

## MISCELLANEOUS

# Growth, Photosynthetic Characteristics, Antioxidant Capacity and Biomass Yield and Quality of Wheat (*Triticum aestivum* L.) Exposed to LED Light Sources with Different Spectra Combinations

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## Keywords

antioxidant capacity; biomass; bioregenerative life support system; light quality; photosynthetic characteristics; wheat

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## Abstract

As a consequence of the increasing importance of crop in Bioregenerative Life Support System (BLSS), there is an interest in enhancing both the productivity and quality of wheat. Lighting systems for growing wheat need to be lightweight, reliable and durable. Light-emitting diodes (LEDs) have these characteristics. Previous studies demonstrated that the combination of red and blue lights was an effective light source for several crops. Yet the appearance of plant in this kind of lighting was purplish grey, and other problems were also accompanied. The addition of other spectra LEDs made better growth and also offer a better visual experience to bring psychological benefit to the crews. The objective of this study was to investigate the influences of different spectra combinations on the wheat growth, photosynthetic characteristics, antioxidant capacity and biomass yield and quality during their life cycle. Four types of different spectra combinations with the same intensity were employed: a single red light (R), a red–blue light (R + B, R : B = 4 : 1), a red–white light (R + W, R : W = 4 : 1) and a white light (W). The results showed that the wheat cultivated in the R + W light was characterized by highest harvest index and lowest lignin in inedible biomass, which was more beneficial to recycle substances in the processes of the environment regeneration. The data were comparable to those under W condition in terms of straw height, relative water content (RWC), membrane stability index (MSI), photosynthetic rate, chlorophyll concentration, antioxidant capacity, thousand kernel weight (TKW) and soluble sugar concentration. Wheat was sensitive to light quality which significantly affected those indices of growth and physiology, especially at earing and flowering stages.

## Introduction

Plants in Bioregenerative Life Support System (BLSS) can provide human beings with fresh air, clean drinking water, nutrient-rich food and necessary spiritual consolation, which are essential for long-term manned space missions (Blüm et al. 1994, Sirko et al. 1994, Lasseur et al. 1996, Tong et al. 2011). However, how to select an ample and suitable light source for plant growth and to supplement artificial light source for less-sunlight areas to improve

plant yield and quality are actual challenges. Generally, high-pressure sodium lamps (HPSL), metal halide lamps (MHL), incandescent lamps and fluorescent lamps are widely used for plants to fulfil a complete life cycle. Nevertheless, these lamps have some limitations for application in BLSS because they are bulky and the energy conversion capability is low. Therefore, seeking a new kind of light source for plant cultivation in BLSS is needed. Light-emitting diode (LED), a solid-state semiconductor, is able to convert electrical energy into visible

light. It is characterized in long lifespan, high photosynthetic efficiency, small size, less thermal radiation as well as high safety performance (Bula *et al.* 1991, Schuerger *et al.* 1997). Customized emission wavelength is also available for LEDs, which can match the most of plant photosynthesis receptors. As a result, crop yields, physiological characteristics and metabolic components are under control (Bourget 2008, Massa *et al.* 2008, Morrow 2008). Thus, LED light is considered as an ideal light source for plant cultivation of BLSS (Barta *et al.* 1992, Bula and Zhou 2000).

The red–blue LED combination as light source was shown to replace in part sunlight for crop growth. Some spectra such as green light (500–600 nm), far-red light (710–740 nm), UV-A (320–500 nm) and UV-B (280–320 nm) have positive impacts on development and growth of plants by triggering physiological and biochemical reaction (Briggs and Olney 2001, Briggs *et al.* 2001, Kim *et al.* 2004). The phytochrome and cryptochrome of plants, which are involved in the control of chloroplast growth (Batschauer 1998), can be affected by far-red light and ultraviolet light, and UV-A, respectively. Besides, the maximum absorption wavelength of proteins is within the range of UV-B radiation wavelength and thus UV-B has a significant impact on protein metabolism (Tevini *et al.* 1981, Mackeress *et al.* 1999). Several studies indicated that adding green light to the red–blue light can improve the growth and quality of lettuce (Kim *et al.* 2004); however, it leads to dim appearance, short of nutrition and accelerated senescence to crops when other spectra with different wavelengths are lack (Bula and Zhou 2000). Therefore, further study on optimizing LED spectra, which is one of the most important factors affecting the growth of wheat plants in controlled environments including BLSS, is required.

In principle, the white LED exhibits obvious advantages on the optimization of plant light source and spectra. On one hand, the white LED is a continuous spectrum which can provide different wave bands of spectra for plant growth and development like far-infrared ray, ultraviolet light and green light. On the other hand, the white LED consists of blue LED wrapped with phosphor powder, which enhances the luminous efficiency of light source and plant utilization rate (Pimputkar *et al.* 2009). Previous studies demonstrate that the red–blue–white LED light source indeed is predominant in improving the output and nutritional quality of crops such as lettuce (Lin *et al.* 2013) and tomatoes (Lu *et al.* 2012) in comparison with the red–blue LED light source.

Wheat is one of the core grain crops in BLSS. Previous study has shown that although wheat is not sensitive to the blue light dose induction (Dougher and Bugbee 2001, Cope and Bugbee 2013), the combination of red–blue LED spectra can improve its output and photosynthetic

rate (Goins *et al.* 1997). However, it is still unclear whether the red–white LED can further improve the output and photosynthetic rate of wheat plants in a controlled environment. In addition, in BLSS, the quality and inedible biomass of wheat are also becoming an important focus of both theory and practical applications. There was still no report on the change of criteria and parameters of crops when different LED spectra light sources were used. The aim of this study was to investigate the influence of different LED light sources on photosynthetic characteristics, enzymatic antioxidant capacity and biomass yield and quality of wheat (*Triticum aestivum* L.) in a controlled environment.

