EVALUATION OF PLANT GROWTH AND YIELD OF PAKCHOI UNDER DIFFERENT LED-LIGHT SPECTRUM IN VERTICAL GROW-HOUSE

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CERTIFICATE

This is to certify that the thesis entitled **"EVALUATION OF PLANT GROWTH AND YIELD OF PAKCHOI UNDER DIFFERENT LED-LIGHT SPECTRUM IN VERTICAL GROW-HOUSE"** submitted to the Department of Horticulture, Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTERS OF SCIENCE (M.S.)** in **HORTICULTURE**, embodies the result of a piece of bonafide research work carried out by **MARJAN JUI**, Registration No. **14-06238** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that any help or source of information, received during the course of this investigation has been duly acknowledged.

June, 2021

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Dedicated

to

Beloved Parents

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The Author

EVALUATION OF PLANT GROWTH AND YIELD OF PAKCHOI UNDER DIFFERENT LED-LIGHT SPECTRUM IN VERTICAL GROW-HOUSE

ABSTRACT

The present study was carried out at the roof top (indoor structure) of Dr. M. Wazed Mia Central Laboratory, Sher-e-Bangla Agricultural University, Dhaka. during the period from November 2020 to December 2020 to study the improvement of plant growth and yield of Pakchoi under different LED-light spectrum in vertical farming. Six treatments of the experiment *viz*. (i) T_{FL-T5} (Fluorescent light – Full; control), (ii) T_W (White - Full), (iii) T_R (Red - Full), (iv) T_B (Blue - Full), (v) T_{RB} (Red and Blue - 4:1) and (vi) T_{RBG} (Red, Blue with Green - 4:1:1) were laid out in a Randomized Complete Block Design (RCBD) with three replications. PPFD of different LED light was maintained at 115 µmol/m^2/s and duration was 16 hrs per day. Results showed that the highest plant height (24.41 cm), number of leaves plant^{-1} (14.33) and leaf breadth (9.10 cm) were found from the treatment T_{RB} (Red and Blue - 4:1) but the highest leaf length (18.23 cm) was from the treatment T_W (White - Full) whereas the lowest plant height (17.68 cm), number of leaves plant^{-1} (9.00), leaf length (16.50 cm) and leaf breadth (4.22 cm) were found from the treatment T_R (Red - Full). Treatment T_{RB} (Red and Blue - 4:1) also registered the highest stem diameter (1.20 cm), average diameter of whole plant (8.24 cm), leaf base breadth (1.92 cm), fresh weight plant^{-1} (336.67 g), total yield (1.29 kg) and total soluble solids (9.24 °Brix) but the highest vitamin-C content (41.81 mg/100g) was achieved from T_{RBG} (Red, Blue with Green - 4:1:1) whereas T_B (Blue - Full) gave the highest SPAD value (42.04) and moisture content (94.12%). The lowest stem diameter (0.70 cm) was recorded from the treatment T_B (Blue - Full) but the lowest diameter of whole plant (6.08 cm), fresh weight plant^{-1} (120.00 g), leaf base breadth (1.36 cm) and total yield (0.18 kg) of the experimental area were recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1). T_W (White - Full) showed lowest SPAD value (34.60) but T_R (Red - Full) gave the lowest vitamin-C content (38.24mg/100g). From the above results, the treatments of different LED-light spectrum in vertical farming, T_{RB} (Red and Blue - 4:1) can be considered as the best treatment followed by control treatment T_{FL-T5} (Fluorescent light - Full) among all the treatments.

LIST OF CONTENTS

LIST OF CONTENTS (Cont'd)

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

LIST OF APPENDICES

ABBREVIATIONS AND ACRONYMS

AEZ	=	Agro-Ecological Zone
BBS	=	Bangladesh Bureau of Statistics
BCSRI	=	Bangladesh Council of Scientific Research Institute
cm	=	Centimeter
CV %	=	Percent Coefficient of Variation
DMRT	=	Duncan's Multiple Range Test
et al.,	=	And others
e.g.	=	exempli gratia (L), for example
etc.	=	Etcetera
FAO	=	Food and Agricultural Organization
g	=	Gram (s)
i.e.	=	id est (L), that is
Kg	=	Kilogram (s)
LED	=	Light Emitting Diode
LSD	=	Least Significant Difference
m^2	=	Meter squares
ml	=	MiliLitre
M.S.	=	Master of Science
No.	=	Number
PPFD	=	Photosynthetic Photon Flux Density
SAU	=	Sher-e-Bangla Agricultural University
var.	=	Variety
^{o}C	=	Degree Celceous
%	=	Percentage
NaOH	=	Sodium hydroxide
GM	=	Geometric mean
P	=	Phosphorus
K	=	Potassium
Ca	=	Calcium
L	=	Litre
µg	=	Microgram
USA	=	United States of America
WHO	=	World Health Organization

CHAPTER I

INTRODUCTION

With the growing global urban population and the emergence of megacities, there is a huge demand for arable land to meet the food demand and reduce malnutrition. Conventional agricultural practices lead to deforestation of the land for crop production and agricultural intensification to produce higher yield per unit area. These activities have been established to have negative impact on the environment thereby causing soil and water pollution. It is important to consider the use of vertical grow-house technology, which utilizes both horizontal and vertical space, and efficiently uses nutrients, water, and time (off season production with artificial lighting) more effectively to produce higher yield per unit volume of space than the conventional outdoor farming (Rajan *et al*., 2019).

Light plays an important role in affecting plant growth (Snowden *et al*., 2016), photochemical biosynthesis (Dou *et al*., 2018; Mickens *et al*., 2019), and gene expression (Son *et al*., 2012). Therefore, lighting environment can be optimized by management of light conditions and proper design of artificial lights, thus increasing the yield and beneficial nutritional values of plants and decreasing the lighting consumption accounts.

Light is one of the most important environmental factors regulating plant growth, development, and photosynthesis (Claypool and Lieth, 2020; Ouzounis *et al*., 2015). Lighting-emitting diodes (LEDs) are regarded as the most effective light source with the highest potential and are being developed to provide powerful, effective, and environmental emission spectra covering the entire photosynthetically active radiation range to precisely regulate numerous types of light combinations (Avercheva *et al*., 2016). Light quality has more complex impacts on plant morphology and metabolism than light intensity or photoperiod (Chen *et al*., 2017). In addition, light quality induces a series of physiological and biochemical reactions that are mainly promoted through signal transduction pathways involving photoreceptors such as phytochrome, cryptochrome, and phototropin receptors, resulting in the up regulation or down regulation of related gene expression (Landi *et al*., 2020; Tissot and Ulm, 2020).

Plants have varied morphological and physiological responses to specific light spectrum, and the current advancement of LEDs enables one to tailor the spectrum to obtain favorable plant growth or nutritional values (Mickens *et al*., 2018; Park and Runkle, 2018). Epidemiological studies have shown that the consumption of Brassicaceae vegetables can effectively prevent cancer and cardiovascular diseases (Bjorkman *et al*., 2011).

Pakchoi (*Brassica campestris* L. ssp. *chinensis* var. communis) is an annual leaf vegetable belonging to the Brassicaceae family that originated in China, whose leaves are rich in nutritional and functional health-related compounds (minerals, vitamins, flavonoids, glucosinolates, and anthocyanins) (Harbaum *et al*., 2008). It is grown successfully under protected environments. Light is a major limiting factor on the productivity of greenhouse vegetables during a prolonged series of cloudy days, especially in the winter.

Recent technological advances in protected culture in artificial light sources have introduced Light Emitting Diodes (LED) (Li, 2012). It reported a great success among growers during last few years as it is a promising greenhouse lighting solution, over traditional lighting sources (Mitchell, 2012). The main advantage of LEDs over all other lamp types is its energy saving ability (Mitchell, 2012). Other than that, fast switching, higher durability, longer life time, lower thermal radiation, narrow variation in specific wavelength for targeted crops, etc. make them more applicable for protected culture (Olle and Virsile, 2013). The LEDs have become the best choice for highly intensive forms of vertical-grow house as a viable artificial lighting solution. In commercial horticulture industry, light-emitting diodes (LEDs) have tremendous potentiality owing to flexibility of spectral configuration, long life spans, and high energy conversion efficiency (Mitchell *et al*., 2015).

LEDs provide various advantages over the traditional lighting systems, (e.g., greenhouse and open-field) for leafy greens, herbs, and transplants production such as longer lifetime, smaller size, higher photosynthetic efficiency, lesser thermal radiation and higher safety performance and its year-round cultivation (Schuerger *et al*., 1997; Bula *et al*., 1991; Kozai and Niu, 2016a). The direct radiation of heat by LEDs is not seen, but LEDs do produce heat that has to be effectively removed from the system for better functionality (Bian *et al*., 2016). The potentials of LED lighting are many, of which the development of a species-specific light recipe for optimizing plant growth and other desirable traits are promising (Bian *et al*., 2015 and Wei *et al*., 2018).

In research on monochromatic light spectra, the morpho-anatomical, photosynthetic, and secondary metabolism characteristics of plants have been shown to be more significantly affected by monochromatic than multispectral light (Landi *et al*., 2020). Among various species, previous studies have suggested that red light results in the highest quantum yield of CO_2 fixation among the wavelengths in the photosynthetically active spectrum (Hogewoning *et al*., 2012; Wu *et al*., 2019). Plants exhibit some degree of undesirable growth characteristics (lower biomass, lower net photosynthetic rate, or downward leaf curling) under red light alone, but these symptoms are ameliorated when blue light is added (Kong *et al*., 2018; Miao *et al*., 2019). It has been reported that blue-light signaling triggers processes such as photomorphogenesis, stomatal opening, and phototropism, which broadly affect the level of photosynthesis (Horrer *et al*., 2016; Huche-Thelier *et al*., 2016). Blue light enhances the accumulation of carotenoids, flavonoids, and anthocyanins without substantially affecting plant morpho-anatomical traits (Landi *et al*., 2020; Zhang *et al*., 2020), but long-term blue light exposure may affect plant growth and morphology, inducing changes in the photosynthetic apparatus (Huche-Thelier *et al*., 2016). However, red light strongly alters morphology and physiology without showing positive effects on secondary metabolites (Zhang *et al*., 2020). A large number of photophysiological studies have verified the importance of the combination of red and blue light for improving plant growth and nutritional quality compared with than monochromatic light in crops such as lettuce, cucumber, soybean seedlings, and pakchoi (Chen and Yang, 2018; Ma *et al*., 2018; Song *et al*., 2020).

