

Photocontrol of Rice (*Oryza Sativa.L*) Development by Excitation of Phytochrome

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ABSTRACT

The different types of abiotic factors play an important role in the growth and development of plants, the light is one of the crucial abiotic factors that enhance plant development. In this assessment, the impact of various light on seed development and interconversion of phytochrome, production of biomass, seedling length on rice varieties Shandar, and IRR-6 were observed. The data indicate under red light 68.09%, far-red light 67.61%, blue light 67.14%, white light 64.29%, and dark 63.80% in Shandar. Similarly, in IRR-6 germination percentage was recorded under far-red light 66.19%, red light 63.33%, white light 60%, blue light 58.57%, and dark condition 58.57%. Plumule and the radical length was maximum in IRR-6 under white light and. The biomass of IRR-6 was maximum under white light 0.33gm followed by red light 0.26gm, far-red light 0.17gm, dark condition 0.2gm, and blue light 0.1gm respectively. Phytochrome is a controlling photoreceptor that works as a dual molecular switch, regulates gene expression of the plant in reaction to signals of light from the surroundings. Five light-harvesting genes of Rice are identifying by using the Arabidopsis thaliana model plant in computational biology/ bioinformatics databases. The discovered genes are cl02879, cl23770, cl21528, cl33336 and cl36851.

Introduction

Rice is a very important grain and main source of food for the world population. It grows approximately one-tenth of the earth's cultivable land. The world's half population relies on rice with the greatest (26.2%) intake of calories [1]. The largest amount of rice is consumed and cultivated in Asia as compared to the world [2]. The third-largest crop of Pakistan is rice after cotton and wheat, and also greater cash earn crop [3] and the most important export commodity. Plants always monitor surrounding lights form utilizing different sensory photoreceptors like phytochromes and modify their metabolism and morphology to adjust to change the conditions of the environment to maximize photosynthesis [4]. In photosynthesis and morphogenesis, light quality has an important role [5]. The yellow, blue, violet, orange, Indigo, and red colors are

release by spectrum visible light. For the photosynthesis of plant 400-700 nm wavelengths are used, which give overall energy for the requirement of the plant.

Spectral of light combination reaching the plants influences their development and growth through the involvement of photoreceptor cells of the plant [6]. Phototropines and cryptochromes are particularly blue light-sensitive, while phytochromes are red light-sensitive than blue light [7]. Red light plays a vital role in the development of the photosynthetic system of plants, may enhance starch aggregation in various species of plant by preventing the photosynthetic translocation away from leaves [8]. Blue light is crucial in chloroplast development, chlorophyll formation, enzyme synthesis, stomatal opening, circadian rhythm (biological clock)

activation of photomorphogenesis, photosynthesis [9,10]. A light-controlled quality about a suitable ratio of far-red or red, the blue light quality is given additional light can enhance biomass and photochemical content of plants under white light [11]. Phytochromes play an important role within all photoreceptors; phytochromes encourage germination, when lights are in favorable condition and control or prevent when light conditions are not favorable, e.g. in the canopy [4]. The phytochromes mainly affect gibberellin signaling and synthesis to encourage germination [12]. The germination study illustrates the different roles of PHY B and PHY A, among PHY A performing a sensor of broadband for down, while PHY B controlling germination of reversible-R/FR induction [13].

Furthermore, PHY E and PHY D also encourage germination [14]. Phytochromes consist of two forms of photo-interconvertible: Far-red-absorbing, an active form, and red-absorbing an inactive form. Chromophores absorb the light cause this to alter the conformation of protein of phytochromes from red-absorbing from to far-red. The first evidence of phytochromes function was observed in lettuce seed germination, in case of legume, the red light was found to activate germination, although far-red light stopped the germination of pulses. Phytochromes work the entire life span of the plant, in function like seedling growth and photoperiodic induction controlling the time of flowering. The regulatory photoreceptor is phytochromes that work as a dual switch of molecular that controlling gene expression of the plant in reaction to the signal of light from the environment [15]. Genes play a vital role during the growth and developmental stages of seeds. They express when some external factors impact on seeds, like the light. Phytochrome observes light and control gene expression. The NCBI aim is to evolve the latest technologies of information to facilitate the understanding of basic molecular and processes of genetics that regulate illness and health [16].

Bioinformatics is described as a tool of analysis and computation to grab and elucidation of biological information. The bioinformatics toolbar comprises the application of computational software including BLAST, which hinge on internet accessibility [17]. Basic Local Alignment Search Tool is the most intensively used resource for analysis of sequence; in the domain of public, it is available. Currently, there is a large number of algorithms of BLAST that may be used to find various database sequences through the web page of BLAST [18]. ORF finder accomplishes a Nucleotide sequence six-frame translation and returns visuals that specify the ORF location founded Conserved domain database is a tool of NCBI database Entrez system and functions as an important tool for interpretation of footprint of a conserved domain in Entrez on the sequence of the protein.