Here, we have designed a single red light (R), a red–blue light (R + B, R : B = 4 : 1), a red–white light (R + W, R : W = 4 : 1) and a white light (W), which are with the same initial photosynthetic photon flux density (PPFD). In such conditions, the physiological and biochemical indexes including photosynthetic characteristics, relative water content, the stability of cell membrane, antiretroviral system, the output, quality and the component of inedible biomass of the edible biomass were studied during the whole life cycle of wheats. The results provide some basic information on the optimal light sources for stable, good quality, high yields of wheat plants in BLSS.

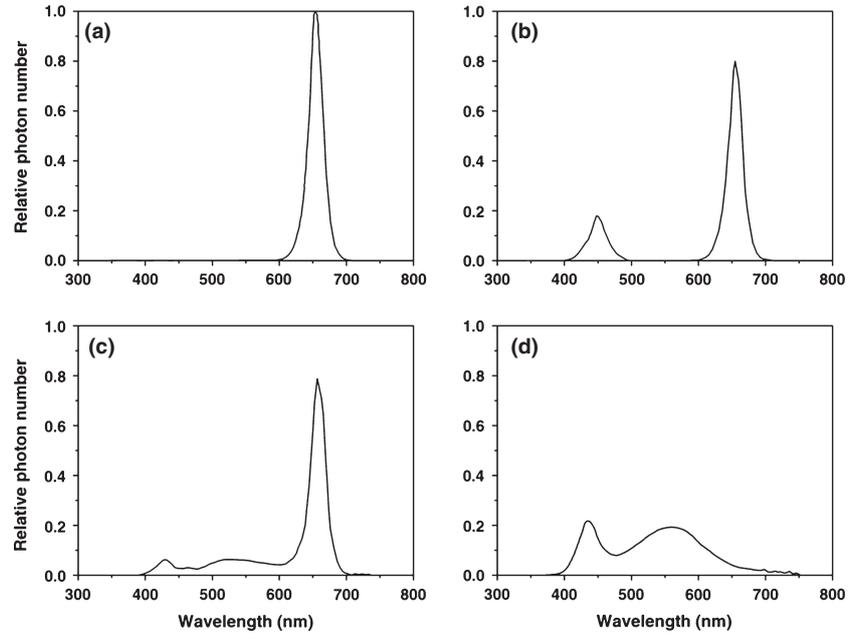
## Materials and Methods

### Light treatments

Red LEDs only (R), mixtures of red plus blue LEDs (R + B, R : B = 4 : 1), mixtures of red plus white LEDs (R + W, R : W = 4 : 1) and white LEDs (W) were used. For all treatments, lighting was continuous (24/0 h light/dark). Photosynthetic photon flux density (PPFD) levels were measured daily at the top of plant canopy with a quantum sensor (Li-250A; Li-Cor, Lincoln, NE, USA). Light sources were moved up and down every day to keep the same light intensity throughout the growth cycle of wheat. PPFD was about  $500 \mu\text{mol m}^{-2} \text{s}^{-1}$  for all the treatments, as calculated (Avaspec-2048-UA; Avantes B.V., Apeldoorn, Netherlands) from Fig. 1 and the spectral absorbance from 300 to 800 nm. All of the treatments were placed in a culture room and were arranged in as separate plots with the same light intensity. Ten samples of those wheat plants were selected randomly when the measurement was in process.

### Cultivation conditions

Spring wheats (*Triticum aestivum* L.) were cultivated on negative pressure porous titanium tubes, which were implemented water supply on demand. The distance



**Fig 1** Spectral distribution of light treatments, including red LEDs only (R, 1a), mixtures of red plus blue LEDs (R : B = 4 : 1, 1b), mixtures of red plus white LEDs (R : W = 4 : 1, 1c) and white LEDs (W, 1d). Photosynthetic photon flux integrations for each light treatment were equal to  $\sim 500 \mu\text{mol m}^{-2} \text{s}^{-1}$

between LED light and plant canopy was 70 cm, which guaranteed that the PPFD of wheat initial canopy was unified. Air temperature, relative humidity and  $\text{CO}_2$  levels were maintained in growth chambers at  $21 \pm 1.3 \text{ }^\circ\text{C}$ ,  $70 \pm 4.6 \%$  and  $500 \pm 48.2 \mu\text{mol mol}^{-1}$ , respectively. The growth period was about 70 days.

The modified Hoagland nutrient solution (Hoagland and Arnon 1950) included:  $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ , 1417 mg/l;  $\text{KNO}_3$ , 910 mg/l;  $\text{NH}_4\text{H}_2\text{PO}_4$ , 172 mg/l;  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , 739 mg/l;  $\text{FeEDTA}$ , 45 mg/l;  $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ , 2.4 mg/l;  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ , 0.12 mg/l;  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ , 0.33 mg/l;  $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$ , 0.03 mg/l;  $\text{H}_2\text{BO}_3$ , 4.29 mg/l, and the pH was 6.3–6.5.

## Morphological and physiological analyses

### Morphology

The height and root length of wheat plants were measured every 2 days by straight scale and vernier caliper. The samples were selected on random within measurement process.

### Determination of relative water content

At the beginning of earing and flowering stages, the RWCs of leaves were separately measured (Weatherley 1951). The fresh leaves were weighted about 0.5 g ( $m_1$ ) and soaked in double distilled water at room temperature for 4 h. Then the leaves were weighted as  $m_2$  and put in the drying oven ( $65 \text{ }^\circ\text{C}$ ) for 48 h. The dried leaves were expressed as  $m_3$ . RWC was calculated as according to the following equation:

$$\text{RWC} = \frac{m_1 - m_3}{m_2 - m_3} \times 100\%$$

### Determination of membrane stability index

To measure the MSI of leaves at the beginning of earing and flowering stages (Sairam and Srivastava 2001), the sample was divided into two equivalent parts (about 0.1 g for each) and soaked in 10 ml double distilled water. Then, one part was heated at  $40 \text{ }^\circ\text{C}$  for 30 min. Conductivity  $C_1$  was determined by conductivity metre (HI8733; Hanna Instruments, Padova, Italy). The other part was heated at  $100 \text{ }^\circ\text{C}$  for 10 min, and conductivity  $C_2$  was determined. Membrane stability index was calculated as the following:

$$\text{MSI} = \left(1 - \frac{C_1}{C_2}\right) \times 100\%$$

## Photosynthetic characteristics analyses

### Determination of chlorophyll

The content of Chlorophyll a and Chlorophyll b was detected by ultraviolet spectrophotometer (SP-75; Shanghai spectrum instruments co., Ltd, Shanghai, China) (Mackinney 1941). Samples were frozen in liquid nitrogen and stored at  $-80 \text{ }^\circ\text{C}$  until use.