To identify the amounts of supplemental red lights, blue lights, or both, combination of white with different fractions of red LEDs, blue LEDs, or both were examined in sweet basil, strawberry, begonia seedlings, geranium seedlings, petunia seedlings, and snapdragon seedlings (Park and Runkle, 2018; Piovene *et al*., 2015). The LEDs with an R:B ratio of 0.7 improved growth and nutraceutical properties in sweet basil and strawberry (Piovene *et al*., 2015). White LEDs had similar impacts on seedling growth and electric energy consumption of artificial lights compared with mixture of red and blue lights (Park and Runkle, 2018). Chen *et al*. (2016) inferred that lettuce yield would be higher with larger red light fraction when white light was applied as background light.

Therefore, the objectives of this study were to figure out the improvement of plant growth, nutrient and energy use efficiency of Pakchoi under different LED-light spectrum in vertical grow-house created by white, fluorescent, red plus blue light from sole-source LEDs by investigating growth, yield and quality parameters of pakchoi with the following objectives:

1. To find out the effects of LED light on growth of pakchoi
2. To find out the effects of LED light on yield of pakchoi
3. To study the photosynthetic performance with quality of pakchoi under different colored LED light spectrum

CHAPTER II

REVIEW OF LITERATURE

Improvement of plant growth and yield using different LED-light spectrum is a new idea of vegetable farming. Among different LED-light spectrum treatments, pakchoi has been grown to find out the best option using LED-light spectrum. Very limited studies have been performed in this aspects. Some of the recent past information on the improvement of plant growth, nutrient and energy use efficiency of Pakchoi under different LED-light spectrum in vertical farming have been reviewed below:

Light is central for the evolution and sustainability of life on our planet. For plants, light can be a source of energy and an environmental signal. Plants harness light energy from the sun to convert carbon dioxide and water into carbohydrates and release oxygen into the atmosphere. Plants have also evolved many types of photoreceptors to perceive different light qualities, such as wavelength, intensity and duration, to regulate a broad range of developmental and physiological processes. In a study of different intervals of alternating red and blue light, treatment with an interval of 1 h was shown to be beneficial for the accumulation of biomass, sucrose, and starch in lettuce and promoted electric efficiency and light use efficiency (Chen *et al*., 2019). However, under alternating red and blue light with intervals of 2 and 4 h, soluble sugar and ascorbic acid levels were significantly increased, but the nitrate content was decreased (Chen *et al*., 2017).

Vertical grow-house facilitates the production of high value crops with higher yield than obtained from conventional farming by efficient utilization of resources such as water, nutrients, space and time, thereby, reducing carbon footprint (Specht *et al*., 2014). Vertical grow-house technology does not require huge arable land to produce crops and thus is agriculturally independent. This innovation utilizes both the horizontal and vertical spaces more effectively, thereby, producing higher yield per unit volume under controlled environmental conditions of temperature, light, carbon dioxide and humidity. There are different types of vertical grow-house innovations like hydroponics, aeroponics, and aquaponics where the nutrients are effectively utilized and monitored for physical and chemical parameters like quality, pH, and solubility in water. Since vertical grow -house is experimented within a closed and controlled environment, sunlight as a source of light for carrying out photosynthesis is replaced by artificial lights with different spectra and intensities. In such a case, LED lights are more effective with high energy use efficiency and durability than traditional light sources like fluorescent lamps (Specht *et al*., 2014).

There are two lighting methods for exposing plants to red and blue light: the familiar method of simultaneous lighting and the Shigyo Method, the core concept of which is the alternation of red and blue light irradiation (Shimokawa *et al*., 2014). Alternating red and blue light was shown to significantly enhance lettuce growth when the total intensity was the same as that under the simultaneous irradiation with red and blue light each day (Shimokawa *et al*., 2014).

Various species exhibit different light-response modes; nevertheless, more experiments need to be conducted to ensure the application of optimal red and blue light with alternating intervals. The effects of supplementary light on growth and health-promoting compounds in Brassica vegetables have been reported; for example, the spectra and intensity of supplemental LED illumination were associated with the enhanced accumulation of lutein and b-carotene in Brassicaceae microgreens (Brazaityte *et al*., 2015). Both plant growth and the accumulation of health-promoting compounds in Chinese kale and pakchoi were increased in association with the supplemental blue light intensity (Li *et al*., 2019; Zheng *et al*., 2018).

Light is one of the most important environmental factors that influence plant growth and development. Terrestrial sunlight consists of ultraviolet (UV), visible light and infrared radiation, in which visible light accounts for almost half of the absorption spectrum (Abe, 2010). The wavelength of the UV radiation lies in the range of 100–400 nm, visible light in the range of 400–700 nm and infrared in the range of 700–1000 nm. Even though the terrestrial sunlight spectrum is wide, plants can only utilize the visible light spectrum as the sole source of energy for photosynthesis, and this narrow spectrum of electromagnetic radiation is defined as photosynthetically active radiation (PAR) (McCree, 1971, 1972). Interestingly, plants can sense and detect variations in the light intensity and spectral composition of their native environment to adjust their growth and developmental processes (Fiorucci and Fankhauser, 2017). This has given rise to various plant responses such as photomorphogenesis, photoperiodism and phototropism (Kendrick and Kronenberg, 2012; Vince-Prue, 1975; Whippo and Hangarter, 2006). Photomorphogenesis refers to the growth and development of plants. Photoperiodism is the ability of plants to track time. Phototropism enables plants to grow towards or away from a light source (Wong *et al*., 2020).

Plants use photosynthetic pigments in their leaves to capture energy from PAR to drive synthesis of sugar molecules. These photosynthetic pigments are present around the thylakoid membranes of chloroplasts to serve as primary electron donors in the electron transport chain (Anderson, 1986). In plants, the most abundant photosynthetic pigments are chlorophyll a and chlorophyll b (Shoaf and Lium, 1976). The chlorophyll content is determined by mainly two methods, which are the absorption of light of isolated chlorophyll in aqueous acetone and the measurement of leaf reflectance and transmission level using a Soil Plant Analysis Development (SPAD) chlorophyll meter (Netto *et al*., 2005). The approximate absorption maxima of chlorophyll a are at 430 nm and 662 nm and those of chlorophyll b are at 453 and 642 nm (Inskeep and Bloom, 1985). Due to the chemical structures of chlorophyll a and b, the absorption spectra are not uniform across PAR and they have minimal absorption in the 500–600 nm range, thus, reflecting the colors of light green and turquoise, respectively. In some plants, accessory pigments, such as carotenoids (carotenes and xanthophylls), are produced to help absorb light in the blue-green spectrum to enhance photosynthesis (Havaux, 1998).

The development in LED lighting technology, allowing the flexible modifications of light spectra, has enabled the research and application of light quality in enhancing leafy green qualities in controlled environment for better growth, colour, flavour and phytonutrient content. The effects of LED spectra on the growth, development and metabolite accumulation of leafy vegetables have been intensively studied, especially in lettuce. However, it is challenging to extract an optimal lighting recipe from all of this research due to inconsistent experimental parameters ranging from the precise spectral composition to the length of treatment (Wong *et al*., 2020). Studies conducted to understand the different spectral composition on improving biomass and quality of leafy vegetables have focused primarily on red and blue wavelengths, the absorptance maxima of chlorophyll. Red light (RL) has the highest quantum yield, whereas blue light (BL) is considerably less efficient in driving photosynthesis (Inada, 1976; McCree, 1972).

There is a significant loss of BL energy resulting from the absorption by non-photosynthetic pigments, including anthocyanin and accessory photosynthetic pigments that have inefficient energy transfer to chlorophyll (Terashima *et al*., 2009). RL induces many physiological responses including leaf development, stomatal opening, chlorophyll and carbohydrate accumulations (Azad *et al*., 2020; Hogewoning *et al*., 2010; Lee *et al*., 2016; Yang *et al*., 2016). BL influences photosynthetic activity by inducing stomatal opening (Zeiger *et al*., 2002) and affecting chloroplast movement within the cell (Kasahara *et al*., 2002) in the short term while increasing stomata number and leaf thickness in the long term (Hogewoning *et al*., 2010; Wang *et al*., 2016). BL is also known to increase the chlorophyll content (Hogewoning *et al*., 2010; Johkan *et al*., 2010; Matsuda *et al*., 2007). A greater fraction of BL is associated with the development of "sun-type" leaf characterized by a high leaf thickness and photosynthetic capacity (Hogewoning *et al*., 2010; Matsuda *et al*., 2007). BL also regulates several plant morphogenic responses including leaf expansion and shoot elongation (Li and Kubota, 2009; Metallo *et al*., 2018).

Kim *et al*., 2004) conducted an experiment to determine the effects of LEDs on net photosynthetic rate, growth, and leaf stomata of chrysanthemum plantlets *in vitro* grown in MS medium, it was found that the net photosynthetic rate was highest under red (650 nm) and blue (440 nm) combination and lowest under blue-far red (720 nm) combination and blue. Red and red-far red combination resulted in the highest stem elongation but with stem fragility. Shoot growth excluding stem elongation was the greatest under red-blue combination and fluorescent light. When *Lactuca sativa* of variety red curly lettuce was grown under different light spectrum, it was found that anthocyanin synthesis, protein content and phenylalanine ammonia-lyase enzyme activity were highest in combined radiation of blue and red-light treatment (Heo *et al*., 2012). In another study, where red and green basil (*Ocimum basilicum*) microgreens were grown with blue and red LED, it was found that growth of microgreens was enhanced with predominantly blue illumination showing larger cotyledon area and higher fresh mass, enhanced chlorophyll a, and anthocyanin pigments contents. Stimulation of phenolic synthesis and free radical scavenging activity were improved by pre1dominantly red light in the green cultivar and blue light in the red cultivar (Lobiuc *et al*., 2017), which indicates that LED light has an influence on the colour of the leaf.

