%, respectively, after 49 days of sowing. After 56 days of sowing an increase of 68.68% (T4), 57.33% (T5), and 45.74% (T7), and at 63 days after sowing, an increase by 73.54%, 61.39% and 48.99%, in T4, T5, and T7, respectively, was observed (**Figure 1C**).

Nitrate reductase (NR) acts as a key enzyme in nitrogen metabolism and its activity affects the rate of nitrogen assimilation and protein synthesis. As can be seen in **Figure 1D**, compared with CK the NR activity of T2, T4, T5, T7 and T8 seedlings was significantly increased, but the difference among T1, T3 and T6 was not significant. The activity trend showed an increase in NR from 42 to 63 days after sowing. Compared with CK, the activity of NR in T2, T4, and T8 increased steadily throughout the experiment by 74.80%, 58.98%, 86.12% and 66.14%, at 42, 49, 56, and 63 days after sowing, respectively (**Figure 1D**).

# Effects of different light quality and GA3 treatments on the content of carbon and nitrogen metabolites

**Figure 1E** shows that the starch content of each treatment had a tendency to rise first and then to decrease. The increase of starch content in T4 treatment was the most significant, which increased by 45.03%, 30.02%, 29.30%, and 45.45%, respectively, followed by that in the T5 treatment, which increased by 42.54%, 23.20%, 27.41%, and 45.67%, at 42, 49, 56 and 63 days after sowing, respectively. The treatment T7 increased by 28.45%, 17.13 %, 25.33% and 29.00%, at 42 days, 49 days, 56 days and 63 days after sowing, respectively, (Figure 1E).

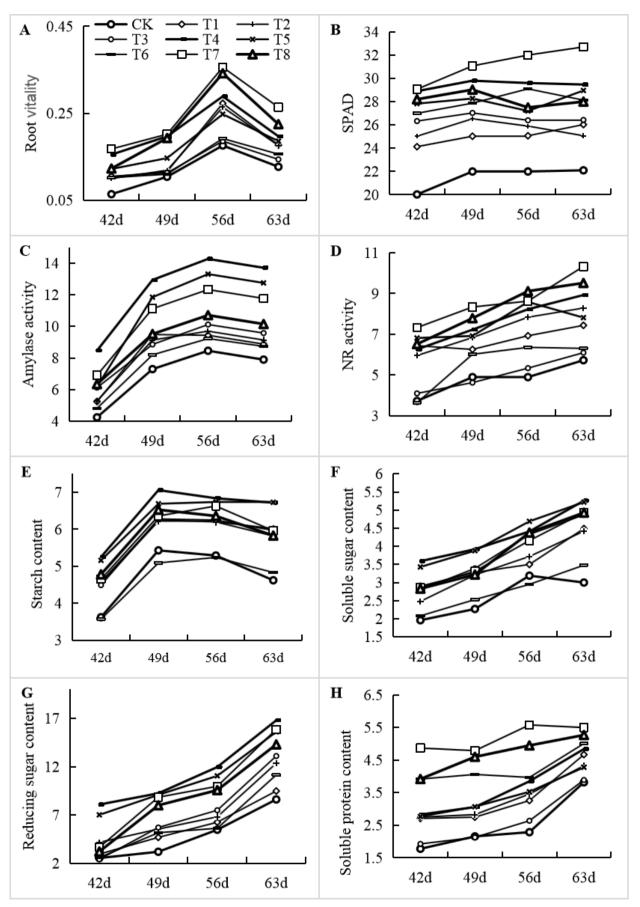


Figure 1: Effects of different light quality and GA<sub>3</sub> treatments on root vitality (A), SPAD values (B), key enzyme activities of carbon and nitrogen metabolism (C, D), and the contents of carbon and nitrogen metabolites (E-H) of tobacco seedlings. (A)  $mg \cdot g^{-1} \cdot h^{-1} \cdot FW$ , (C) amylase activity, mg maltose  $\cdot g^{-1}FW \cdot min^{-1}$ , (D) nitrate reductase activity,  $\mu g \text{ NO}^{2-} \cdot g^{-1} FW \cdot h^{-1}$ , (E) starch content (%), (F) soluble sugar content (%), (G) reducing sugar content (%), (H) soluble protein content ( $mg \cdot g^{-1}$ )