### Determination of photosynthetic efficiency

Portable photosynthesis instrument (Li-6400XT) was used for the determination of photosynthetic characteristics. Leaf gas exchange parameters included photosynthetic rate

(A) and stomatal conductance (gs) of the second leaf at the wheat terminal bud. The intrinsic water-use efficiency ( $A/g_s$ ) was calculated by dividing A by  $g_s$  (Pérez-López *et al.* 2013). These parameters were analysed every 2 days.

#### *Stomata observation*

Samples were excised from the leaves of ten wheat plants at a similar position for each treatment. To observe the stomata, samples were taken from fully expanded leaves in each plant. The slides made by the leaf epidermal fingerprint of cotton with the transparent nail polish method were observed using an optical microscope (Zeng *et al.* 2008). Slides were analysed with an Olympus DP71 microscope (Olympus Inc., Tokyo, Japan). The length, width and frequency of stomata were measured with Motic Images Plus 2.0. 10 images per leaf, one leaf per plant and 10 plants per treatment were analysed.

#### **Antioxidant capacity analyses**

##### *Determination of superoxide dismutase, peroxidase and catalase activity*

Superoxide dismutase (SOD) activity was determined based on the method as previously described by Dhindsa *et al.* (1981). Briefly, 0.5 g fresh weight of wheat leaves was ice bathed in homogenate and centrifuged at 15 000 g for 20 min. 50 ml of supernatant constant volume was added to 10 ml reaction mixtures containing 5 % phenols, 5 %  $H_2O_2$  and 87.5 % distilled water. Then, 0.25 ml enzyme solution was added into the reaction mixture, and the reaction was carried out at a constant temperature (25 °C) for 20 min. The optical density was determined at  $\lambda = 470$  nm.

Peroxidase (POD) activity was analysed spectrophotometrically at 470 nm using guaiacol as a phenolic substrate with hydrogen peroxide (Diaz *et al.* 2001). The reaction mixture contained 0.15 ml of 4 % (v/v) guaiacol, 0.15 ml of 1 % (v/v)  $H_2O_2$ , 2.66 ml of 0.1 M phosphate buffer (pH = 7.0) and 40  $\mu$ l of enzyme extract. Blank sample contained the same mixture without enzyme extract.

Catalase (CAT) activity was determined according to the method described by Kumar and Knowles (1993). Catalase reaction solution consisted of 100 mM  $Na_2HPO_4$ – $NaH_2PO_4$  buffer solution (pH = 7.0) and 0.1 M  $H_2O_2$ . The optical density was determined every 1 min at  $\lambda = 240$  nm.

##### *Determination of malondialdehyde*

Determination of malondialdehyde (MDA) depended on the method of Stewart and Bewley (1980). Briefly, 10 ml 0.1 % TCA pestled homogenate was used to centrifuge wheat leaves (0.5 g) at 4000 rpm for 10 min. 2 ml supernatant was added to 4 ml 5 % TBA which was made up by

20 % TCA. The mixture was heated at 95 °C for 30 min and then cooled in ice bath rapidly. The supernatant was obtained by centrifuging at 10 000 g for 10 min. When  $\lambda = 532$  nm and  $\lambda = 600$  nm, the specificity optical density was determined. The content of MDA was calculated by absorptivity of 155  $mm^{-1} cm^{-1}$ .

#### **Biomass yield and quality analyses**

##### *Determination of edible biomass*

The crude fibre (Van Soest *et al.* 1991, Li *et al.* 2013), sugar, protein and fat of wheat seeds were determined, respectively, under different conditions according to the method described by Gao (2000). The TKW of wheat seeds was weighed, respectively, under four different light sources (Groos *et al.* 2003).

##### *Determination of inedible biomass*

For the determination of inedible biomass components, plant tissues were dried in an oven for 48 h at 70 °C before weighing. The content of neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL) and acid-insoluble ash (Ash) in wheat straw was determined according to Van Soest *et al.* method (Van Soest *et al.* 1991) using FIWE six raw fibre extractor (VelpScientifica, Milan, Italy).