Materials and Methods

The dry seeds of different rice varieties (Shandar, IRR6) were collected from the seed department of Islamabad, Pakistan. The seeds were sterilized by using a 10% hydrogen peroxide solution. The seeds were dipped in 10% hydrogen peroxide for 5 minutes and then washed them with distilled water three times. Seeds were air dry for 30 minutes [19]. Sterilized imbibed seeds (12h at night in dark) of rice varieties (Shandar, IRR6) were taken for the germination test in vitro. In each petri plate (diameter 9cm) double layer of Whatman filter paper was placed carefully and sterilized seeds were kept with gaps on moist filter paper in three replicates for germination under different light treatments (blue, red, far-red and white) [20].

Irradiation

White light fluorescent (Philips TLD18W/54-764) is used for white light treatment [21]. For red, blue, and far-red light Plexiglas filters were used under the white tube light (Philips TLD18W/54-764). Provide two red light the 3mm thick Plexiglas filter was used, one 3mm thick red Plexiglas filter was used between two blue Plexiglas filters for far-red light conditions. Similarly, two blue filters were used for blue light treatment. Provide dark conditions with the help of aluminum foil and placed in the cotton box. Under these light treatment germinations of seeds were observed within one week.

Measurement of Plumule and Radical

Length of Plumule and radical of the seedlings was measure by the help of scale (30 cm), Wet and dry weight of seedlings were recorded by the help of digital balance.

Gene Collection, Identification, and Homology Exploration

The light-harvesting genes are collected from the NCBI (National center of biotechnology information). The 5 genes of model plant *Arabidopsis thaliana* were collected to compare with sample plant Rice (*Oryza sativa*.L) LHG genes [16]. For gene identification of Rice, used Nr to generate relationships among Rice and *Arabidopsis thaliana*.

BLAST (Basic Local Alignment Search Tool)

Open NCBI database BLAST and paste the *Arabidopsis thaliana* selected gene nucleotide sequence in the box. Select Nr and inter sample plant name (*Oryza sativa*). Chosen the option somewhat similar sequence and show results in a new window, after selection clicks on BLAST and wait for the result. The result appeared only chosen the plant that has maximum query cove [22].

ORF- Finder

Arabidopsis thaliana and Rice FASTA sequences were pasted separately in ORF-finder and submit, in few seconds the coding regions of the selected sequence displayed.

Conserve Domain Database

The ORF resulted in sequences of Arabidopsis thaliana and Rice was copy and paste into the conserved domain database and submitted. The results were displayed within seconds; they were shown the conserve domain family, accession, and description of both plants [23].

Statistical Analysis

The data were analyzed with analysis of variance (ANOVA) using the statistical package Genstat 9.2 (VSN International Ltd., Hemel Hempstead, Hertfordshire, UK). The mean values were compared

and separated based on Fisher's least significant difference (LSD) test.

Results

Seed Germination of Rice Verities at Optimal (20°C) Temperature Under Various light stresses

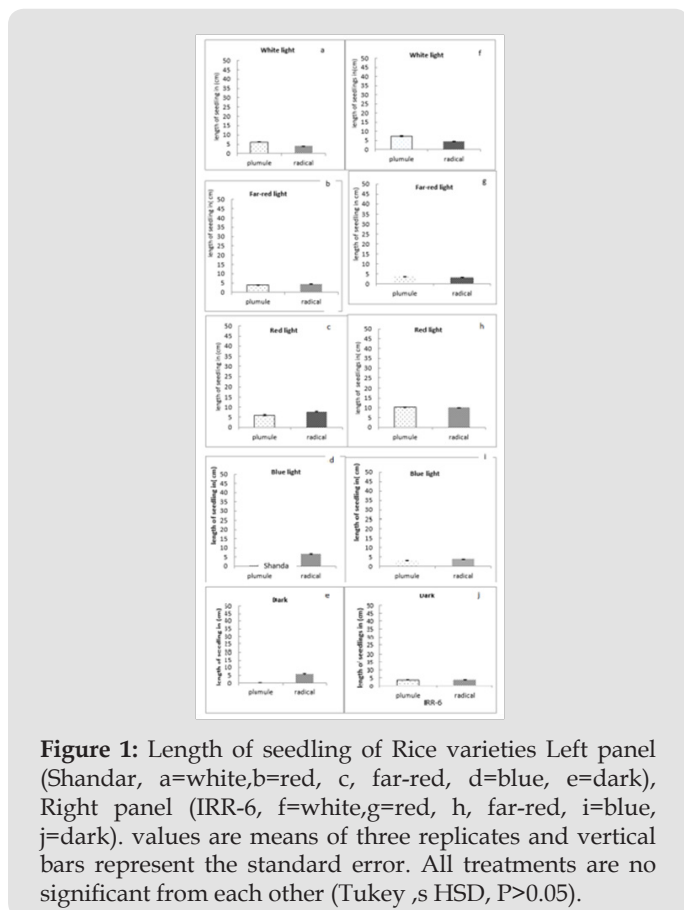
The two verities Shandar and IRR-6 were growing under different light stresses (white light, red light, far-red light, blue light, and dark) at the optimal temperature (20°C). The germination was started at 2nd day in all light conditions. The maximum seeds germination was seen in IRR-6 under all light conditions as compared to the Shandar seeds. Maximum germination observes under red light (68.09%). Other light germination percentage was (67.61%) under white light, (67.14%) under blue light (64.29%) under far-red light and (63.80%) under dark condition (Table 1).