Moreover, soluble sugar content and reducing sugar content of each treatment showed an upward trend. Compared with CK, the soluble sugar content and reducing sugar content of T4 and T5 treatments were significantly high. The soluble sugar content of T4 treatment increased by 83.16%, 72.69%, 38.24% and 75.33%, respectively, and the reducing sugar content increased by 217.25%, 189.06% and 118.43%, 95.01%, at the four incremental time periods, respectively. The treatment T5 caused an increase in soluble sugar content by 75.00%, 70.93%, 47.02%, and 74.00%, and increase in reducing sugar content by 175.29%, 184.06%, 101.82%, 81.09%, at 42, 49, 56 and 63 days after sowing, respectively; and likewise the treatment T7 resulted in an increase in soluble sugar content by 45.92% 45.81%, 30.41%, and 64.33%, and in reducing sugar content by 43.53%, 176.56%, 82.12%, and 84.11%, at 42, 49, 56 and 63 days after sowing, respectively (Figure 1E and F).

As can be seen from **Figure 1G**, compared with CK, the soluble protein content in T7 increased by 173.60%, 121.76%, 143.67%, and 43.98%, followed by an increase in T8 by 120.22%, 112.96%, 116.16%, and 37.96%, at 42, 49, 56 and 63 days after sowing, respectively (**Figure 1G**).

# **Correlation analysis**

**Table 3** shows that the soluble sugar content and reducing sugar content were significantly and positively correlated with starch content, amylase activity and SPAD at 42, 49, 56, and 63 days after sowing. There was a significant positive correlation between starch content and amylase activity and SPAD values. There was a significant positive correlation between soluble protein content and NR activity and SPAD values. NR activity and amylase activity were significantly and positively correlated with SPAD values.

Measuring time	Item	Soluble sugars	Reducing sugars	Starch	Soluble proteins	Nitrate reductase activity	Amylase activity
	Reducing sugars	0.774**	-	-	-	-	-
sowing	Starch	0.852**	0.714**	-	-	-	-
	r Soluble proteins	0.064	-0.013	0.068	-	-	-
	NR activity	0.674**	0.475*	0.665**	0.433*	-	-
	Amylase activity	0.770**	0.685**	0.762**	0.227	0.475*	-
	SPAD values	0.560**	0.438*	0.575**	0.551**	0.421*	0.628**
	Reducing sugars	0.722**	-	-	-	-	-
	Starch	0.843**	0.710**	-	-	-	-
49 d after sowing	r Soluble proteins	0.027	0.489**	0.046	-	-	-
	NR activity	0.369	0.670**	0.423*	0.697**	-	-
	Amylase activity	0.861**	0.842**	0.805**	0.251	0.553**	-
	SPAD values	0.533**	0.721**	0.443*	0.618**	0.580**	0.631**
	Reducing sugars	0.749**	-	-	-	-	-
56 d after sowing	Starch	0.848**	0.753**	-	-	-	-
	r Soluble proteins	0.302	0.466*	0.403*	-	-	-
	NR activity	0.514**	0.700**	0.632**	0.715**	-	-
	Amylase activity	0.735**	0.898**	0.821**	0.432*	0.609**	-
	SPAD values	0.344	0.538**	0.425*	0.769**	0.543**	0.605**
63 d after sowing	Reducing sugars	0.777**	-	-	-	-	-
	Starch	0.863**	0.724**	-	-	-	-
	r Soluble proteins	0.283	0.412*	0.10	-	-	-
	NR activity	0.579**	0.609**	0.452*	0.664**	-	-
	Amylase activity	0.776**	0.879**	0.788**	0.336	0.543**	-
	SPAD values	0.616**	0.734**	0.465*	0.729**	0.648**	0.713**