##### *Data statistics*

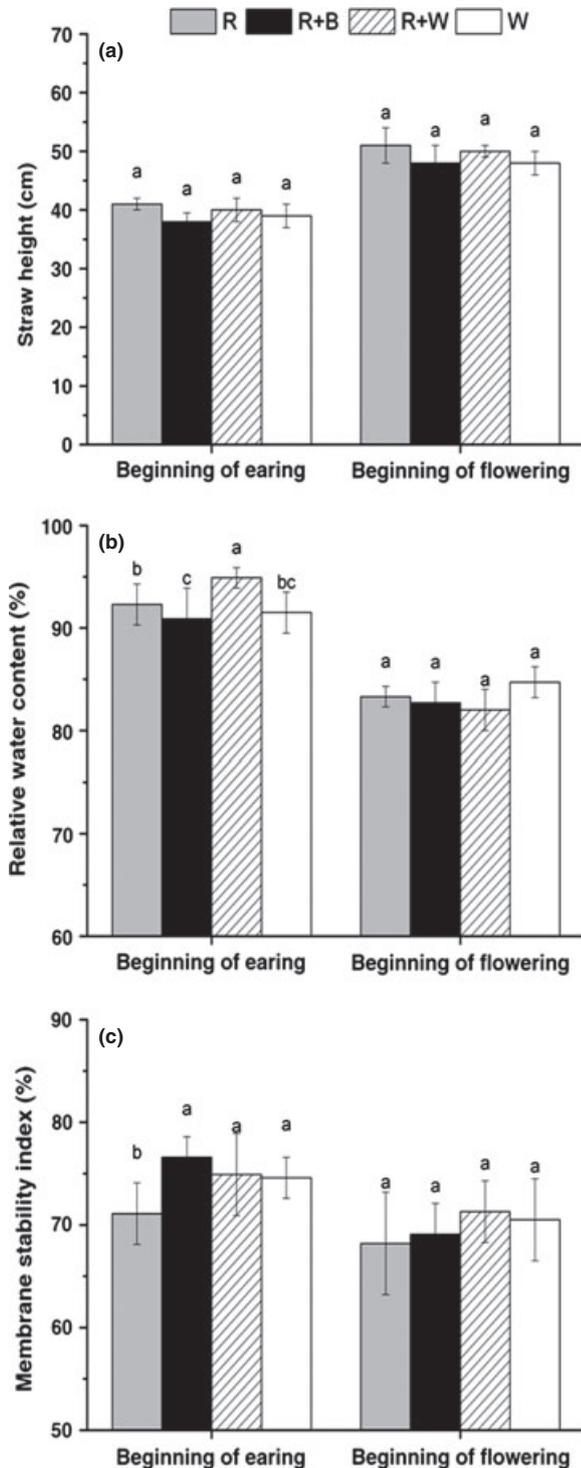
All experiments were performed in triplicate. The average value of total six measurements  $\pm$  standard deviation was regarded as the final result. All statistical analyses were performed using SPSS 18.0.  $P < 0.05$  was considered statistically significant.

## **Results and Discussion**

### **The response of wheat growth to different treatments**

There was no significant difference in straw height of wheat plants as indicated in Fig. 2A. However, in particular, the wheat height was higher only when the red light was used. Once the blue light was added, the plant height was suppressed at seeding stage. The growth-induced ability of the red light was probably related to the low activity of POD, which may make the stem become extended (Normanly *et al.* 1997). In contrast, the blue light was able to dwarf the plant. From earing to flowering period, the plant height of R + B was 1–1.5 cm lower than that of W and 3–4 cm lower than the single red light. The compound light was beneficial to wheat growth at seedling stage.

The most obvious influence of different light sources on RWC of wheat leaves happened at the beginning of earing and flowering (Fig. 2B). With the condition of R + W, the



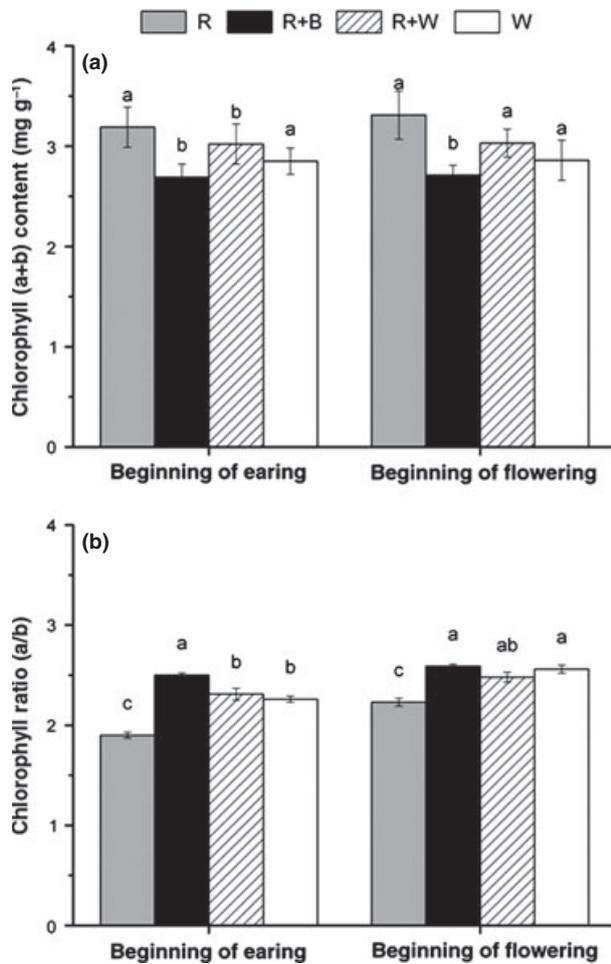
**Fig 2** Response of straw height of wheat plants to different light treatments (a). Relative water content (b), membrane stability index (c) of leaves in wheat plants at different stages of ontogenesis under red LEDs only (R), mixtures of red plus blue LEDs (R + B), mixtures of red plus white LEDs (R + W) and white LEDs (W). Vertical bars are means  $\pm$  S.D. Within each graph, bars labelled with lowercase letters are significantly different at  $P \leq 0.05$ .

RWC was the highest at earing stage, wherein the transpiration strengthened and the plant growth was vigorous. However, the more RWC existed in leaves, the less reflectivity of leaves was, which would affect the optical property. At flowering stage, the minimum RWC occurred, which was more beneficial for accumulating energy to perform self-pollination. Therefore, photosynthesis, transpiration and water-use efficiency are strongly linked to the water regime of plants.

MSI gradually reduced during the development and growth of wheat plants (Fig. 2C). In the single red light, the level of MSI was low during the whole life cycle of wheat plants.

