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Original Article

Perceived Impact of Light Quality on Seed Germination and Photosynthetic Pigments in Rice (*Oryza sativa* L.)

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Abstract

The present investigations attempted to study the effects of different colour of light on seed germination, hypocotyl growth, mobilization efficiency (ME), vigor index (VI), biomass production and content of photosynthetic pigments (Chl a, Chl b and Carotenoids) in Rice (*Oryza sativa* L.) cv. Sugandha 5, an important staple food crop of the world. Germination rate was found maximum in red light (98%), followed by blue (94%) and natural light (93%) at 84 hours while, almost no germination was shown in green light even after 96 hours. Yellow light caused significant reduction in % germination (85). The seedlings obtained under different lights revealed variation in biomass production (fresh weight of root and shoot). Root and shoot growth were observed highest in red light and the order of biomass production was red > natural > yellow > blue > green. ME and VI of rice seedlings recorded maximum (128.22 and 43.26 respectively) in red and minimum in yellow light (116.09 and 1.20 respectively). Both, ME and VI could not be estimated under green light due to absence of well-marked hypocotyl. The contents of photosynthetic pigments in seedlings indicate synthesis of photosynthetic pigments highly dependent on light quality. Chlorophyll b and carotenoids were recorded highest in natural light whereas total chlorophyll and chlorophyll a were highest in red light in comparison to other light treatments. These findings indicate possibility for exploring light quality for manipulation of germination and seedling health of crop plants in general and *O. sativa* in particular.

Keywords: Germination; Light Wavelengths; *Oryza Sativa*; Photosynthetic Pigments; Rice; Seedling Vigour.

Introduction

Light is an indispensable factor for plants, being the energy source for their growth and development, however, besides being essential to photosynthesis, it also serves as an environmental signal which, when perceived, triggers changes in plant metabolism and development (Jiao *et al.* 2007). The effects of light on a plant community, especially in terms of environmental signaling, are not only related to the magnitude of the photosynthetic photon flux i.e. the amount of light, but also to the direction, duration, and particularly, the quality of light available to plants (Majerowicz and Peres, 2004). Besides importance for growth, development, and environmental perception, experiments have shown that light has a connection with a number of other plant processes, including related to biotic and abiotic stress tolerance (Svyatyna and Riemann, 2012). In this regard, light and other environmental stimuli often work together to trigger the development of specific responses in plants (Jiao *et al.* 2007). The light quality reaching the soil/plants and the absorbing organs varies according to many factors which include the time of the day, season, geographic location, atmospheric gases and moisture, clouds, smoke, dust, and other pollutants in the air, topography, presence of barriers including plants, plant architecture, and location of absorbing plant organs within the canopy. Nature has produced a

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number of light absorbing molecules that enable organisms to respond to changes in the natural light environment. The changes in the light signal/quality (wavelength) influences various physiological processes (i.e. intra- and inter-cellular differentiation, seed germination and seedling growth, photosynthesis, flowering etc.), depending on the developmental stage and plant species or studied plant part (He *et al.* 2017). Green light, in the process of seed germination of *Arabidopsis*, stimulates the early elongation of the stems, antagonizing the growth inhibition by light whereas the *white* and *red* light, in ferns can cause delay of the chlorophyll loss due to senescence (Burescu *et al.* 2015). It has been mentioned earlier that light is absolute factor regulating the seed germination process in numerous plant species (Jala, 2011; Lal and Sachan, 2017).

Over a study period of 4 weeks, Jala (2011) found that the seeds of *Nepenthes mirabilis placed* under white and red light germinated first and those placed under green light were the last once to germinate and the highest average speed of emergence was also recorded highest for seedlings under red light. Pigments are biomolecules that absorb light usually in the range of 320 to 760 nm and their biosynthesis in seedlings is highly dependent on light quality they perceive.