#### **Table 3. Correlation analysis**

\*, \*\* significant at 0.05 and 0.01 levels of probability, respectively

# Discussion

# Effects of different light quality and GA<sub>3</sub> treatments on agronomic traits of tobacco seedlings

The effects of light quality on plants are mainly reflected in the photosynthesis characteristics, carbon metabolism, stomatal conductance, transpiration rate and plant cell antioxidant system. As a light-loving plant, tobacco has a significant effect of light on its growth and development. It has been

shown that the effect of red and blue lights on tobacco plants is higher (Kasperbauer, 1971; Naznin et al., 2019; Li et al., 2020). Researchers believe that in the growth period of tobacco seedlings, supplementing the red LED light source can increase the stem height of the tobacco seedlings in the greenhouse, and supplementing the blue LED light source can reduce the stem height of the tobacco seedlings and increase the stem diameter of tobacco seedlings (Li et al., 2011). Tobacco plants treated with red light can increase the internode spacing and change the leaf color and leaf thickness. The treatment with blue light can promote the growth of callus and stimulate the occurrence of buds (Yang et al., 2015). Gibberellins can promote growth, germination and flowering of tobacco and the spraying at the seedling stage is conducive to achieve enhanced growth of stems and leaves. Zhang et al. (1988) have shown that hormones determine plant organogenesis, and light induction regulates plant physiological and biochemical reactions by regulating plant endogenous hormones (Zhang et al., 1988). The results of the present study showed that, supplementation of red, blue and GA<sub>3</sub> 63 days after sowing had no significant effect on the stem diameter and leaf number of tobacco seedlings. The plant height at 50 mg  $L^{-1}$  GA<sub>3</sub> (T1, T4, and T7) increased, and at T1 (white light + spray 50 mg L<sup>-1</sup> GA<sub>3</sub>) it increased by 28.18%, at T4 (red light + 50 mg  $L^{-1}$  GA<sub>3</sub>) increased by 29.48%, at T7 (blue light + 50 mg  $L^{-1}$  GA<sub>3</sub>) it slightly increased being only 24.68%; but at a foliar spray of 100 mg L<sup>-1</sup> GA<sub>3</sub> (T2, T5, and T8) the plant height increased significantly.

This indicates that gibberellin is effective in promoting the vertical growth of tobacco seedlings, which is consistent with the results of previous studies that gibberellin can increase the auxin content in plants, regulate cell elongation and promote the physiological role in cell expansion (Ren et al., 2022). The GA-induced increase in plant height as observed in the present study is consistent with the results of some earlier studies, e.g., in rice (Li et al., 1994), cabbage seedlings (Cao, 2013), and *Phalaenopsis* tissue culture seedlings (Dai et al., 2010).

# Effects of different light quality and GA<sub>3</sub> treatments on root growth and vigor of tobacco seedlings

Roots are the main organs for plants to absorb and transport nutrients and water from the soil. Their growth status and vitality are regulated by light and hormone levels, in addition to soil fertility, water, permeability and microbes. The present study found that the effects of different light qualities on the root system were significant, and the combined treatment of red and blue light could effectively increase the root growth and root activity of plants as earlier observed in cucumber seedlings (Yan et al., 2016). It has been reported that the combination of red and blue light improves root growth and root activity (Guo et al., 2011). Moreover, it has been shown that the root activity, root absorption area and root crown growth rate of blue-red combined light source or blue light treated tobacco seedlings are significantly higher than that under red light treatment (Xu et al., 2015). Blue light can significantly increase the root length, surface area and volume of tobacco seedlings. Exogenous GA<sub>3</sub> promoted the plant height, fresh weight, root length, lateral root length and number of lateral roots in the tobacco seedlings. Overall performance of the tobacco seedlings was recorded to be the best under blue light + spray of 50 mg·L<sup>-1</sup> GA<sub>3</sub>. Therefore, supplementation of blue and red light as well as spray of GA<sub>3</sub> can enhance the root activity of tobacco seedlings, thereby increasing the absorption rate of nutrients and finally promoting the growth of tobacco seedlings. Earlier studies with maize have shown that GA<sub>3</sub> solution can significantly promote the development of the main roots, lateral roots and stems and leaves of seedlings (Suo et al., 2013; Xu et al., 2018).