#### The response of photosynthetic characteristics to different treatments

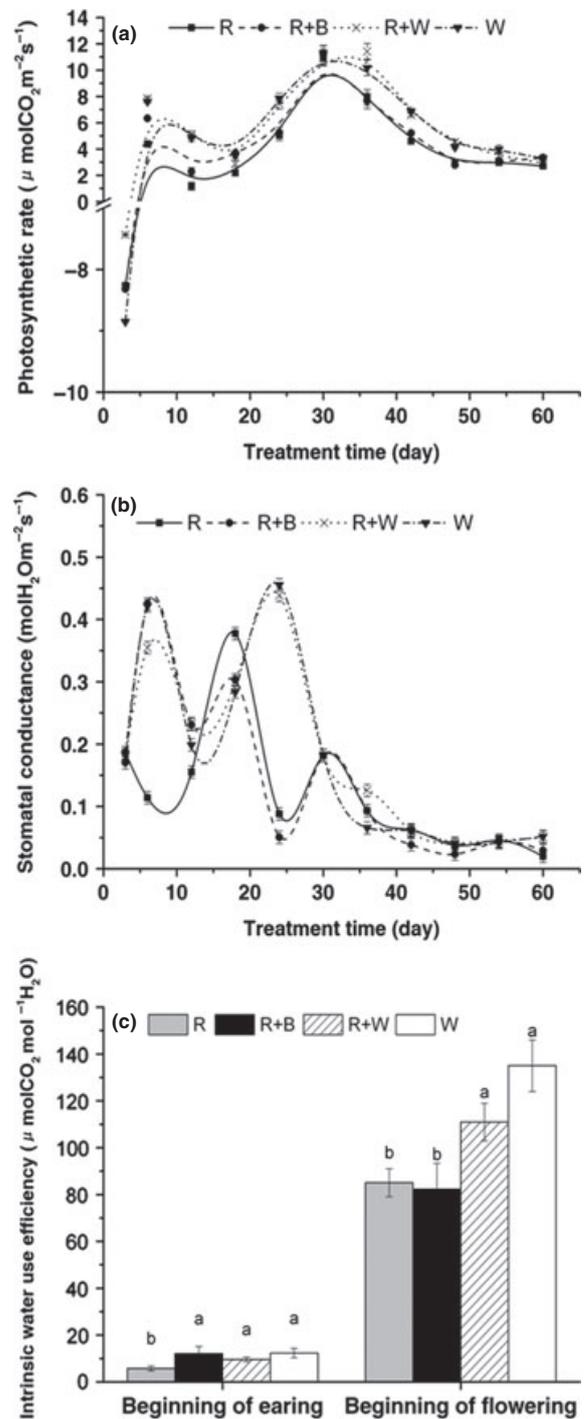
Lighting system is a very important element for chlorophyll synthesis. Light sources with different wavelengths affect different photoreceptors of plants to control pigment synthesis (Stuefer and Huber 1998). Our results showed that the red light was in favour of increasing the total content of chlorophyll (Chl a and Chl b) in wheat leaves (Fig. 3A). By contrast, the addition of the blue light resulted in the relatively low content of chlorophyll. These findings contrast with previous studies where the chlorophyll content of birch leaves was shown to reach peak in the blue light, which was twice more than that in the red light (Saebo et al. 1995). This suggests that light quality has different effects on cytochrome accumulation of different plant species. In addition, the ratio of Chl a/Chl b in wheat leaves was increased under the blue light, but for Chl b/Chl, a ratio was increased under the red light (Fig. 3B). The red light promoted chlorophyll synthesis more effectively than leaf growth. However, the blue light promoted chlorophyll synthesis slightly, mainly because the promotion on leaf growth was weak so that the accumulation of the chlorophyll was restricted indirectly. Besides, compared with the single red light, the promotion of chlorophyll synthesis was weaker when the blue light or white light was mixed into red light, indicating the purity of light quality also affects chlorophyll content. The total chlorophyll content maximized after 30 days of wheat germination during which the photosynthetic intensity peaked. The time of high photosynthetic efficiency in wheat plants was longer under R + W condition than that under W condition (Fig. 4A). This could be because the high stomatal conductance resulted in more fixed content of  $\text{CO}_2$  (Fig. 4B), which was more conducive to wheat jointing and heading and the accumulation of organics. In the single light or some compound lights, both photosynthetic rate and stomatal conductance were lower than those in the white light. Supplementary



**Fig 3** Response of chlorophyll (Chl a and Chl b) contents (a) and ratio (b) of wheat plants to different light treatments. Vertical bars are means  $\pm$  S.D. Within each graph, bars labelled with lowercase letters are significantly different at  $P \leq 0.05$ .

lighting was known to increase shoot and root dry weight via increased photosynthetic rate in celery, tomato, broccoli, lettuce and scallion (Masson et al. 1990, Levine and Paré 2009).

For plants grown under white light conditions,  $A/g_s$  was  $\sim 12 \mu\text{mol CO}_2 \text{ mol}^{-1} \text{ H}_2\text{O}$  at the beginning of earing and  $\sim 134 \mu\text{mol CO}_2 \text{ mol}^{-1} \text{ H}_2\text{O}$  at the beginning of flowering (Fig. 4C). Under R + W condition,  $A/g_s$  increased 10 times from earing to flowering and 11 times vs. W conditions. Under R and R + B conditions,  $A/g_s$  also increased, but there was a significant difference from earing to flowering compared to measurements taken under W and R + W conditions (Fig. 4C). Compared with R and R + B conditions, under R + W and W conditions, photosynthetic rate was higher during their whole life cycle. From Fig. 4B, there were two peaks of stomatal conductance under both R + W and W conditions from 3 to 30 days



**Fig 4** Curves of variations in the photosynthetic rate (a), stomatal conductance (b) and  $A/g_s$  (c) for the flag leaves of wheat plants in different light treatments. Vertical bars are means  $\pm$  S.D. Bars labelled with lowercase letters are significantly different at  $P \leq 0.05$ .