Burescu *et al.* (2015) studied the effect of different wavelengths LED lights on the growth of Spruce (*Picea abies* L.) plantlets and observed increased biosynthesis of chl a, chl b under blue and yellow light, respectively. Carotenoid synthesis was also significantly enhanced in yellow light treated plantlets. However, all pigments analyzed were found lower in plants raised under green light than in other light treatments.

Studies on effect of light quality on early development of rice cultivars under laboratory and green house environment have shown that red light has promontory effect where as green light turns to be inhibitory, and interaction between light x temperatures proved role of red light in promotion of cold tolerance (Venske *et al.* 2013).

However, the information on effect of light quality (wavelengths) on seed germination, seedling health and photosynthetic pigment is not available in literature for most of the crop plants including rice. Considering these diverse effects of light, the present study was carried to investigate the effect of different colours of light (natural, red, blue, yellow, and green) on seed germination, hypocotyl growth, biomass production, mobilization efficiency (ME), vigor index (VI) and photosynthetic pigments in Rice (*Oryza sativa* L., Family- Poaceae), a monocotyledonous angiosperm and most important staple food crop of the world, mainly cultivated and consumed in Asian countries.

Materials and Methods

Rice (*Oryza sativa*) cv. Sugandha 5 was used as experimental material for germination using Knop's nutrient solution. Knop's solution (10X) was prepared by dissolving 0.8 mg of $Ca(NO_3)_2.4H_2O$, 0.2 mg of KNO_3, 0.2 mg of K_2HPO_4, 0.2 mg of $MgSO_4.7H_2O$ and traces of $FeSO_4$ in 50 ml of distilled water (DW). This stock solution was further diluted with DW for the preparation of 250 ml nutrient solution for germination of seeds.

Hundred surface sterilized seeds of *O. sativa* were grown in a series of 5 petri dishes, each containing equal amount of sand and moistened with 30 ml of nutrient solution. These petri dishes were exposed to light of different wavelength (i.e. natural, red, yellow, green and blue light) provided using with different color LEDs of Philips Company (different wavelengths) for the duration of 96 hours. The experiment was designed to assess the effect of different types of light: (white (fluorescent), red light with peak emission of 660 nm, green light with peak emission of 550 nm, blue light with peak emission of 490 nm, and yellow light with peak emission of 600 nm light). The adopted photoperiod in the experiments was 16 hrs light/24 hrs.

Mobilization efficiency (ME) in germinating seedlings of each light treatment was estimated by the method of Mohan *et al.* (1996) with the following formula:

ME = Dry weight of seedlings/ Dry weight of cotyledon X100

Vigor index (VI) of germinating seedlings against each light treatment was estimated by the method of Abdul-Baki and Anderson (1973) with the following formula:

VI = % germination X average hypocotyl length

The amounts of chlorophyll a, chlorophyll b, total chlorophyll and carotenoid were quantified in mg/gm fresh weight according to method of Arnon (1949) with sight modification (Bansal *et al.* 1976). On 5^{th} day 80% (v/V) acetone homogenate of plants was incubated at 4°C for 24 hours, and then the homogenate was centrifuged at 5000 revolution per minute for 15 minutes. The supernatant was used to determine OD of each sample/treatment at 480, 510, 630, 645, 652, 663 and 665 nm using Spectronic 20 Bousch and Lomb spectrophotometer. The pigment values were calculated as:

Total Chlorophyll = $O.D_{652}$ X 1000/ 34.5

Chlorophyll a = 15.6 X ($O.D_{665}$) – 2 X ($O.D_{645}$) – 0.8 X ($O.D_{630}$)

Chlorophyll b = Total Chl – Chl a

Carotenoids = 7.6 ($O.D_{480}$- 1.4 X $O.D_{510}$)

The actual pigment content (mg/g FW) was computed as pigment value X V/1000 X 1/W

Where

V - volume of acetone extract (in ml) and

W - weight of the leaf tissue used (in g).