# Effects of different light quality and GA<sub>3</sub> treatments on SPAD values of tobacco seedling leaves

Plant photosynthetic pigments mainly include chlorophyll and carotenoids. Among them, chlorophyll is the main pigment molecule for photosynthesis that is responsible for the absorption, transmission and transformation of light energy in photosynthesis. The increase of chlorophyll content can promote the absorption and transformation of plants by light energy, and speed up the photosynthetic rate (Yan et al., 2014; Ahmed et al., 2022). Su et al. (2007) have found that the correlation between SPAD values and chlorophyll content in the leaves of potato buds has reached a very significant level. Li et al. (2010) showed that the SPAD values and chlorophyll content of apple leaves were linearly correlated. The blue LED light treatment can increase the CO<sub>2</sub> concentration of the substrate cells, and the red LED light treatment improves the conversion efficiency of the original light radiation intensity of the greenhouse tobacco seedling leaves, thereby increasing the photosynthetic rate of the tobacco seedlings (Xue et al., 2014). Previously, Ni et al. (2009) have shown that the photosynthetic rate of cucumber leaves under red LED fill light is higher than that under blue LED fill light treatment, and Fu et al. (2007) have shown that rice photosynthetic rate under blue LED fill light was higher than under red LED fill light treatment, indicating different crop photosynthetic systems exhibiting different responses to light quality. Exogenous GA<sub>3</sub> stimulates endogenous GA<sub>3</sub> activity, promotes the levels of endogenous IAA in leaves, enhances the function of plant chlorophyll synthesis, and increases the number of chloroplasts,

thereby accelerating photosynthetic rate (He et al., 1979; Wen et al., 2018). Chlorophyll content and photosynthetic rate of winter jujube increased after treatment with 20 mg  $L^{-1}$  and 40 mg  $L^{-1}$  GA<sub>3</sub> (Cao, 2012). Huang (2006) has shown that the contents of chlorophyll a and chlorophyll b increased significantly in lily plants after treatment with 200 mg  $L^{-1}$  or 500 mg  $L^{-1}$  GA<sub>3</sub> resulting in improved photosynthesis. The results of the present study showed that the application of different light qualities and the application of GA<sub>3</sub> had a significant effect on the SPAD values of Yunvan 99 tobacco seedlings. Under the conditions of blue light or red light with appropriate spray of GA<sub>3</sub> at the same time could increase the SPAD values of leaves. This indicates that an increase in chlorophyll content has a significant influence on photosynthetic rate. The amount of leaf pigment during the growth process of tobacco leaves not only affects the growth and development of tobacco leaves, but also affects the aroma components and intrinsic quality of tobacco leaves, thereby determining the flavor of tobacco leaves. At 42, 49, 56 and 63 days after sowing, the SPAD values of the leaves of the tobacco seedlings varied significantly between the treatments. The results of the four periods were compared with the control (white light + foliar-applied 0 mg  $L^{-1}$  GA<sub>3</sub>). Compared with the other 8 treatments, the SPAD values increased significantly, and the best results were obtained by blue light + spray of 50 mg  $L^{-1}$  GA<sub>3</sub> treatment, followed by blue light + 100 mg  $L^{-1}$  GA<sub>3</sub>.

# Effects of different light quality and GA3 treatments on carbon and nitrogen metabolism enzyme activities and products of tobacco seedlings