Table 1: Germination % rate of shandar and IRR-6 under various light conditions.

Treatments (Germination %)	White light	Red light	Far-red light	Blue light	Dark
Shandar	63.33±1.09 ^a	60±0.85 ^a	60±0.85 ^a	58.57±0.23 ^{ab}	58.57±0.23 ^{ab}
IRR-6	67.61±0.96 ^a	68.09±0.83 ^a	64.29±1.04 ^a	67.14±1.09 ^a	63.80±0.90 ^a

Note: Three replicates mean values ± SE and values bearing various letters in a similar column are not significantly various from one another.

Length of Seedlings Rice Verities at Optimal (20°C) Temperature Under Various light stresses



The seedling was shown different length of plumule and radical in Shandar and IRR-6 under white light the maximum length of plumule (7.45cm) show in IRR-6 and also the radical (4.55cm) show maximum length. The shandar seedling show minimum length as measure up to IRR-6. The plumule show (6.08cm) and radical show (3.99cm) under white light (Figures 1a & 1f) The seedling was shown different lengths of plumule and radical in Shandar and IRR-6 under Red light the maximum length of plumule (10.32cm) show in IRR-6 and also the radical (10.08cm) show maximum length. The Shandar seedling show minimum length as compare to IRR-6. The plumule show (6,02cm) and radical show (7.8cm) under red light (Figures 1b & 1g). The seedling was shown different lengths of plumule and radical in shandar and IRR-6 under far-red light the maximum length of plumule (4.01cm) show in shandar and also the radical (4.37cm) show maximum length in shandar. The IRR-6 seedling determines minimum length as measure up to shandar. The plumule (3.55cm) and radical show (3.12cm) under far-red light (Figures 1c & 1h).

The seedling was shown different lengths of plumule and radical in shandar and IRR-6 under blue, Red light the maximum length of plumule (3.32cm) determine in IRR-6 and the radical (6.57cm) give maximum length in shandar. The minimum length of plumule (0.24cm) in shandar while the minimum length of radical (3.87cm) measure in IRR-6 (Figures 1d & 1i). The seedling was shown different lengths of plumule and radical in shandar and IRR-6 under Dark condition the maximum length of plumule (3.87cm) determine in IRR-6 and the radical (5.78cm) show maximum length

in Shandar. The minimum length of plumule (0.18cm) in Shandar while the minimum length of radical (3.89cm) measure in IRR-6 (Figures 1e & 1j).

Biomass of Rice Verities at Optimal (20°C) Temperature Under Various light stresses

The IRR-6 and Shandar show variation in biomass. The maximum biomass was showed in IRR-6 under white light 0.33gm, followed by red 0.26gm, far-red 0.2gm, blue 0.17gm, dark, 0.1gm, respectively. In Shandar under white light 0.21gm, red light 0.18gm, far-red 0.16gm, blue 0.18gm, dark 0.4g respectively (Table 2).

Table 2: Shandar and IRR-6 biomass under various light conditions.

Treatments	Shandar (gm)	IRR-6 (gm)
White light	0.21±0.01 ^{ab}	0.33±0.01 ^b
Red light	0.18±0.01 ^{ab}	0.26±0.01 ^{ab}
Far-red light	0.16±0.01 ^{ab}	0.2±0.01 ^{ab}
Blue	0.18±0.01 ^{ab}	0.17±0.00 ^{ab}
Dark	0.07±0.01 ^a	0.1±0.02 ^a

Table 3: Relative study of Arabidopsis thaliana with *Oryza sativa*.