Results and Discussion

The effects of different light treatments on germination of rice (*Oryza sativa*) seeds are summarized in Table 1. The extent/rate of germination has been found to depend on light quality (wavelength) and exposure period. The germination was recorded maximum (98%) in red light at 84 hours duration and this wavelength was found most suitable than other lights tested. In the study by Abdullateef and Osman (2011), red light (660 nm) had better influence on germination in *Stevia rebaudiana* Bertoni seeds than white light (400-700 nm) and present result are in conformity with germination in *Stevia rebaudiana*. Thereafter, in natural light the rate of germination was relatively much faster than other light treatments with maximum germination of 93% at 84 hours. Sharma and Sen (1975) also recorded highest percentage of germination in *Merremia* species with red light treatment. Blue light resulted in germination (94%) comparable to natural light and yellow light showed moderate germination (85%) at 84 hours. Almost no germination was noticed under green light even after 96 hours. The germination process did stop at 84 hours in all the light treatments. It has been observed previously in case of *Nepenthes mirabilis* and *Vigna unguiculata* that seeds placed under white and red light germinated first and those placed under green light were the last ones to germinate (Jala, 2011; Lal and Sachan, 2017). However, upon completion of germination, Jala (2011) recorded highest % germination in yellow light followed by red and natural light. The present results on germination show deviation from Jala (2011) but conform to findings of Colbach *et al.* (2002) on *Alopecurus myosuroides* and Ambika (2007) on *Chromolaena odorata* seeds.

The effect of different light wavelengths on biomass production in rice seedlings is summarized in Table 2 in terms of root fresh weight (FW), shoot fresh weight (FW), seedling dry weight, cotyledon dry weight and hypocotyl length. Seedlings under red light showed highest biomass yield in terms of root, shoot and cotyledon fresh weight. The order of biomass production under different light qualities was red > natural > yellow > blue > green. Irradiation with blue was not suitable for biomass growth (particularly root) in *O. sativa* although it turned superior over yellow light in respect of % germination. Blue light

Table 1: Germination (%) of rice (*O. sativa*) cv. Sugandha 5 in different light wavelengths

Treatments	% Germination							
	12 h	24 h	36 h	48 h	60 h	72 h	84 h	96 h
Natural Light	0	14	32	61	92	93	93	93
Red Light	0	23	81	91	95	98	98	98
Blue Light	0	21	36	75	84	90	94	94
Yellow Light	0	02	51	65	77	85	85	85
Green Light	*	*	*	*	*	*	*	*

*No germination

Table 2: Growth response of root, shoot and hypocotyls of germinating seedlings of rice (*O. sativa*) cv. Sugandha 5 in different light wavelengths

Treatments	Root Length (cm)	Root FW (g)	Shoot Length(cm)	Shoot FW (g)	Seedling Dry Wt. (g)	Cotyledon Dry Wt. (g)	Hypocotyl length (cm)
Natural light	1.72± 0.22	0.008±0.004	2.92± 0.17	0.146± 0.017	0.011± 0.004	0.013± 0.006	1.20± 0.19
Red light	1.92± 0.29	0.011± 0.002	3.76± 1.07	0.150± 0.013	0.012± 0.005	0.017± 0.009	1.58± 0.23
Blue light	0.40± 0.10	0.006± 0.003	0.32± 0.07	0.102± 0.016	0.004± 0.001	0.007± 0.003	0.66± 0.29
Yellow light	1.56± 0.23	0.012± 0.004	3.20± 0.64	0.129± 0.013	0.007± 0.002	0.012± 0.006	1.34± 0.17
Green light	*	*	*	*	*	*	*

*Parameters not measurable, Data on FW and DW is shown up to third place after decimal to reveal the differences resulted in smaller cotyledons and leaves with relatively low FW and Seedling dry weight and caused significant reduction in hypocotyls length. Horizontal and vertical expansion of shoot, particularly leaves is genetically controlled developmental process (Tsukaya, 1998) and irradiation with blue light seems to cause imbalance in expression of concerned genes leading to inhibition of leaf expansion. For the lettuce crop, the fresh and dry weight accumulations were higher under the RB (red-blue) treatment (Mickens, 2012). Snowden (2015) also observed significant reduction in dry biomass in radish under green light at the high level among the comparable treatments. In contrast to present findings, green light is reported to stimulate the spruce (*Picea abies* L.) seed germination and plant growth whereas the blue light inhibits hypocotyl elongation (Burescu *et al.* 2015).