Carbon and nitrogen metabolism is the most basic physiological metabolic process of plants. The metabolism of plants during each growth period directly relates to absorption and assimilation of minerals, synthesis of nutrients and formation and transformation of photosynthetic products (Tang et al., 2011; Lambers and Bassham, 2022). Amylase and NR are the rate-limiting enzymes of carbon and nitrogen metabolism. Their activity is important for the formation of carbon compounds such as starch, reducing sugars and soluble sugars in the tobacco leaves, as well as the formation of nitrogen-containing compounds such as proteins, amino acids and nicotine. The role of light quality has a regulatory effect on plant carbohydrate and nitrogen metabolism (Ning et al., 2015). Guo et al. (2011) showed that blue light significantly inhibited the growth of rice seedlings, but increased the seedling index and soluble protein content in seedlings. Red light significantly increased the starch and soluble sugar content of the rice seedlings. Ning Yu and others (Ning et al., 2015) have shown that increasing proportion of red light can promote nitrogen absorption and carbon metabolism cycle, inhibit protein synthesis, increase the proportion of blue light which can enhance the regulation of nitrogen metabolism, but the net photosynthetic rate is lowered down, and the accumulation and metabolism of carbon in plants are inhibited. Xiao et al. (2013) have reported that the combined light source based on red light can significantly increase the soluble total sugar content of tobacco seedlings; in the combined light source based on blue light, it can significantly enhance the NR activity of the tobacco seedlings and enhance the soluble protein content. During the vegetative growth of lily, the soluble sugar content of leaves was less variable with GA<sub>3</sub> treatment, while soluble sugar content maintained a high level for a long period of time with a pronounced effect of high concentration of  $GA_3$  treatment (Huang, 2006). The present study showed that Yunyan 99 seedlings had significant effects on their key enzyme activities of carbon and nitrogen metabolism pathways and the synthesis and transformation of their metabolites after treatment with different light qualities and  $GA_3$ . The carbon metabolism amylase activity and its products showed that the supplemental red light and sprayed  $GA_3$  treatment were better than the supplemental blue light and sprayed  $GA_3$  treatment, and both were better than the supplemental white light and sprayed  $GA_3$ treatment. It indicates that the supplementation of red light at the tobacco seedling stage and spraying of suitable concentration of GA<sub>3</sub> can effectively enhance the key enzyme activities of carbon and nitrogen metabolism, increase the rate of carbon and nitrogen metabolism, increase the content of starch, soluble sugars and reducing sugars, thereby promoting the growth of tobacco seedlings. The nitrogen assimilation key enzyme NR activity and nitrogen assimilation substrate soluble protein content were both enhanced by blue light and foliar-applied GA<sub>3</sub>

# Conclusion

This research indicated that light quality and gibberellins improve the quality of tobacco plants under different light sources. Based on our results, it can be concluded that light quality and gibberellins significantly enhanced the fundamental morphological traits, e.g., leaf number, plant height and shoot biomass were significantly enhanced compared to that by the control treatments. Our results could provide a better understanding of the combined effects of light quality and gibberellins under different light sources on growth, root characteristics, and carbon and nitrogen metabolism of tobacco seedlings.

# Author(s), Editor(s) and Publisher's declarations

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#### **Supplementary material**

No supplementary material is included with this manuscript.

#### **Conflict of interest**

The authors declare no conflict of interest.

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### **Contribution of authors**

Conceptualization and designing the study: YL, ZC. Conduction of experiment: NB. Data collection, visualization and interpretation: SA. Preparation of initial draft: MAA. Review of initial draft: LZ. Proof reading and approval of the final version: PW.

# **Ethical approval**

This study does not involve Human/animal subjects and no ethical approval is needed.

## Handling of bio-hazardous materials

The author(s) certify that all experimental materials were handled with care during collection and experimental procedures. After completion of experiment, all materials were properly discorded to minimize any types of bio-contamination(s).

#### Availability of primary data and martials

As per editorial policy, experimental materials, primary data or software codes are not submitted to the publisher. These are available with corresponding author and/or with other author(s) as declared by the corresponding author of this manuscript.

### Author's consent

All authors contributed in designing and execution of the experiment. All contributors have critically read this manuscript and agree for publishing in IJAEB.

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# References

Ahmed, N., Hussain, H.Z., Ali, M.A., Rahi, A.A., Saleem, M., Ahmad, F. (2022). Effect of zinc on chlorophyll contents, gas exchange attributes and zinc concentration in rice. *Pakistan Journal of Botany* 54(1):17-24.