A combination of red and blue LEDs routinely used in indoor farming has a relatively higher production efficiency compared to other light sources such as fluorescent lamps with the same light irradiance (Amoozgar *et al*., 2017; Johkan *et al*., 2010; Lee *et al*., 2016). The optimal ratio between BL and RL is crucial in determining plant productivity and a low B/R ratio generally favours biomass accumulation. In a background of RL, 25% of BL (B25R75) has produced red pakchoi with greater biomass, leaf area and anthocyanin accumulation compared to white LED (Mickens *et al.*, 2019), while lettuce (‘Grizzly’) grown under B30R70 has 75% increase in biomass when compared to those under white LED (Amoozgar *et al.*, 2017). Kale grown under B20R80 increases by 12.5% in dry mass compared to those under white light and shows significant morphological alteration with shorter and more compact plants, which is consistent with BL’s roles in inhibiting extension growth (Metallo *et al.*, 2018). The above-ground dry weight is almost doubled in tatsoi when RL is supplemented with 10% BL (Virsile *et al.*, 2019). In Chinese kale, a 2–3 fold increase in leaf area and shoot dry weight has been reported when RL was supplemented with 18% BL (He *et al.*, 2015).

Monochromatic RL environment, however, triggers abnormal plant morphology in lettuce, spinach, kale and basil that includes the development of elongated hypocotyl, long petioles and thin wide leaves with reduced chlorophyll content (Amoozgar *et al.*, 2017; Johkan *et al.*, 2010; Kang *et al.*, 2016; Naznin *et al.*, 2019). The impaired development seen in these studies resembles those associated with shade avoidance response that is triggered by low light, a high red to far-red (R/Fr) ratio, low BL or high green light levels in the environment (Wang *et al.*, 2020).

Too much BL in the irradiance can also have an adverse effect on plant growth and development. When the proportion of blue LED exceeds 11% in a broad-spectrum background, dry mass and leaf area are decreased for lettuce (‘Waldmann’s Green’), radish and pepper (Cope *et al.*, 2014). Similar parameters in two other cultivars of lettuce (‘Rouxai’ and ‘Green Skirt’) as well as kale are also negatively correlated with the amount of BL in a red background (Dou *et al.*, 2020; Kang *et al.*, 2016; Meng *et al.*, 2020). An increase from 16 to 24% of BL leads to the reduction in the leaf area and dry weight of Chinese kale grown in a red background (He *et al*., 2015).

A lower level of irradiance does not seem to negate the effect of high fraction of BL on lettuce ('Lollo Rosso') growth as the leaf area and shoot dry weight decrease significantly as BL fraction is increased from 25% to 50% at 90 μmol $m^{-2}s^{-1}$ (equivalent to 5% of full sunlight) (Azad *et al*., 2020). Spinach seems to be more vulnerable to BL-induced negative effects compared to lettuce and komatsuna (*Brassica rapa* var. perviridis) as severe reduction in dry weight has been observed under blue fluorescent lamp (Ohashi-Kaneko *et al*., 2007).

Green light (GL), unlike RL and BL, can penetrate deeper into a leaf (Sun *et al*., 1998) and canopy (Massa *et al*., 2015). A higher fraction of GL may thus stimulate photosynthesis deep within a leave and canopy layer, increasing whole-plant photosynthesis. Such increase in photosynthesis is not possible with excess RL or BL due to their strong absorption by chlorophyll in the upper part of the leaf (Terashima *et al*., 2009). This unique contribution of GL to photosynthesis suggests that it may be beneficial to plant growth and development when used to supplement red and blue irradiation, especially for vegetables that form thick canopy. GL can also act as a shade signal (Sellaro *et al*., 2010; Zhang *et al*., 2011) and it can antagonize a number of BL-induced responses including inhibition of extension growth (Folta, 2004) and stimulation of stomata opening (Talbott *et al*., 2002).

There is a considerable lack of studies on the effect of supplemental GL on leafy vegetables species other than lettuce, and even these are inconclusive regarding the physiological benefits of GL. A blue and red background (B16R84) with 24% GL from green fluorescent lamp (B15G24R61) has been shown to increase lettuce ('Waldmann's Green') yield (Kim *et al*., 2004), whereas in another study, an increase of up to 30% green LED light does not influence the dry mass in the same cultivar (Snowden *et al*., 2016). This discrepancy may be attributed to the different sources of GL used since fluorescent light could confound the effect of GL with associated increases in diffused light and leaf temperature (Snowden *et al*., 2016). The effect of GL on growth has also been examined at different growth stages. For example, the biomass and shoot diameter of lettuce (‘Outredgeous’) are significantly increased at 14- and 21-day-after-sowing (DAS) but such effects are no longer significant at maturity (28 DAS) compared to those grown under white LED, suggesting that GL stimulates an early rapid growth of lettuce (Mickens *et al*., 2018). Similarly, there is significant increase of leaf area in lettuce (‘Red Cos’) seedlings (3-w-old) with the inclusion of GL in the blue and red irradiance compared to those grown under blue and red only, but this fails to translate to an increase in biomass for mature plants (5-w-old) (Samuoliene *et al*., 2020). These imply that supplemental GL favours growth at seedling stage but as the plant grows and matures, it becomes less sensitive to the growth-promoting effect of GL. In addition, a negative effect of GL (33.3%) on growth of lettuce (‘Rouxai’) has been found to be dependent on the high BL fraction (33.3%) since lower BL fraction (11.1% or 0%) diminishes such effect (Meng *et al*., 2020). This inhibitory effect of GL on growth that is dependent on high BL fraction has also been reported in kale (Dou *et al*., 2020). Thus, the interaction between GL and BL further adds to the complexity in delineating the GL effect quantitatively. Nonetheless, the inclusion of GL is helpful in the dichromatic background of red and blue in a closed production system as it can improve the visual quality by creating a more pleasant white light output that is less harsh on human eyes, making crop inspection more straightforward (Massa *et al*., 2008).

Yellow light (YL, 580–600 nm) is poorly absorbed by any of the photosynthetic pigments and we know relatively little about the effect of YL on photosynthesis as compared to other wavelengths. Research using green fluorescent lamp includes 500–600 nm wavelengths and therefore contains YL (Dougher and Bugbee, 2001; Kim *et al*., 2004). These coupled with the low efficiency of yellow LEDs (Jiang *et al*., 2019) could explain the scarcity of studies exploring the use of YL in the cultivation of leafy greens. Yellow wavelength (ranging from 20 to 30% of photosynthetic photon flux density (PPFD) = 200 and 500 μmol $m^{-2}s^{-1}$) from high pressure sodium lamp and metal halide lamp has been reported to inhibit growth in lettuce (‘Grand Rapids’) by suppressing chlorophyll formation (Dougher and Bugbee, 2001), while a more recent study shows minimal effect of YL (6.7% of PPFD = 300 μmol $m^{-2}s^{-1}$) on lettuce (‘Red and Green Cos’) growth and biomass production (Virsile *et al*., 2020). In another cultivar (‘Green Oakleaf’), supplemental YL (30% of PPFD = 135 μmol $m^{-2}s^{-1}$) inhibits growth compared to those under white LED (Chen *et al*., 2016). Thus, YL negatively impacts on lettuce growth at high fraction. The effect of YL on the growth of other leafy greens as well as on the accumulation of phytonutrients await further investigation. Such studies will be facilitated with the recent development of a more efficient light source (Jiang *et al*., 2019).

Plants have varied morphological and physiological responses to specific light spectrum, and the current advancement of LEDs enables one to tailor the spectrum to obtain favorable plant growth or nutritional values (Mickens *et al*., 2018; Park and Runkle, 2018). Compared with other wavelengths, red and blue lights were paid more attention to lettuce (Lee *et al*., 2010; Son and Oh, 2015; Stutte *et al*., 2009; Wang *et al*., 2016b), cucumber seedlings (Hernandez and Kubota, 2016), tomato seedlings (Hernandez *et al*., 2016; Liu *et al*., 2018), Mesembryanthemum crystallinum (He *et al*., 2017), and sweet basil (Pennisi *et al*., 2019) in past and recent decades, as red and blue lights were considered more effectively absorbed by chlorophylls of plant leaves (McCree, 1971). Moreover, plants grown under the combination of red and blue lights had bigger stem diameter (Yang *et al*., 2018), higher net photosynthetic rate (Pn) (Wang *et al*., 2016b), anthocyanin (Lee *et al*., 2010; Stutte *et al*., 2009), and arginine (Zhang *et al*., 2018a) compared with those grown under monochromatic red light. In addition, red and blue LEDs had higher photosynthetic photon efficiency (Park and Runkle, 2018). As a result, red and blue LEDs are commonly applied for commercial production in PFALs. Previous studies also investigated the suitable R:B ratio created by mixture of red and blue lights for dry weight accumulation in lettuce (Wang *et al*., 2016b), cucumber seedlings (Hernandez and Kubota, 2016), and M. crystallinum (He *et al*., 2017).