Variation was recorded in mobilization efficiency (ME) and vigor index (VI) in seedlings of rice (*O. sativa*) obtained in different light treatments and both ME and VI were highest (128.22 and 3.26, respectively) under red light followed by blue light (Table 3). Under green light, both ME and VI could not be determined due to lack of well differentiated hypocotyl. Contrary to these findings, Jala (2011) reported that seedling vigor index and germination index were highest under yellow light, followed by red light. In another similar study on *V. unguiculata* (a dicot plant), Lal and Sachan (2017) recorded highest total chlorophyll, chl b and carotenoids in natural light whereas chl a was highest in red light. These findings indicate pigment biosynthesis/content in response to different light wave lengths as a species/genotype specific trait.

The seedlings formed under different lights revealed differences in the quantity of photosynthetic pigments (Table 4). Chlorophyll b and carotenoids were recorded highest in natural light whereas total chlorophyll and chlorophyll a was recorded highest in red light in comparison to other treatments. Natural light also recorded maximum synthesis of carotenoids followed by red, yellow and blue light, respectively. Green light had no measurable pigments because of insufficient growth and development of seedlings. In some instances, green light may function by informing the plant of photosynthetically unfavorable conditions, allowing plants to adjust their compositions and physiology to the available light quality. The chlorophyll a : b ratio also varied in different light treatments. In red light, chlorophyll a:b ratio was found maximum (2.07) whereas total chlorophyll : carotenoid ratio was highest (2.72) in blue light in comparison to other lights. Saebo *et al.* (1995) reported that red light is important for the development of the photosynthetic apparatus (plastid differentiation) of plants and a combination of red and blue light is important in the biosynthesis of chlorophyll. The use of red-LED light to drive photosynthesis has been widely accepted due to fact that red wavelengths (600–700 nm) are efficiently absorbed by photosynthetic pigments (Sager and McFarlane, 1977) and the same is evident from the present results. Likewise, the highest inhibition of all the assimilating pigments in spruce (*Picea abies* L) plantlets was observed when exposed to green LEDs (Burescu *et al.* 2015). In a recent study, He *et al.* (2017) explained that the suitable combination of red- and blue-LED light enhances plant growth and

Table 3: Mobilization efficiency and Vigour index of seedlings of Rice (*Oryza sativa*) cv. Sugandha 5 grown in different light wavelengths

Treatments	Mobilization Efficiency (ME)	Vigour Index (VI)
Natural Light	117.03±2.184	2.58
Red Light	128.22±4.344	3.26
Blue Light	120.29±3.245	3.04
Yellow Light	116.09±2.135	1.20
Green Light	*	*

* ME and VI could not be determined because no differential/well-marked hypocotyl present

Table 4: Photosynthetic pigment contents of rice seedling leaves in different light wavelengths

Treatments	Total chl (mg/g FW)	chl a (mg/g FW)	chl b (mg/g FW)	Carotenoids (mg/g FW)	chl a: b ratio	Total chl: carotenoids ratio
Natural Light	0.850	0.526	0.324	0.456	1.62	1.86
Red Light	0.936	0.632	0.304	0.368	2.07	2.54
Blue Light	0.638	0.419	0.219	0.234	1.91	2.72
Yellow light	0.736	0.375	0.361	0.296	1.03	2.48
Green Light	*	*	*	*	*	*

*Not measurable pigment values, Chl- Chlorophyll photosynthetic capacities of *M. crystallinum* compared to red- or blue-LED alone. This observation indicates need for further experiments in range of crop plants using combination of two or more wavelengths to improve germination and seedling vigour.

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