- Cao, G., (2013). Effects of different LED light quality on seedling growth, light and characteristics and endogenous hormones of cucumber and cabbage. *Gansu Agricultural University, Lanzhou*, China.
- Cao, L.Q., (2012). Effects of different concentrations of gibberellin on chlorophyll content of winter jujube leaves. *Modern Horticulture* (22):10.
- Chen, L., Wang, Y. F., Zhang, X.J., Wang, M.L. (2015). Effects of different red and blue combined light sources on root growth and root vigor of peanut seedlings. *Abstract of papers of the 2015 Annual Conference*. Beijing: China Crop Society 208.

- Dai, Y.J., Wang, Q.L., Zhang et al. (2010). Effects of LEDs with different spectra on the growth of *Phalaenopsis* seedlings. *Jiangsu Agricultural Sciences* 5:227-231.
- Franklin, K.A. (2009). Light and temperature signal crosstalk in plant development. *Current Opinion in Plant Biology* 12(1):63-68.
- Fu, C.M., Huang, N.Z., Zhao, Z.G., Tang, F.Q., Huang, Z.M. (2007). Effects of light quality and light supplementation on growth and photosynthetic rate of rice seedlings. *Guangxi Plant* 27(2):255-259.
- Gao, J.F. (2006). "Experimental Guidance of Plant Physiology". Beijing Higher Education Press.
- Gu, Z.X., Chen, Z.G., Jiang, Z.H. (2003). Effects of gibberellin treatment on germination and changes of main components of brown rice. *Journal of Nanjing Agricultural University* 26(1):74-77.
- Guo, Y.S., Gu, A.S., Cui, W. (2011). Effects of light quality on growth and physiological characteristics of rice seedlings. *Chinese Journal of Applied Ecology*. 22(6):1485-1492.
- Han, Y., Zhu, X.D., Yan, X.D., Wang, Z. (2005). Effects of light on root growth of hydroponic hyacinth. *Journal of Yangzhou University* 32(2):79-82.
- He, Z.P., Luo, S.W. (1979). The relationship between gibberellin promoting tobacco tissue growth and nitrogen metabolism. *Chinese Journal of Plant Physiology* 5(4):319-326.
- Huang, J., Zhang, Y.Z., Li, H.G., Fang, M., Wang, S.C, Kong, F.Y. (2014). Effect of warming and supplementation on the seedling quality of flue-cured tobacco in Wuzhou. *China Tobacco Science* 2:100-103.
- Huang, Y.H. (2006). Effect of gibberellin on photosynthetic characteristics and growth status of lily tissue culture seedlings. *Guangxi University*, Nanning, China.
- Kang, P., Wang, S.S., Ma, W.G., Long, Z.L., Yang, H.Q., Song, B.Q, Gong, M. (2014). Gibberellin induces simultaneous improvement of drought resistance and cold resistance of tobacco seeds and seedlings. *Seed* 33(2):30-34.
- Kasperbauer, M.J. (1971). Spectral distribution of light in a tobacco canopy and effects of end-of-day light quality on growth and development. *Plant Physiology* 47(6):775-778.
- Ke, X., Li. J.Y., Li, X.Y., Qi, C.F., Xu, C.H., Jin, Y., Gong, M. (2011). Effects of different light qualities on growth, light and characteristics of tobacco leaves. *Acta Physiologica Sinica* 47(5):512-520.