Plants grown under mixed red and blue LEDs appeared purplish, thus making visual assessment of disease symptoms and growth disorder difficult to observe (Kim *et al*., 2004). One solution for this limitation was to apply white LEDs, alone or with blue LEDs, red LEDs, or both (Park and Runkle, 2018; Yan *et al*., 2019). The world's first white LED was created in 1996 as a phosphor coating used by blue LED (Bourget, 2008) and the present white LEDs showed increased energy efficiency as the highly efficient blue-emitting diodes were invented (Pust *et al*., 2015). White LEDs were suggested as a substitute lighting sources compared with fluorescent lamps for lettuce production in PFALs (Park *et al*., 2012), and green leaf lettuce grown under white LEDs had higher leaf fresh weight (Zhang *et al*., 2015), root fresh weight, and phenolic concentration (Son *et al*., 2012) than those grown under monochromatic red LEDs. Moreover, white LEDs were used as background lighting in 'Green Oak Leaf' lettuce reported by Chen *et al*. (2016), and the effects of red, green, blue, yellow, and far red lights as supplemental lights were examined. Results indicated that lettuces grown under white light combined with supplemental red light appeared vigorous and compact compared with those grown under white LEDs alone. Mickens *et al*. (2018) also observed similar results in red romaine lettuce at harvest. To identify the amounts of supplemental red lights, blue lights, or both, combination of white with different fractions of red LEDs, blue LEDs, or both were examined in sweet basil, strawberry, begonia seedlings, geranium seedlings, petunia seedlings, and snapdragon seedlings (Park and Runkle, 2018; Piovene *et al*., 2015).

Daily light integral is the total amount of light received by plants in 1 d (Bruggink and Heuvelink, 1987; Kozai *et al*., 2018). Linear relationships were often observed between DLI and biomass of lettuce (Gent, 2014; Zhang *et al*., 2018b), average shoot dry weight per average internode number of Celosia seedlings (Pramuk and Runkle, 2005), and nutritional values of sweet basil (Dou *et al*., 2018). The suitable DLIs for hydroponic lettuce production were recommended at 12.67 mol $m^{-2}d^{-1}$ (Fu *et al*., 2017) and 14.40 mol $m^{-2}d^{-1}$ (Zhang *et al*., 2018b), respectively. However, few studies examined the relationships between DLI and carbohydrate accumlation under LEDs with different R:B ratios when white LEDs were used as base lighting source, and there were few bases for the spectrum design of relatively broad and wide LEDs for lettuce cultivation. In addition, few researchers focused on the energy use efficiency for lettuce production in PFALs.

In the commercial greenhouse, light supplementation using artificial light can significantly increase crop yield and nutrition quality, especially in low-light-intensity seasons like winter and late autumn (Lu *et al*., 2012; Yorio *et al*., 2001). With the development of urban agriculture, artificial light has become the most important way to control the light conditions. For a long time, people were using fluorescent lamps, filament lamps and high-pressure sodium lamps (HPL), and much research was carried out to test their effects (Tibbitts *et al*., 1983). However, these kinds of light tend to consume large amounts of electrical energy and release a lot of heat (which will also increase the cooling system cost), and their spectra are not very suitable for plants, which leads to excessive waste of energy (Randall and Lopez, 2014).

The most important element in controlling artificial farming costs is supplying light for photosynthesis and growth by light sources with high photoelectric efficiency. Light-emitting diodes (LEDs) have been proposed as alternative light sources in controlled agricultural environments since, compared with traditional horticulture light sources (e.g., HPL), LEDs have drastic advantages, such as superior lifetime, reduced size, cooler emitting temperature, and reduced energy consumption (Massa *et al*., 2008).

An exciting potential of using LED lighting is the development of species-specific light recipes comprising the optimum proportion of specific narrow-band wavelength light that can optimize plant growth, development and other desirable traits (e.g., increase phytochemical content) (Bian *et al*., 2015, 2016), whilst significantly reducing the energy input compared with traditionally used horticulture light sources. Recently, the effect of LEDs on plant growth and development has aroused increasing interest. However, the results of related studies are sometimes different, and even contradictory (Avercheva *et al*., 2009; Bian *et al*., 2016; Hogewoning *et al*., 2010; Urbonaviciute *et al*., 2007).

Plants grown in vertical grow-house systems are surrounded by walls and receive no sunlight; therefore, artificial lamps provide the only light source. However, conventional light sources have drawbacks to their use, causing excessive heat on leaf surfaces and leading to undesirable effects on plant growth (Martineau *et al*., 2012). Thus, there is a need for the development of innovative artificial lighting for optimizing the light environment. Light-emitting diodes (LEDs) have seen great development with many technological advancements and have incomparable advantages, and their application to lighting in plant cultivation has increased rapidly (Xu, 2019). The price of LEDs has decreased remarkably over the past several years, and many studies have been concentrated on defining the optimal light environments to enable the high-quality, high-speed production of various plant species (Massa *et al*., 2008). This makes LED lighting systems a cost-effective solution for controlled environment agriculture systems.

Natural sunlight contains a wide continuum of wavelength and fluence and is optimal for plants (Darko *et al*., 2014). Therefore, manipulating the light conditions of artificial light sources is essential for growing plants in vertical farming to obtain electricity cost savings and balance the yield and quality of plants (Dou *et al*., 2018). It is well documented that the various regions of light spectra have different efficiencies in enhancing the plant photosynthetic process and plant morphological, physiological, and biochemical responses (Folta and Maruhnich, 2007). Within the visible light spectral range (400–700 nm), many researchers have focused on studying the role of red (R) (600–700 nm) and blue (B) (400–500 nm) light and on defining their optimal combination ratios because their wavelengths are close to the absorbance of photosynthetic pigments that effectively drive photosynthesis (Sabzalian *et al*., 2014). Many studies have confirmed the role of R LEDs in increased biomass accumulation, stem elongation, and leaf expansion, as well as the effect of B LEDs in chlorophyll production, stomata opening, and photosynthesis (Muneer *et al*., 2014). Therefore, monochromatic R or B LEDs and combined RB LEDs have been widely used in scientific research and commercial vertical farming (Ouzounis *et al*., 2015). However, plants exposed to combination R and B lights normally appear purplish-grey to the human eye, which leads to difficulties in the visual assessment of plant health (e.g., disease symptoms, nutritional deficiencies, and physiological disorders) (Kim *et al*., 2005). The addition of green (G) (500–600 nm) light is considered a possible solution to this limitation. It is reported that G light has little impact on plant photosynthesis and photomorphogenesis, but it has a greater ability to penetrate the folded layers of leaves and the lower canopy, which can increase photosynthesis in the lower parts of leaves as well as carbon assimilation (Smith *et al*., 2017). However, supplementary G LED light is not widely applied in practical plant cultivation due to the inefficiency in converting electricity into photons. Hence, another strategy is the application of white (W) light that contains G light.

Advanced LED technology enables broad-spectrum W LED light that consists of R, G, and B lights. This could be effective for use in vertical grow-house to improve plant growth and provide desirable lighting for human vision. Several approaches have been used to achieve W LED light. The most common and successful approach is the use of a B LED chip with phosphors to convert a part of the B light to R and G lights. The B light from the LED chip and the R and G lights converted by phosphors create W light, leading to the steady increase in the efficiency of B LEDs and consequently improving the W LED efficiency (Cope and Bugbee, 2013). W LED light can also be created by combining several LED chips that emit monochromatic R, B, and G lights (Chang *et al*., 2012), enabling control of the ratios of R, B, and G lights desirable for human vision and plant growth responses. This approach has become feasible with the highly efficient use of B and G LEDs and possesses high reliability and durability as well as low energy consumption (Pimputkar *et al*., 2009).

Different crops e.g. lettuce crops grown with red and blue LED lighting (95% red and 5% blue) used 50% less energy per unit dry biomass accumulated than under traditional light sources, which indicates that the significant reduction in energy consumption for plant-growth by using LED than traditional light sources (Poulet *et al*., 2014). In an experiment on the indoor cultivation of basil and strawberry, it was found that the plants expressed increased biomass, fruit yield, antioxidant content and reduced nitrate content when treated with LED with highest energy use efficiency than traditional fluorescent lamps and spectral red: blue ratio of 0.7 was essential for proper plant growth with improved nutraceutical properties (Piovene *et al*., 2015).

Huang *et al*. (2021) conducted a study aimed to evaluate the effects of alternating red (660 nm) and blue (460 nm) light on the growth and nutritional quality of two-leaf-color pakchoi (*Brassica campestris* L. ssp. *chinensis* var. communis). Four light treatments (supplemental alternating red and blue light with intervals of 0, 1, 2, and 4 hours, with a monochromatic light intensity of 100 µmol $m^{-2}s^{-1}$ and a cumulative lighting time of 16 hours per day) were conducted in a greenhouse under identical ambient light conditions (90 to 120 µmol m^{-2} s^{-1} at 12:00 AM) for 10 days before green- and red-leaf pakchoi were harvested. The results showed that the two-leafcolor pakchoi receiving alternating red and blue light exhibited more compact canopies and wider leaves than those under the control treatment, which was attributed to the shade avoidance syndrome of plants. The trends of both biomass and the soluble sugar content were highest under the 1-hour treatment. The contents of chlorophyll a and total chlorophyll in both cultivars (green- and red-leaf pakchoi) were significantly increased compared with control, without significant differences among the 1-, 2-, and 4-hour treatments, whereas chlorophyll b exhibited no significant difference in any treatment. Alternating red- and blue-light treatment significantly affected the carotenoid content, but different trends in green and red-leaf pakchoi were observed, with the highest contents being detected under the 1-hour and 4-hour treatments, respectively. Compared with 0 hours, the contents of vitamin C, phenolic compounds, flavonoids, and anthocyanins in two-leaf-color pakchoi were significantly increased, but no significant differences were observed in vitamin C, phenolic compounds, and flavonoids among the 1-, 2-, and 4-hour treatments, similar to what was found for the anthocyanin content of green-leaf pakchoi. Taken together, treatment with an interval of 1 hour was the most effective for increasing the biomass of pakchoi in this study, but treatment with a 4-hour interval should be considered to enhance the accumulation of health-promoting compounds.