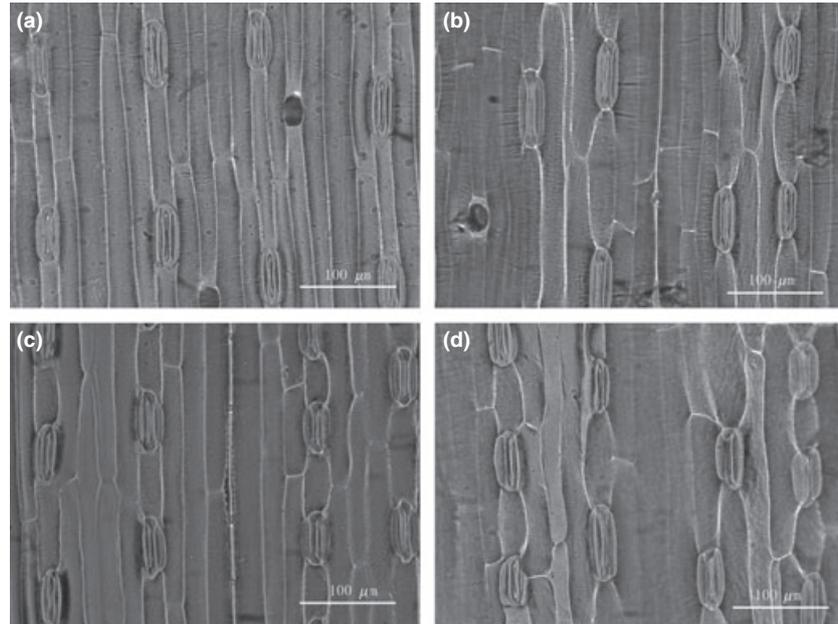
after planting. However, under R and R + B conditions, the number of peaks decreased and the process exhibited hysteresis, which means the controlling gene and active

protein may need to be activated by light except red and blue spectra in photoreceptors. From earing to flowering, the  $A/g_s$  increased, at least in wheat plants (Fig. 4C), allowing the plant to more efficiently use water under controlled conditions.

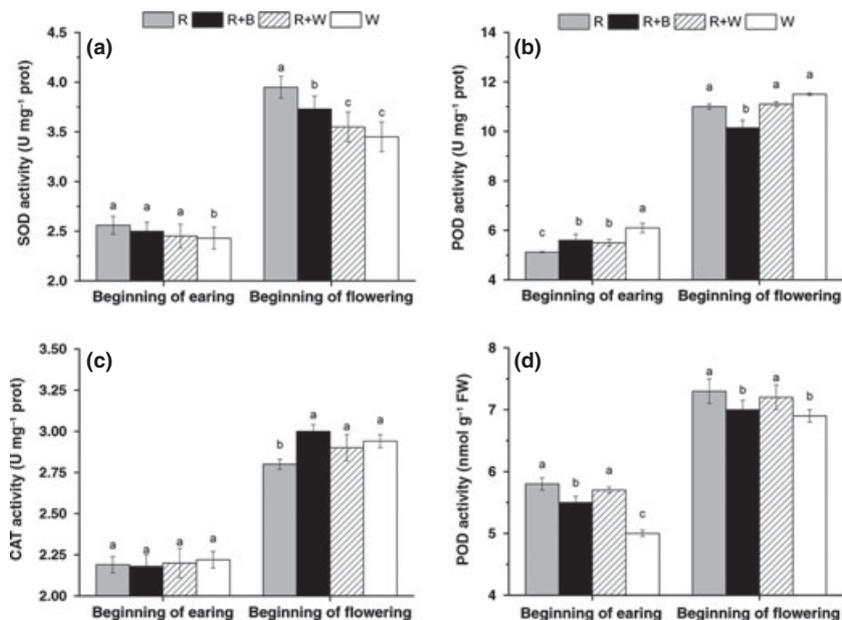
Increased  $CO_2$  typically increases the rate of photosynthesis in many  $C_3$  species by increasing the intercellular  $CO_2$  concentration, enhancing the efficiency of carboxylation, and reducing photorespiration (Bowes 1993, Robredo et al. 2007). Our cultivars under R + W and W conditions

showed greater photosynthetic rate (Fig. 4A) and  $A/g_s$  (Fig. 4C) at increased  $CO_2$  permitting a higher availability of carbon skeletons to produce more biomass (carbon allocation to synthesize new biomass) in cultivars. However,  $A/g_s$  was lower under R and R+B conditions, indicating that under these circumstances, the wheat plants needed more water to fix the same amount of carbon and thus they were less efficient in using water.

At seeding stage, the length of epidermal cells between stomata was getting longer, with the tendency



**Fig 5** Effects of different light qualities on wheat leaf stomata. a (R), b (R+B), c (R+W), d (W), Bar = 100  $\mu m$ .



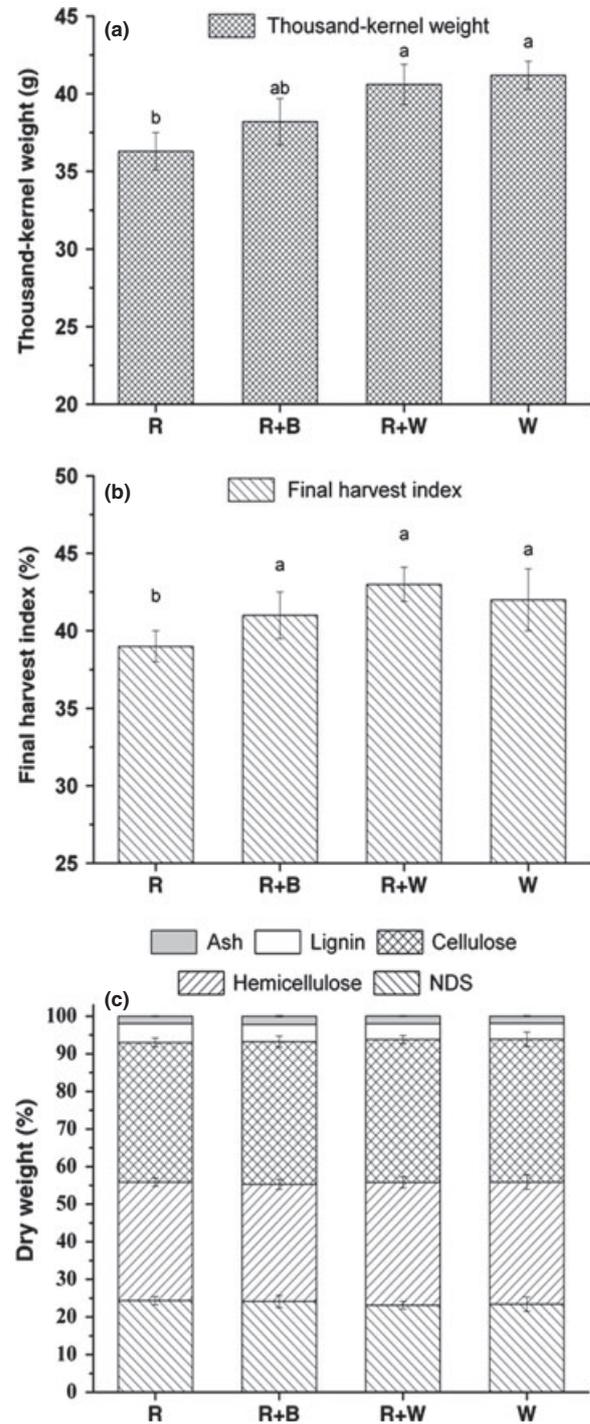
**Fig 6** Correlation of activity of SOD (a), POD (b), CAT (c) and MDA content (d) in leaves of wheat plants at different stages of ontogenesis under red LEDs only (R), mixtures of red plus blue LEDs (R + B), mixtures of red plus white LEDs (R + W) and white LEDs (W). Vertical bars are means  $\pm$  S.D. Bars labelled with lowercase letters are significantly different at  $P \leq 0.05$ .