- Lambers, H., Bassham, J.A. (2022). "Photosynthesis", *Encyclopedia Britannica* https://www.britannica.com/science/photosynthesis
- Lencucha, R., Drope, J., Magati, P., Sahadewo, G.A. (2022). Tobacco farming: overcoming an understated impediment to comprehensive tobacco control. *Tobacco Control* 31:308–312.
- Li, H.S. (2000). "Principles and Techniques of Plant Physiological and Biochemical Experiments". *Higher Education Press*, Beijing 153-157.
- Li, J., Liu, F.J., Xu, J. (2011). Effects of different light-emitting diode (LED) light sources on the growth, fruit yield and quality of autumn tomato plants in greenhouse. *Jiangsu Journal of Agricultural Sciences* 27(6):339~343.
- Li, M.X., Zhang, L.S., Li, B.Z., Zhang, H.Y., Guo, W. (2010). Relationship between hyperspectral properties and chlorophyll content and SPAD value of apple leaves. *Journal of Northwest Forestry University* 25(2):35-39.
- Li, X.B., Xu, J.C., Zhu, L.H. (2004). Gibberellin signal transduction and plant-dwarfism. *Journal of Chinese Biotechnology* 24(12):26-31.
- Li, Y., Xin, G., Liu, C. *et al.* (2020). Effects of red and blue light on leaf anatomy, CO<sub>2</sub> assimilation and the photosynthetic electron transport capacity of sweet pepper (*Capsicum annuum* L.) seedlings. *BMC Plant Biology* 20:318. https://doi.org/10.1186/s12870-020-02523-z
- Li, Y.S., Pan, R.C. (1994). Study on the effect of blue light on the growth of rice seedlings. *China Rice Science* 8(2):115-118.
- Long, Z.K., Long, J.Z., Wu, M.L. (2010). Study on the effect of gibberellin on the growth of *Eustoma* seedlings. *Chinese Horticultural Digest* 26(6):17-19.
- Mao, J.S., Yang, D.Q. (1991). Effects of light quality on rice seedling growth. *Journal of Southwest Agricultural University* 13(4):446-449.
- Martins, G.Z., Silva, S.R., Kölln, O.T. (2022). Does a hormonal plant growth promoter (KIN, GA<sub>3</sub>, and IBA) affect grain yield and N, P, K, Ca, and Mg uptake in wheat crop in Southern Brazil? *Journal of Plant Nutrition* DOI: 10.1080/01904167.2022.2058535.
- Naznin, M.T., Lefsrud, M., Azad, M.O.K., Park, C.H. (2019). Effect of different combinations of red and blue LED light on growth characteristics and pigment content of *in vitro* tomato plantlets. *Agriculture* 9:196. https://doi.org/10.3390/agriculture9090196.
- Nguyen, C.T., Dang, L.H., Nguyen, D.T., Tran, K.P., Giang, B.L., Tran, N.Q. (2019). Effect of GA<sub>3</sub> and Gly plant growth regulators on productivity and sugar content of sugarcane. *Agriculture* 9:136. https://doi.org/10.3390/agriculture9070136
- Ni, J.H., Chen, X.H, Chen, C.H., Xu, Q. (2009). Supplementation of different light qualities on the growth, photosynthesis and early yield of cucumber in greenhouse. *Chinese Agricultural Sciences* 42(7):2615-2623.