S.No	Gene ID	Accession No.	Query cover	Families of domain	Model plant Arabidopsis thaliana	Rice (<i>Oryzasativa</i>)
1	LHB1B1351914	cl02879	100%	Chloroa_b-bind	Present	Present
2	LHCA2329064	cl23770	91%	FliH	Present	Absent
3	NPQ1354854	cl21528	87%	Lipocalin	Present	Present
4	FTSH8360212	Cl33336	79%	ftsH	Present	Present
5	FTSH1363727	cl36851	77%	FtsH_fam	Present	Present

Discussion

In this trial, as it was expected, the best results for germination rate, biomass, and seedling length increased by the supplementary lights (red light and white light), which are also observed by [24]. Phytochromes are involved in both light entrainments of plant photoperiodic control and circadian clocks in altered parts of plants. Biological reactions facilitated by plant pigments have been completely presented in seed plants. The Plants have full-grown a factual mechanism that connects various senescence-influencing indications therefore they can raise their versatility in diverse ecological environments [25,26]. The light recipient group accomplishes blue light deduction. They perform immoderately in encouraging the induction of flowering circadian clock, an extension of the primary root, and opening of stomata in Arabidopsis thaliana. Light availability on the development of root it would be the elementary effect of availability of variance sugar to roots because, in shoot existence of photosynthesis, there is important proof for more complex indicating effects of different light environments characteristics In our study the same result observed in the case of root elongation.

Note: Three replicates mean Values ± SE and values bearing various letters in a similar column are not significantly various from one another.

Relative Study of Arabidopsis thaliana with Rice (*Oryza sativa* .L)

The research work carried out on 5 genes of Arabidopsis thaliana that give the information about light-harvesting genes, by using the web site NCBI (national center for biotechnology information), BLAST, ORF and CCD. In the BLAST tool found out the query cover of sample plant Arabidopsis thaliana, the query cover was 100%, 91%, 87%, 79%, and 77%. The BLAST result obtained through Nr (non-redundant). Using ORF to find the coding regions of the sequence. On the conserved domain I am putting the model plant and main plant coded sequence to discover the super families. Four families are present in both sample plant Arabidopsis thaliana and main plant *Oryza sativa* like Chloroa_b-bind, Lipocalin, ftsH, and FtsH_fam. Only one family FliH present in sample plant (Arabidopsis thaliana) and absent in reference plant *Oryza sativa* (rice) (Table 3).

In the case of a far-red light study, our result was revealed the positive characteristics. Through Phytochromes, the Signaling of r/fr light was used by plants that control germination, senescence, and shade avoidance, [6]. The inactive Phytochrome absorbed red light that results in photoconversion into a form of active phytochrome far-red then to inactivate it absorbs far-red light. To identify light connected phytochromobilin is used by phytochrome and light signal is moved which goes complete from convertible conformation differences [27,28]. The blue light strengths are very significant for the growth of vascular plants and especially in the manner of plant development and elongation. Corresponding results concerning the biomass assay and seed germination were approved by the present study. In the same way under the red-light treatments, it is reported that supplementation of blue light can enhance photosynthesis on the leaf of rice (*Oryza sativa*) [29]. In the case of a far-red light study, our result was showed the positive aspects. Through Phytochromes, the Signaling of r/fr light was used by plants that regulate shade avoidance, germination, and senescence [6]. The red light absorbed by inactive phytochrome red that result in photoconversion into a form of active phytochrome far-red then to inactivate it absorbs far-red light.

To detect light connected phytochromobilin is utilized by phytochrome and light signal is transferred which goes through from convertible conformation variations the blue light intensities are very important for the development of vascular plants and especially in the process of plant growth and elongation. Parallel outcomes about the seed germination and biomass assay were confirmed by the present study. The present study explains the mechanisms of light-dependent gene expression for the role of the photosynthesis reaction center in seeds of two Rice species. Up-regulated genes are related with an altered kind of dark and light are doing not only the mechanism of indicator transduction with specific photoreceptor molecules but also the photosynthetic reaction, in the form of Phytochrome in plant *Arabidopsis thaliana*. The receiver of blue light leads chloroplast to evade light [30,31]. Our results created on five light-harvesting genes of Rice compared with the model plant *Arabidopsis thaliana*. It has been revealed that it could also be concerned in response to various light conditions. Rice can respond to beneath the active metabolism of different phytohormones, e.g., JA, ABA, and SA that are planned to be involved in the adjustment of light [32].

Conclusion

In this study, levels of phytochrome, as well as growth properties, were greatly influenced by different types of lights. The maximum seedling length and dry biomass in rice seedlings grown under blue and white light may be related to the fact that the photoreceptor absorbs blue light. Our data also show that different types of lights have marked effects on the various biosynthetic pathways. Most genes were down and up-regulated due to a higher energy level of blue light, however, the relevant mechanism is unknown. Expression levels of different genes were quite change under different light impulsive, that may alter retrograde signaling which sends signals to the nucleus via various cell organelles where control expression of nuclear-encoded genes. Further studies are needed to elucidate the signaling pathway involved in the light wavelength-dependent physiological modifications in plants.

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