of growth elongation (Fig. 5A), leading to the reduction in CO<sub>2</sub> assimilation efficiency when photosynthesis was on. The epidermal stomatal density in the red light was sharply lower than that in a combined light source. These observations demonstrate that different combinations of light sources impact differently the morphology and the distribution of epidermal cells and stomata of wheat leaves.

#### Response of antiretroviral system to different light treatments

The reactive oxygen defensive system in creatures consists of SOD, POD and CAT. This system plays a critical role in preventing or decreasing the form of hydroxyl radicals and eliminating superoxide radicals, H<sub>2</sub>O<sub>2</sub> and peroxide (Fridovich 1986, Halliwell 1987, Wise and Naylor 1987, Imlay and Linn 1988, Becana *et al.* 1998). As for the response of antioxidant enzyme to different light sources, the level of SOD in wheat plants was an important index of ageing and death. At the beginning of earing stage, the response of enzymes to the light sources with different wavelengths was quite sensitive. SOD in the white light was usually at the lowest level (Fig. 6A). In contrast, the contents of CAT and POD were at the highest level (Fig. 6B–C). When plants were under a single light or some compound lights, the burst out of reactive oxygen resulted in the increase of radicals, peroxidation of cell membrane, which might bring damage to cell membrane. Compared with CAT, the activity of POD and SOD in the red–blue light was connected to the light quality.

Peroxidase, SOD and CAT are key protective enzymes in plants, which are involved in important physiological activities such as antimicrobial damage, antipathogen invasion and growth (Davies 1987, Wojtaszek 1997, Maffei *et al.* 2006). The level of these enzymes reflects the situation of physiological activity of plants. Peroxidase has been proven to have the function of oxidase IAA (Normanly *et al.* 1997). Low level of POD promoted the growth of over-ground part of wheats, especially for the elongation growth. Peroxidase also could prevent the poison of internal metabolites such as H<sub>2</sub>O<sub>2</sub>, avoid degradation of chlorophyll and the generation of reactive oxygen. SOD was able to avoid the poison of radical. Moreover, the activities of SOD, POD and CAT were related to plant senescence. With the senescence of plants, the activity of them dropped very fast. In the present study, we found that the activities of these three enzymes altered under different light qualities as shown in Fig. 6A–C. At the beginning of flowering, activity of the three enzymes was higher than that at the beginning of earing. The reason may be that the expression of wheat enzyme genes is much more promoted during the flowering stage, ensuring a good growth of plants. Meanwhile, it



**Fig 7** TKW (a), final harvest index (b) and components of inedible biomass (c) of samples in different light treatments. Vertical bars are means  $\pm$  S.D. Bars labelled with lowercase letters are significantly different at  $P \leq 0.05$ .

also indicates that there might be complementary effect and additive effect in combined lights. However, the mechanism needs to be further investigated.

At the beginning of earing and flowering stages, when the red light was added, MDA was accumulated in wheat leaves (Fig. 6D). This resulted in peroxide effect to cells and thus reactive oxygen was accumulated. The single red light showed strongest stress effect in wheat seeding stage. When the blue light was jointed, stress effect became weaker.

### The responses of biomass yield and quality to different treatments

When wheat was in the single red light, TKW was 15.3 % lower than that in the white light during the whole life cycle (Fig. 7A). Compared with the white light source, the harvest index of wheat was higher than that in the red–white light source, and the percentage of inedible biomass was lower (Fig. 7B). These results were much more beneficial to continuous cultivation under energy confinement and high-recycling conditions.

The soluble sugar content of the edible part of wheat plants decreased with narrowing spectrum such as in R or R + B treatments, and the accumulation of carbohydrate also decreased (Table 1). This finding was similar to the cases of birch blades reported by Saebo et al. (1995). The accumulation of starch grain in mesophyll cells in the blue light was less in comparison with that in the red light. It might be because that the red light restrains the export of

photosynthate from blades, thereby increasing the accumulation of starch grain. However, the excessive accumulation of starch grain was helpless for blade photosynthesis (Bondada and Syvertsen 2005).