Ni, W. (1980). Effect of different light quality on rice seedling growth. Yunnan Plant Research. 2(2):194-201.

- Ning, Y., Deng, H.H., Li, Q.M., Mi, Q.H., Han, B., Ai, X.Z. (2015). Effects of red and blue light on carbon and nitrogen metabolism and key enzyme activities of celery. *Chinese Journal of Plant Physiology* 51(1):112-118.
- Ren, Y., Zhang, S., Xu, T., Kang, X. (2022). Morphological, transcriptome, and hormone analysis of dwarfism in tetraploids of *Populus alba × P. glandulosa*. *International Journal of Molecular Sciences* 23:9762. https:// doi.org/10.3390/ijms23179762.
- Shafiq, I., Hussain, I., Raza, M.A., Iqbal, N., Asghar, M.A., Raza, A., Fan, Y-F., Mumtaz, M., Shoaib, M., Ansar, M., Manaf, A., Yang, W-Y., Yang, F. (2021). Crop photosynthetic response to light quality and light intensity. *Journal* of Integrative Agriculture 20(1):4-23,

- Su, Y.S., Guo, H.C, Chen, Y.L. (2007). Correlation between SPAD value and chlorophyll content and yield in potato leaves. *Southwest Agricultural Journal* 20(4):690-693.
- Suo, W.L., Zheng, Y., Ma, W.G., Niu, Y.Z., Song, B.Q., Li, Y.J. (2013). Effects of different pH gibberellin priming on tobacco seed germination and seedling quality. *Chinese Tobacco Science* (6):60-63.
- Tang, X.M., Zhong, R.C., Jie, H.K., Liu, C., Wang, Z.P., Han, Z.Q. (2011). Effect of intercropping peanut on carbon and nitrogen metabolites and key enzyme activities of cassava. *Chinese Agricultural Science Bulletin* 27(3):94-98.
- Wang, G.P., Hu, G.P., Han, T.S., Liu, Z.H., Hu, G. (2013). Effect of gibberellin treatment on seed germination and seedling growth of wild jasmine. *Forestry Science and Technology* 38(5):10-13.
- Wang, R., Liu, G.S., Chen, G.H., Xiang, D., Wu, Y.P. (2010). Effects of light intensity on photosynthesis and dry matter production of flue-cured tobacco in seedling stage. *Chinese Journal of Applied Ecology* 21(8):2072-2077.
- Wen, Y., Su, S-C., Ma, L-Y., Wang, X-N. (2018). Effects of gibberellic acid on photosynthesis and endogenous hormones of *Camellia oleifera* Abel. in 1st and 6th leaves, *Journal of Forest Research* 23:309-317.
- Xiao, C.S., Chen, W., Zhong, Y.F., Yang, H.Q., He, M.J., Li, F., Yan, X.D. (2013). Effects of red and blue light on the growth and carbon and nitrogen metabolism of tobacco seedlings. *Chinese Agricultural Science Bulletin* 29(22):160-166.
- Xu, G.P., Liu, X.F., Yuan, W.Y., Wang, W., Lou, C.J., Yang, Z.S. (2018). Effects of gibberellin soaking on the low-temperature emergence rate of maize. *Journal of North China Agricultural University* 33(4):147-152.
- Xu, J.G., Ren, S.H., Zhang, L., Zhang, W., Zu, C.L., Cao, Y., Liu, B.R., Wang, W.J. (2015). The effect of supplemental light source on root growth of tobacco seedlings under the model of sports seedlings—2015; Excellent papers collection. Beijing, *China Tobacco Learn* 1-8.
- Xue, C.Q., Wang, J.W., Yang, L.J. (2014). Effects of different LED fill light on light environment and growth of tobacco seedlings in solar greenhouse. *Chinese Journal of Tobacco* 20(2):54-58.
- Yan, M.M., Wang, M.L., Wang, H.B., Wang, Y.F., Zhao, C.X. (2014). Effects of light quality on photosynthetic pigment content and photosynthetic characteristics of peanut seedling leaves. *The Journal of Applied Ecology* 25(2):483-487.
- Yan, X.H., Yu, J.H., Yan, J.M. (2016). Effects of light-filling time and light quality on growth and root vigor of cucumber seedlings. *Journal of Nuclear Agricultural Sciences* 30(6):1211-1217.
- Yang, L.Y., Li, J.Y., Wang, L.P., Duan, S.Z., Ma, J.H., Ma, E.D., Gong, M. (2015). Research progress on the effects of light environment on tobacco growth and material metabolism. *Genomics and Applied Biology* 34(5):1114-1128.
- Zhang, F.P., Zeng, Y.Q. (2009). Effects of plant growth regulators on seed germination and seedling growth of amaranth. *Guizhou Agricultural Sciences* 37(11):160-163.
- Zhang, Y.F., Dong, C.X., Ni, D.X., Wang, Q. (1988). Effects of different hormone concentrations and light quality on adventitious bud formation in tobacco leaves. *Journal of Experimental Biology* 21(4):541-544.
- Zhou, M.B., Tang D.Q. (2004). Research progress of gibberellin biosynthesis and its key enzymes in higher plants. Journal of Zhejiang Forestry College 21(3):344-348.

Zou, Q. (1995). "Plant Physiology and Biochemistry Experiment Guide", China Agriculture Press, Beijing, 26-108.

Zou, W., Shi, J.X., Jiang, W., Meng, L., Feng, X.Y., Liu, M.H., Rao, X.Y., Chen, W. (2014). LED filling effect of stereo floating seedlings of flue-cured tobacco. *Chinese Tobacco Science* 35(6):17-20.