Nguyen *et al*. (2021) carried out a study aimed to examine the effect of W LED light sources on the growth and quality of butterhead and romaine lettuce and reported that white (W) light-emitting diode (LED) light has been used as an efficient light source for commercial plant cultivation in vertical farming. Three W LED light sources including normal W light (NWL) which has 450 nm as its pumping wavelength and two specific W lights (SWL1 and SWL2) with shorter blue peak wavelength (437 nm) were used to grow lettuce in comparison to a red (R) and blue (B) LED combination. As a result, SWL1 and SWL2 treatments with the same electrical power or photosynthetic photon flux density (PPFD) resulted in more growth of both lettuce cultivars compared to RB treatment. Some phenolic and flavonol contents were increased in the RB treatment, whereas SWL2 treatment stimulated the accumulation of other phenolic and flavonol compounds. Meanwhile, neither NWL nor SWL1 treatments increased the individual phenolic and flavonol contents in either cultivar (except for some flavonols in romaine lettuce in the SWL1 group). In addition, light and energy use efficiencies were also highest in the SWL1 and SWL2 treatments. These results illustrate the positive effects of specific W LED light on lettuce growth and quality, and suggest that the specific W LED light sources, especially SWL2, could be preferably used in vertical farming.

Agricultural production in controlled vertical grow-house offers a reliable alternative to food and nutrition supply for densely populated cities and contributes to addressing the impending food insecurity. Leafy vegetables, rich in vitamins, minerals, fibres and antioxidants, account for over half of the indoor farming operations worldwide. Light is the foremost environmental factor for plant growth and development, and the success of vertical grow-house largely depends on lighting qualities. The energy efficient light-emitting diode (LED) has been increasingly used in indoor vertical grow-house systems. A study was carried out by Wong *et al.* (2020) for seeing the lights for leafy greens in indoor vertical grow-house and was provided an updated overview of the current indoor vertical grow-house systems, the mechanisms of light perception by photoreceptors, and the effects of LED spectra or intensity on growth and phytonutrient accumulation of leafy greens for this study. It was reported that by Wong *et al.* (2020) that lighting quality and quantity can be manipulated to improve yield and phytonutrient contents of leafy greens. As responses of leafy greens to light are dependent on genotype and developmental stage, light recipe targeting different developmental stages should be formulated for different species for maximizing yield. While it has been known that blue wavelength has a more prominent positive impact on phytonutrient accumulation than red, little is known for other wavelengths.

Moreover, recent findings that green wavelength inhibits plant growth in a blue-wavelength-dependent manner highlight the need for future research to investigate interactive effects of different wavelengths on modulating plant growth and metabolism.

Yan *et al*. (2020) conducted an experiment in order to examine the effects of addition light added in red plus blue LEDs or white LEDs, green and purple leaf lettuces (Lactuca sativa L. cv. Lvdie and Ziya) were hydroponically cultivated for 20 days under white LEDs, white plus red LEDs, red plus blue LEDs, and red plus blue LEDs supplemented with ultraviolet, green or far-red light, respectively. The results indicated that the addition of far-red light in red plus blue LEDs increased leaf fresh and dry weights of green leaf lettuce by 28% and 34%, respectively. Spectral absorbencies of purple leaf lettuce grown under red plus blue LEDs supplemented with green light were lower in green light region compared with those grown under red plus blue LEDs, which was associated with anthocyanin contents. White plus red LEDs significantly increased leaf fresh and dry weights of purple leaf lettuce by 25%, and no significant differences were observed in vitamin C and nitrate contents compared with white LEDs. Fresh weight, light and electrical energy use efficiencies of hydroponic green and purple leaf lettuces grown under white plus red LEDs were higher or no significant differences compared with those grown under red plus blue LEDs.

Yan and He (2019) conducted a study to investigate the effects of daily light integrals (DLI) and light emitting diodes (LEDs) light quality (LQ) on growth, nutritional quality, and energy use efficiency of hydroponic lettuce (*Lactuca sativa* L.) in a plant factory with artificial lighting (PFAL). Hydroponic lettuce plants (cv. Ziwei) were grown for 20 days under 20 combinations of five levels of DLIs at 5.04, 7.56, 10.08, 12.60, and 15.12 mol m^{-2} d^{-1} and four LQs: two kinds of white LEDs with red to blue ratio (R:B ratio) of 0.9 and 1.8, and two white LEDs plus red chips with R:B ratio of 2.7 and 3.6, respectively. Results showed that leaf and root weights and power consumption based on fresh and dry weights increased linearly with increasing DLI, and light and electrical energy use efficiency (LUE and EUE) decreased linearly as DLI increased. However, no statistically significant differences were found in leaf fresh and dry weights and nitrate and vitamin C contents between DLI at 12.60 and 15.12 mol $m^{-2}d^{-1}$. White plus red LEDs with an R:B ratio of 2.7 resulted in higher leaf fresh weight than the two white LEDs. LUE increased by more than 20% when red light fraction increased from 24.2% to 48.6%.

Manawasinghe and Weerasekara (2020) carried out a research and examined the plant growth and yield of vertically grown pakchoi (*Brassica rapa* var. *chinensis*) (in nutrient film technique (NFT) culture) under supplementary lighting with two different combinations of blue to red color LEDs (1:9 and 1:2 ratios) in comparison with horticulture grade and non-horticulture grade (recommended for general use) white (full spectrum) LED while keeping sunlight as the control treatment. Meanwhile NFT culture was compared to plant growth, yield and nitrate accumulation of basil (*Ocimum basilicum* L.) in comparison with conventional soil, culture and compost mixed cocopeat substrate in a replicated trial, conducted under greenhouse conditions with intensive micro climate control. A significantly high vegetative growth and total to yield could be found in the NFT grown basil. The nitrate accumulation in basil leaves was well below the maximum permissible limit (MPL), set-fourth by the recommendations of the European Health Commission. Meanwhile, the highest overall leaf quality of pakchoi was achieved by the normal LEDs. Horticulture graded to LED maintained fairly high chlorophyll a and b contents contributing to its characteristic leaf color.

Bian *et al*. (2017) carried out a study and found that light supplementation can increase crop yield in greenhouses by promoting photosynthesis and plant growth. However, the high energy costs associated with light supplementation are a predominant factor that limits development and profit improvement of controlled environment agriculture. Light-emitting diodes (LEDs) are a promising technology that has tremendous potential to improve irradiance efficiency and to replace traditionally used horticultural lighting. Compared with traditional light sources (e.g., high-pressure sodium lamps and metal halide lamps) used in crop production, LEDs have distinct advantages, such as their small size, long lifetime and high photoelectric conversion efficiency. The results showed that the highest fresh and dry weight and leaf area were observed under red and blue LED light, with the blue light percentage at 23%. Compared with fluorescent lamps (FL) with photosynthetic photon flux density (PPFD) at 220 μmol m^{-2} s^{-1}, the light-use efficiency increased by 55, 114 and 115% for mixed red and blue LEDs with PPFD at 100, 150 and 220 μmol m^{-2} s^{-1}, respectively. Monochromatic red and blue light LEDs resulted in significant decreases in Pn of tomato plants, but the stomatal conductance (Gs) for monochromatic blue LEDs was higher than that for FL. The effect of light spectrum composition on lettuce nutrition quality was also studied. Continuous light with combined red, green and blue LEDs exhibited a remarkable decrease in nitrate. Moreover, continuous LED light for 24 h significantly increased phenolic compound content and free-radical scavenging capacity in lettuce leaf.

CHAPTER III

MATERIALS AND METHODS

The present research work was conducted at the roof top (indoor structure) of Dr. M. Wazed Mia Central Laboratory, Sher-e-Bangla Agricultural University, Dhaka during the period from November 2020 to December 2020. Brief descriptions of materials and methods that are used in carrying out the experiment have been presented in this chapter.

3.1 Experimental site

The experimental site is located at 90°22′E longitude and 23°41′N latitude at an altitude of 8.6 meters above the sea level. The experimental site is presented in (Appendix I).

3.2 Climatic condition

The experimental area is under the sub-tropical climate that is characterized by less rainfall associated with moderately low temperature during rabi season, (October-March) and high temperature, high humidity and heavy rainfall with occasional gusty winds during kharif season (April-September). Details of weather data in respect of temperature (^{0}C), rainfall (mm) and relative humidity (%) for the study period was collected from Bangladesh Meteorological Department, Agargoan, Dhaka-1207 and presented in Appendix II.

3.3 Planting material used for the experiment

Seeds of the pakchoi variety BARI China Shak-1 was used for the experiment. The seeds were collected from Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur, Bangladesh.

3.4 Treatments of the experiment

One factor experiment consisting six treatments is as follows:

1. T_{FL-T5} = Fluorescent light (control) – Full (L_6 Worm)
2. T_W = White – Full (L_5)
3. T_R = Red – Full (L_4)
4. T_B = Blue – Full (L_3)
5. T_{RB} = Red and Blue - 4:1 (L_2)
6. T_{RBG} = Red, Blue with Green - 4:1:1 (L_1)

- The full light was measured using PPFD (Photosynthetic Photon Flux Density) for single light treatment of different LED light which was maintained by 115 µmol/m^2/s by quantum flux PAR meter and duration was 16 hrs per day maintained by timer started at 6:00 am to 10:00 pm. All this LED light treatment were set up in several rakes vertically where each rake treated with any one of this treatment.

- Fluorescent light was light yellow and it was used as control treatment as an alternative of sunlight.

- Ratio of 4:1 Red and Blue light was calculated by defining the relative areas of the spectrum within the Red and the Blue regions. For each rake, total 15 LED lights were used, among them 12 were Red and 3 were Blue (4:1). The total PPFD for Red and Blue LED light was 115 µmol/m^2/s and duration was 16 hrs per day.