To investigate the influence of combined light sources with different wavelengths on inedible part of wheats, we determined the contents of lignin, cellulose and hemicelluloses. The results showed that the single red light (R) was beneficial for the increase of lignin content (Fig. 7C), with maximal mass fraction of 4.98 %. However, when the compound light was involved, the lignin content decreased to 4.55 % in R + B, 4.13% in R + W and 4.25 % in W treatment. The content of cellulose and hemicelluloses increased in R + W and W treatments. Furthermore, the percentage of cellulose and hemicelluloses from high to low was R + W, W, R + B and R, respectively, which was helpful for wheat straw degradation in BLSS. These observations are consistent with previous study, wherein the red light was shown to lead to the enhancement of cortical cell activity and the accumulation of lignin in broad bean seeding (Badiani et al. 1990).

The results of impact of light source with different wavelengths on the dry weight of each part of wheats showed that the red light was conductive to the growth of overground part in wheats, including the thickening of stem, the increase of overground biomass (Table 2). Adding the blue light was beneficial for the development

**Table 1** The contents of nutrients of wheat seed in different treatments (g/100 g) (Mean±S.E.)

Items	Treatment			
	R	R + B	R + W	W
Soluble sugar	7.45 ± 0.11 <sup>b</sup>	7.65 ± 0.08 <sup>b</sup>	8.03 ± 0.12 <sup>a</sup>	8.17 ± 0.16 <sup>a</sup>
Carbohydrate	69.51 ± 4.61 <sup>a</sup>	72.17 ± 3.95 <sup>a</sup>	72.39 ± 6.11 <sup>a</sup>	73.87 ± 4.64 <sup>a</sup>
Rough protein	22.68 ± 1.99 <sup>a</sup>	23.03 ± 1.91 <sup>a</sup>	22.14 ± 1.97 <sup>a</sup>	21.09 ± 0.99 <sup>a</sup>
Rough fat	2.01 ± 0.21 <sup>a</sup>	2.05 ± 0.16 <sup>a</sup>	2.12 ± 0.45 <sup>a</sup>	2.08 ± 0.33 <sup>a</sup>
Ash	0.33 ± 0.03 <sup>b</sup>	0.63 ± 0.11 <sup>a</sup>	0.66 ± 0.13 <sup>a</sup>	0.17 ± 0.10 <sup>b</sup>
NDS	33.52 ± 3.10 <sup>a</sup>	36.47 ± 3.33 <sup>a</sup>	34.34 ± 3.89 <sup>a</sup>	31.54 ± 2.09 <sup>a</sup>
Hemicellulose	2.44 ± 0.35 <sup>b</sup>	3.14 ± 0.13 <sup>a,b</sup>	2.82 ± 0.19 <sup>b</sup>	3.67 ± 0.28 <sup>a</sup>
Cellulose	4.01 ± 0.12 <sup>b</sup>	3.17 ± 0.26 <sup>c</sup>	4.99 ± 0.09 <sup>a</sup>	4.06 ± 0.25 <sup>b</sup>
Lignin	0.76 ± 0.02 <sup>b</sup>	1.19 ± 0.07 <sup>a</sup>	0.47 ± 0.05 <sup>c</sup>	0.51 ± 0.04 <sup>c</sup>
Nitrogen	3.62 ± 0.12 <sup>a</sup>	3.84 ± 0.32 <sup>a</sup>	3.54 ± 0.21 <sup>a</sup>	3.38 ± 0.31 <sup>a</sup>

Mean values with the same letter were not significantly different, based on ANOVA followed by Tukey's test at  $P \leq 0.05$ .

**Table 2** The proportion of different parts of wheat plants (DW, %) (Mean ± S.E.)

Treatment	Root	Leaf	Stem	Spike	Seed
R	7.11 ± 0.21 <sup>c</sup>	8.16 ± 0.58 <sup>a</sup>	32.21 ± 1.91 <sup>a</sup>	52.52 ± 3.87 <sup>a</sup>	37.51 ± 1.63 <sup>b</sup>
R+B	9.21 ± 0.27 <sup>a</sup>	8.26 ± 0.42 <sup>a</sup>	27.56 ± 1.58 <sup>b</sup>	54.97 ± 4.17 <sup>a</sup>	42.23 ± 2.66 <sup>a</sup>
R+W	8.13 ± 0.19 <sup>b</sup>	8.34 ± 0.44 <sup>a</sup>	28.52 ± 0.95 <sup>a</sup>	55.01 ± 5.89 <sup>a</sup>	44.12 ± 2.33 <sup>a</sup>
W	7.91 ± 0.17 <sup>b</sup>	8.21 ± 0.37 <sup>a</sup>	29.01 ± 2.44 <sup>a</sup>	54.87 ± 4.71 <sup>a</sup>	42.51 ± 2.93 <sup>a</sup>

Mean values with the same letter were not significantly different, based on ANOVA followed by Tukey's test at  $P \leq 0.05$ .

and growth of crop root system. In the blue light, wheat seeding was easy to form roots and the root system was strong. The underground part of biomass was increased by 1.3 % compared with that in the white light. In addition, the harvest index of R + W reached 43.12 %, which was the highest among different light sources.

## Conclusion

Energy consumption and output are two very important standards for evaluating the reliability of artificial light sources when wheats are planted in a controlled environment. Therefore, it is our target of engineering practice to optimize the light source of wheat production module on the ground or in the space plant factory. The light quality is a very significant environmental factor that affects wheat growth. Our study demonstrates that the plants were spindling and the output was very low when wheat was in the single red light. However, after a certain amount of the blue light or white light was mixed into the red light, the tendency of spindling was restrained gradually. The wheat harvest index arrived at the highest and the lignin content of inedible biomass was the lowest in R + W condition, which was more conducive to substance recycling. Therefore, the addition of white light is an important way to guarantee good quality, high yield and stable yield of crops in BLSS. Furthermore, compared with W, R + W can largely reduce energy consumption and thermal radiation, which provides an important reference value for establishing the related testing devices of space flight.

Our findings might be used to design specifically balanced LED system for supporting plant growth, especially for specialized applications, such as in space. Moreover, our present research provides a pathway of improving crop quality from the light source view for engineering technicians who are in charge of facility cultivation. It also offers new thoughts about straw degradation. These will play a critical role in further development of ground, space plant factory and integral experiments.

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