- Ratio of 4:1:1 Red, Blue and Green light were calculated by defining the relative areas of the spectrum within the Red, Blue and Green regions. For each rake, total 15 LED lights were used, among them 10 were Red, 2.5(2) was Blue and 2.5(2) was Red (4:1:1). The total PPFD for Red, Blue and Green LED light was 115 µmol/m^2/s and duration was 16 hrs per day.

- Each treatment of the experiment was set up vertically in 6 racks and each rake contained three boxes. Each box was considered as one replication and five plants were transplanted in each box. The size of the unit box was 25 inch ×10 inch.

3.5 Design and layout of the experiments

The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications. One factor (LED light treatments) was considered for the present study. The layout of experiment is presented in Appendix III.

3.6 Details of the experimental operations

The particulars of the experimental operations carried out during the experiment are presented below:

3.6.1 Seed sowing and growing of seedlings

For germination, at first seeds of BARI China Shak-1 (Pakchoi) was taken into a bowl full of water to select viable seed that laying on the bottom of bowl. To breakdown seed coat and to help germination all the selected seeds were taken into water for 4 hrs. On the other side, germination media and the indoor room by controlling temperature and humidity were prepared.

Rock wool Grow Cubes Starter Sheets as germination media cutting was used in order to the area of clear germination box with small cube shape unit. By placing the sheets into the clear box, germination media is prepared. Then sowing 2/3 seeds per pit on the Rock wool Grow cubes Starter Sheets .Then these sheets were covered with some heavy materials (one box upon another) to warm the area which easily doing germination. All of these boxes were taken under warm room condition. Some water was also offered to keep the media moisturized. Moisture condition was checked by pressing the media by finger.

3.6.2 Preparation of growing condition/frame

After 24 hrs, the seedlings were emerged. To go into a matured seedling stage and also for hardening purposes the seedlings were kept in this germination media under warm condition. At the same time, the final media such as transplanting media was started to make. As this research is totally based on Hydroponic solution and it will be performed in indoor, several racks (6) were prepared in vertical condition and hydroponic solution prepared was used by HRC BARI.

In each racks, 3 solution box (25L) as 3 replications were placed. For lighting purposes, different colored LED was used light which acted as treatment. Lighting structured on the roof of each rack was placed. To maintain PPFD, 115 µmol/m^2/s was tried. To maintain temperature and humidity respectively at 18-20°C and 60%-80% were also tried. Lighting duration was 16 hrs/day.

Plate 1. Pakchoi seedlings after germination

3.6.3 Transplanting the seedlings

After 7 days of germination, the seedlings were ready to transplant. Individual cubes into several net pots were placed in such a way so that hydroponic solution can soak the roak wool which helps supply nutrient to the plant. In this way, all the seedlings were prepared. On the other side, after arranging the entire box in racks, all of them were filled with 25L hydroponic solution. To provide oxygen and to create circulation into the solution, electric motor and some air stoned were used which was placed one for one box. A white partex board was used over the solution box to hold the net pot with seedlings. Each board contained 5 net pots. Placing all the net pots over the solution, transplanting was done. Three electric ventilation fans were used for each rack.

3.6.4 Harvesting

After 35 days of transplanting all of the seedlings were ready to harvest. After harvesting the fresh pakchoi were kept in a cool place to avoid heat stress.

3.7 Recording of data

Then, every one week, the physical data were taken for one day but environmental data was taken for one day interval. The solution level was checked regular basis either plant's roots get solution or not. Data were collected on the following parameters

- **A. Growth parameters**
  1) Plant height (cm)
  2) Number of leaves plant^{-1}
  3) Leaf length (cm)
  4) Leaf breadth (cm)
- **B. Yield contributing parameters and yield**
  1) Stem diameter (cm)
  2) Leaf base breadth
  3) Whole plant diameter plant^{-1} (cm)

4) Fresh weight plant^{-1} (g)
5) Total yield (kg)

C. Quality parameters

1) SPAD value
2) Vitamin-C content (mg/100g)
3) Brix value (°Bx)
4) Moisture content (%)

3.8 Procedure of recording data

3.8.1 Plant height (cm)

The height of the plants was measured of each replication after 7 days of transplanting to at 35 DAT with 7 days interval. The height was measured in centimeter (cm) from the ground level to the tip of the longest leaf and average height was calculated in centimeter.

3.8.2 Number of leaves plant^{-1}

Number of leaves plant^{-1} was calculated from each replication and mean was recorded. Number of leaves plant^{-1} was measured from each replication after 7 to 35 DAT with 7 days interval.

3.8.3 Leaf length (cm)

Leaf length was measured by using a meter scale. The measurement was taken from base of leaf to tip of the petiole and average length of leaves was recorded from plants for each replication. Data was recorded at 7 to 35 DAT with 7 days interval. Mean was expressed in centimeter (cm).

3.8.4 Leaf breadth (cm)

Leaf breadth was recorded as the average of five leaves selected at random from the plant of each replication at 7 - 35 DAT. Thus mean was recorded and expressed in centimeter (cm).

3.8.5 Stem base diameter (cm)

The diameter of stem base in centimeter (cm) was recorded from each plant of each replication at the time of harvest (at 35 DAT) at the base portion of the plant with slide calipers. The average value is termed as stem diameter.

3.8.6 Leaf base breadth (cm)

Leaf base breadth was recorded as the average of five leaves selected at random from the plant of each replication at harvest. Thus mean was recorded and expressed in centimeter (cm).

3.8.7 Whole plant diameter plant^{-1} (cm)

Whole plant diameter plant^{-1} was recorded as the average of five plants of each replication at harvest. Thus mean was recorded and expressed in centimeter (cm).

3.8.8 Fresh weight plant^{-1} (g)

At the time of harvest, whole plant weight of each replication was taken after removing roots and other stables from the plants and then mean was recorded and expressed in gram (g).

3.8.9 SPAD value

SPAD value was measured from pakchoi leaves from each replication with the help of SPAD meter.

3.8.10 Vitamin-C content (mg/100g)

Vitamin C concentration in leaf sample of pakchoi was determine by the method of redox titration using iodine.

3.8.11 Brix value (°Bx)

Brix (symbol °Bx) is a measure of sugars, vitamins, minerals, proteins and other solid content in plant extract. Brix is determined by extracting the juices of a plant and using light refraction to determine the density of the sugars and other dissolved solid content.

3.8.12 Moisture content (%)

To measure the moisture content, 100 g fresh pakchoi was taken and was oven dried and data was recorded. Percent moisture content was measured by the following formula:

$$\% \text{ moisture content} = \frac{\text{Oven dry weight (g)}}{\text{Fresh weight (g)}} \times 100$$

Plate 2. Sample data collection

3.9 Statistical analysis

The collected data on various parameters under study were statistically analyzed using Statistix10 computer package programme. The means for all the treatments were calculated and analysis of variance for all the characters was performed by the F- variance test (Gomez and Gomez, 1984). Significance of difference between means was evaluated by Least Significance Difference (LSD) and the probability level 5% and 1% for the interpretation of results.

CHAPTER IV

RESULTS AND DISCUSSION

The experiment was conducted to find out the Improvement of plant growth, nutrient and energy use efficiency of pakchoi under different LED-light spectrum in vertical grow-house. Data on different growth, yield contributing parameters, yield and quality parameters of pakchoi were recorded. The results have been presented and discusses with the help of table and graphs and possible interpretations given under the following headings:

4.1 Growth parameters

4.1.1 Plant height

Significant variation was found for plant height of pakchoi at different growth stages as influenced by different LED-light spectral ratios (Figure 1 and Appendix IV). Results exhibited that the highest plant height at 7 DAT (6.43 cm) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was significantly different from other treatments followed by control treatment T_{FL-T5} (Fluorescent light - Full) and T_B (Blue - Full) whereas the lowest plant height (3.39 cm) was found from the treatment T_R (Red - Full) which was statistically identical with T_{RBG} (Red, Blue with Green - 4:1:1). Similar trend was found for plant height at 14, 21, 28 and 35 DAT. At 14 DAT, the highest plant height (15.50 cm) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was statistically identical with control treatment T_{FL-T5} (Fluorescent light - Full), T_W (White - Full) and T_B (Blue - Full) whereas the lowest plant height (10.07 cm) was found from the treatment T_R (Red - Full) which was statistically identical with T_{RBG} (Red, Blue with Green - 4:1:1). At 21 DAT, T_{RB} (Red and Blue - 4:1) gave the highest plant height (19.85 cm) which was statistically identical with control treatment T_{FL-T5} (Fluorescent light - Full), T_W (White - Full) and T_B (Blue - Full) whereas T_R (Red - Full) showed the lowest plant height (14.41 cm) which was statistically identical with T_{RBG} (Red, Blue with Green - 4:1:1). Similarly, at 28 DAT, treatment T_{RB} (Red and Blue - 4:1) showed the highest plant height (21.75 cm) which was statistically identical with T_{FL-T5} (Fluorescent light – Full; control), T_W (White - Full) and T_B (Blue - Full) whereas T_R (Red - Full) showed the lowest plant height (15.81 cm) which was statistically identical with T_{RBG} (Red, Blue with Green - 4:1:1). Again, at 35 DAT the highest plant height (24.41 cm) was found from the treatment T_{RB} (Red and Blue - 4:1) which was significantly similar with T_W (White - Full) followed by T_{FL-T5} (Fluorescent light – Full; control) and T_B (Blue - Full) whereas the lowest plant height (17.68 cm) was found from the treatment T_R (Red - Full) which was statistically identical with T_{RBG} (Red, Blue with Green - 4:1:1).

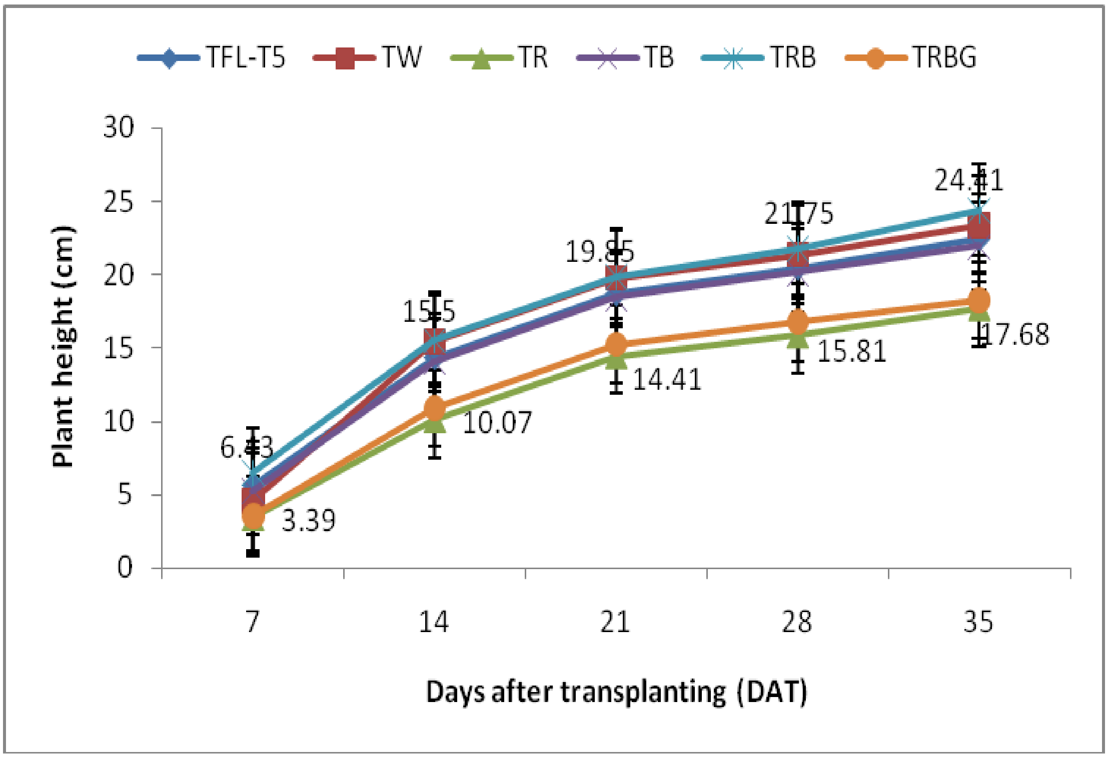

Figure 1. Plant height of pakchoi at different growth stages influenced by different LED-light spectral ratios

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

As a result, briefly it was found that at 7, 14, 21, 28 and 35 DAT, the highest plant height (24.41, 15.50, 19.85, 21.75 and 24.41 cm, respectively) was found from the treatment T_{RB} (Red and Blue - 4:1) followed by T_W (White - Full) whereas the lowest plant height (3.39, 10.07, 14.41, 15.81 and 17.68 cm, respectively) was found from the treatment T_R (Red - Full). The result obtained from the present study was similar with the findings of Bian *et al*. (2017) and they found that light supplementation can increase crop yield in greenhouses by promoting photosynthesis and plant growth and also obtained higher plant growth with red and blue LED light combination in pakchoi. Similar result was also observed by Poulet *et al*., 2014 and Piovene *et al*., 2015.

4.1.2 Number of leaves plant^{-1}

Number of leaves plant^{-1} of pakchoi at different growth stages varied significantly due to different LED-light spectral ratios (Figure 2 and Appendix V). It was observed that at 7 DAT, the highest number of leaves plant^{-1} (7.00) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was significantly different from other treatments followed by T_{FL-T5} (Fluorescent light – Full; control) and T_W (White - Full) whereas the lowest number of leaves plant^{-1} (4.00) was found from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was significantly different from other treatments. With the advancement of cropping duration, similar trend was found for number of leaves plant^{-1}. At 14 DAT, the highest number of leaves plant^{-1} (9.00) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was statistically identical with T_{FL-T5} (Fluorescent light – Full; control) and T_W (White - Full) whereas the lowest number of leaves plant^{-1} (5.33) was found from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was statistically identical with T_R (Red - Full) and T_B (Blue - Full). At 21 DAT, T_{RB} (Red and Blue - 4:1) gave the highest number of leaves plant^{-1} (10.33) which was statistically identical with control treatment T_{FL-T5} (Fluorescent light - Full) and T_W (White - Full) whereas T_{RBG} (Red, Blue with Green - 4:1:1) showed the lowest number of leaves plant^{-1} (6.67) which was statistically identical with T_R (Red - Full) and T_B (Blue - Full). Likewise, at 28 DAT, treatment T_{RB} (Red and Blue - 4:1) showed the highest number of leaves plant^{-1} (12.33) which was statistically similar with T_W (White - Full) whereas T_{RBG} (Red, Blue with Green - 4:1:1) showed the lowest number of leaves plant^{-1} (8.33) which was statistically identical with T_R (Red - Full) and T_B (Blue - Full). Again, at 35 DAT, the highest number of leaves plant^{-1} (14.33) was found from the treatment T_{RB} (Red and Blue - 4:1) which was significantly similar with T_W (White - Full) whereas the lowest number of leaves plant^{-1} (9.00) was found from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was statistically identical with T_R (Red - Full) and T_B (Blue - Full).

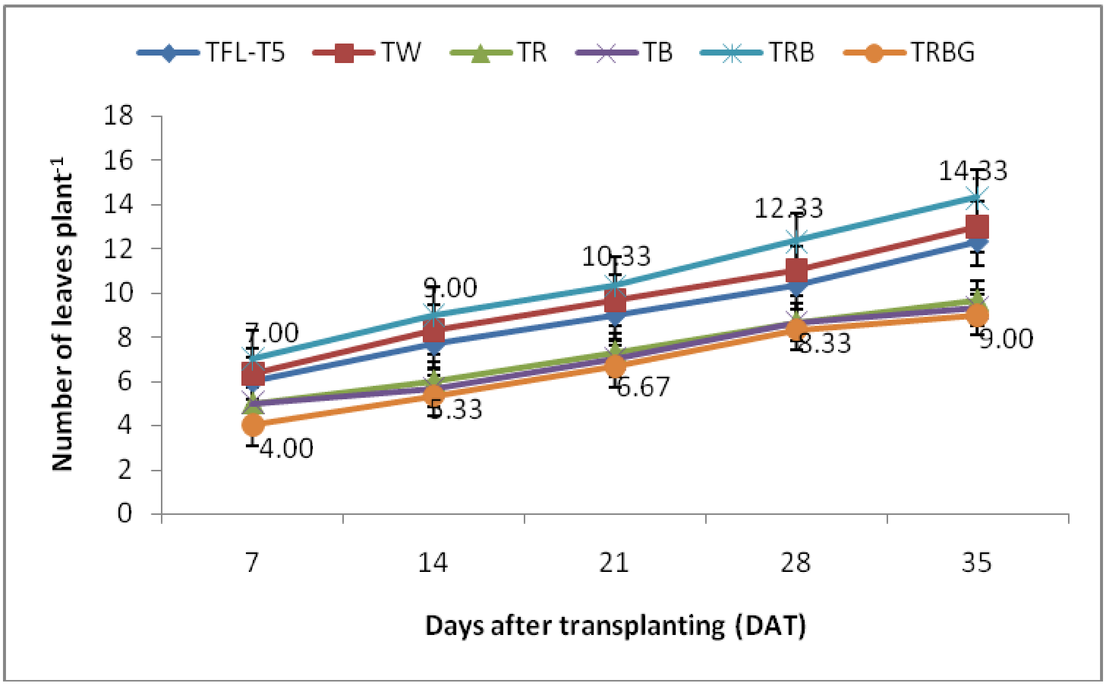

Figure 2. Number of leaves plant^{-1} of pakchoi at different growth stages influenced by different LED-light spectral ratios

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

So, briefly it can be summarized that the highest number of leaves plant^{-1} (7.00, 9.00, 10.33, 12.33 and 14.33 at 7, 14, 21, 28 and 35 DAT, respectively) was found from the treatment T_{RB} (Red and Blue - 4:1) followed by T_W (White -

Full) whereas the lowest number of leaves plant^{-1} (34.00, 5.33, 6.67, 8.33 and 9.00 at 7, 14, 21, 28 and 35 DAT, respectively) was found from the treatment T_R (Red - Full). Supported result was also observed by Huang *et al*. (2021) and reported that two-leafcolor pakchoi receiving alternating red and blue light exhibited more compact canopies than those under the control treatment.

4.1.3 Leaf length (cm)

Non-significant variation was found for leaf length of pakchoi at different growth stages except at 7 DAT due to different LED-light spectral ratios (Figure 3 and Appendix VI). It was observed that at 7 DAT, the highest leaf length (4.17 cm) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was statistically identical with $T_{FL\text{-}T5}$ (Fluorescent light – Full; control) and T_W (White - Full) whereas the lowest leaf length (1.56 cm) was found from the treatment T_R (Red - Full) which was statistically identical with T_B (Blue - Full) and T_{RB} (Red and Blue - 4:1). At 14, 21, 28 and 35 DAT, non-significant variation was found for leaf length (Appendix VI). However, at 14 DAT, the highest leaf length (15.31 cm) was recorded from the treatment T_{RB} (Red and Blue - 4:1) and the lowest leaf length (13.19 cm) was found from the treatment T_R (Red - Full). But at 21 DAT, T_W (White - Full) gave the highest leaf length (17.11 cm) whereas T_R (Red - Full) showed the lowest leaf length (15.38 cm). At 28 and 35 DAT, treatment T_W (White - Full) also showed the highest leaf length (17.71 and 18.23 cm, respectively) whereas T_R (Red - Full) showed the lowest leaf length (15.98 and 16.50 cm, respectively). So, briefly it can be reviewed that the highest leaf length at 7 and 14 DAT (4.17 and 15.31 cm, respectively) was found from the treatment T_{RB} (Red and Blue - 4:1) but at 21, 28 and 35 DAT, the highest leaf length (17.11, 17.71 and 18.23 cm, respectively) was found from the treatment T_W (White - Full) whereas at 7, 14, 21, 28 and 35 DAT, the lowest leaf length (1.56, 13.19, 15.38, 15.98 and 16.50 cm, respectively) was found from the treatment T_R (Red - Full). The result of the present study on leaf length suggested that different LED-light spectral ratios had significant effect on leaf length of pakchoi and among the treatments T_W (White - Full) showed highest leaf length which indicated that white LED light was best for higher leaf length.

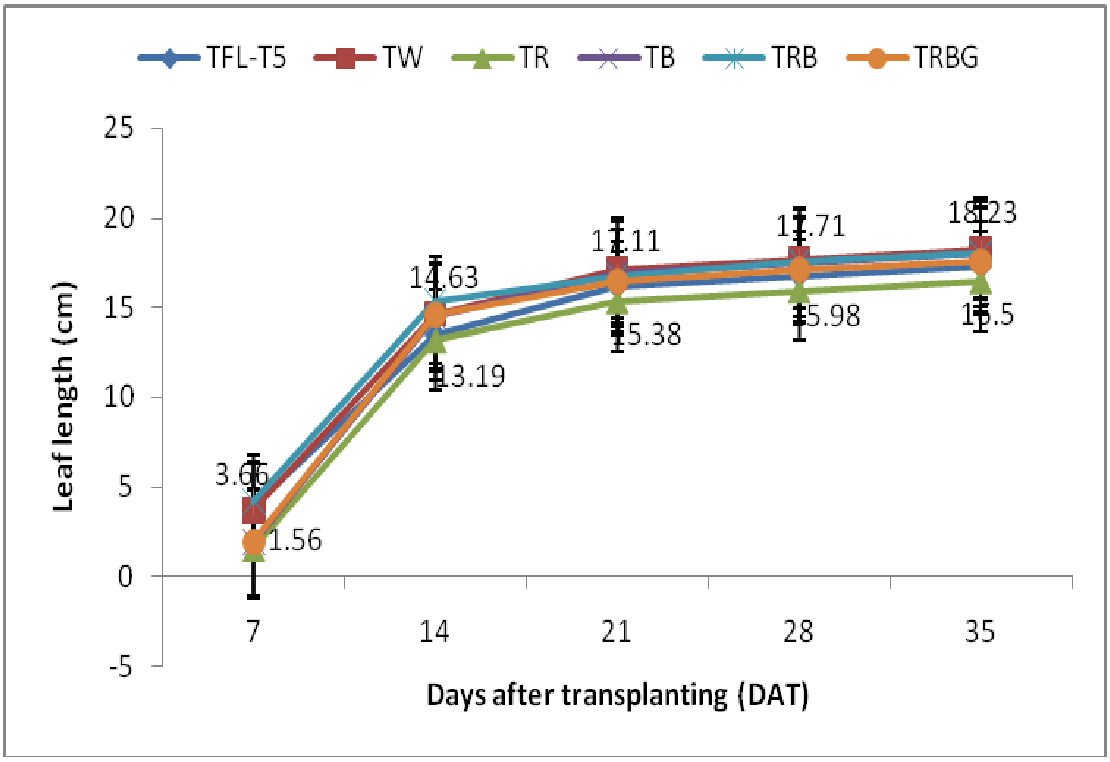

Figure 3. Leaf length of pakchoi at different growth stages influenced by different LED-light spectral ratios

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

4.1.4 Leaf breadth (cm)

Leaf breadth of pakchoi at different growth stages varied significantly due to different LED-light spectral ratios (Figure 4 and Appendix VII). At 7 DAT, the highest leaf breadth (2.27 cm) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was significantly same with T_{FL-T5} (Fluorescent light – Full; control) and T_W (White - Full) whereas the lowest leaf breadth (0.83 cm) was found from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was significantly same with T_R (Red - Full) and T_B (Blue - Full). With the advancement of cropping duration, similar trend was found for leaf breadth. At

14 DAT, the highest leaf breadth (6.97 cm) was recorded from the treatment T_{RB} (Red and Blue - 4:1) which was statistically similar with $T_{FL\text{-}T5}$ (Fluorescent light – Full; control) and T_W (White - Full) whereas the lowest leaf breadth (3.21 cm) was found from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was statistically similar with T_R (Red - Full) and T_B (Blue - Full). At 21 DAT, T_{RB} (Red and Blue - 4:1) gave the highest leaf breadth (7.77 cm) which was statistically similar with control treatment $T_{FL\text{-}T5}$ (Fluorescent light - Full) and T_W (White - Full) whereas T_{RBG} (Red, Blue with Green - 4:1:1) showed the lowest leaf breadth (3.70 cm) which was statistically similar with T_R (Red - Full) and T_B (Blue - Full). Likewise, at 28 DAT, treatment T_{RB} (Red and Blue - 4:1) showed the highest leaf breadth (8.01 cm) which was statistically similar with control treatment $T_{FL\text{-}T5}$ (Fluorescent light - Full) and T_W (White - Full) whereas T_{RBG} (Red, Blue with Green - 4:1:1) showed the lowest leaf breadth (3.85 cm) which was statistically identical with T_R (Red - Full) and T_B (Blue - Full). Again, at 35 DAT, the highest leaf breadth (9.10 cm) was found from the treatment T_{RB} (Red and Blue - 4:1) which was significantly similar with control treatment $T_{FL\text{-}T5}$ (Fluorescent light - Full) whereas the lowest leaf breadth (4.22 cm) was found from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was significantly same with T_B (Blue - Full). So, briefly it can be stated that at 7, 14, 21, 28 and 35 DAT, the highest leaf breadth (2.27, 6.97, 7.77, 8.01 and 9.10 cm, respectively) was found from the treatment T_{RB} (Red and Blue - 4:1) whereas the lowest leaf breadth (0.83, 3.21, 3.70, 3.85 and 4.22 cm, respectively) was found from the treatment T_R (Red - Full). Similar result was also observed by Huang *et al*. (2021) who reported that two-leafcolor pakchoi receiving alternating red and blue light exhibited more compact canopies and wider leaves than those under the control treatment.

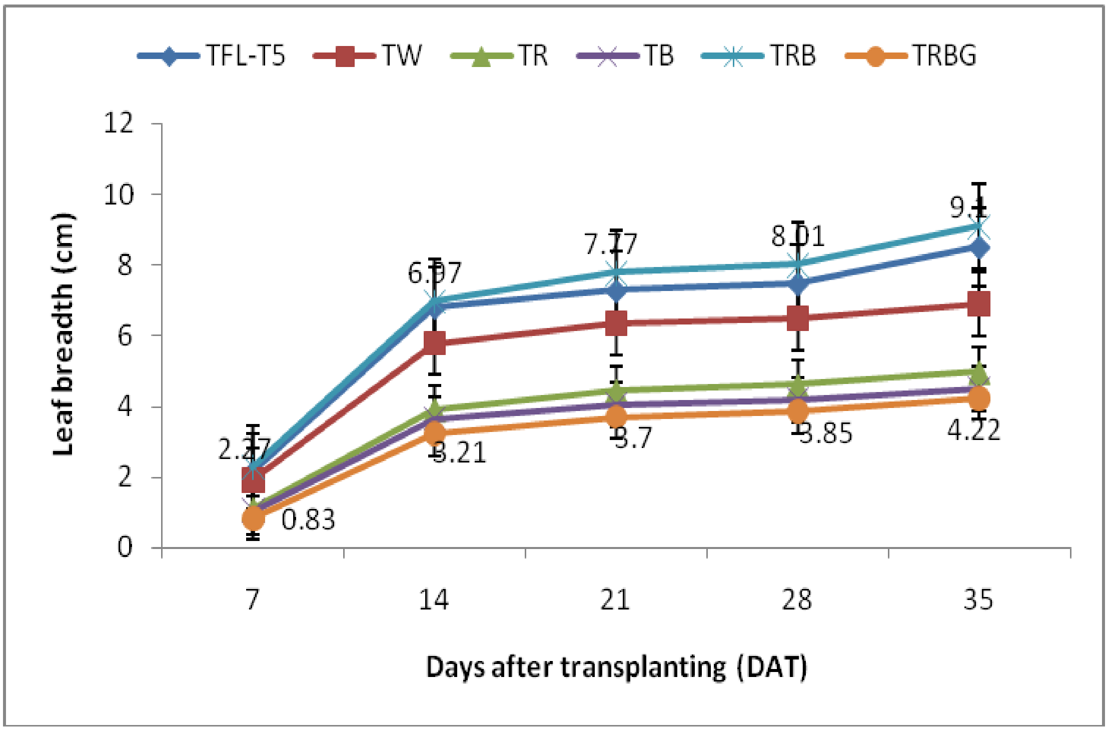

Figure 4. Leaf breadth of pakchoi at different growth stages influenced by different LED-light spectral ratios

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

4.2 Yield contributing parameters and yield of pakchoi

4.2.1 Stem diameter (cm)

At the time of harvest, stem diameter of pakchoi differed significantly with the effect of different LED-light spectral ratios (Figure 5 and Appendix VIII). Results indicated that the highest stem diameter (1.20 cm) was found from the treatment T_{RB} (Red and Blue - 4:1) which was significantly similar with control treatment T_{FL-T5} (Fluorescent light - Full) and T_W (White - Full). On the other hand, the lowest stem diameter (0.70 cm) was recorded from the treatment T_B (Blue - Full) which was significantly same with T_R (Red - Full). The result of the present study suggested that lighting effect of Red and Blue - 4:1 ratio was best for stem diameter of pakchoi compared to other treatments including control which indicated that for stem diameter of pakchoi, the treatment T_{RB}

(Red and Blue - 4:1) is the best choice; because stem diameter is a yield contributing character that influence yield of pakchoi significantly.

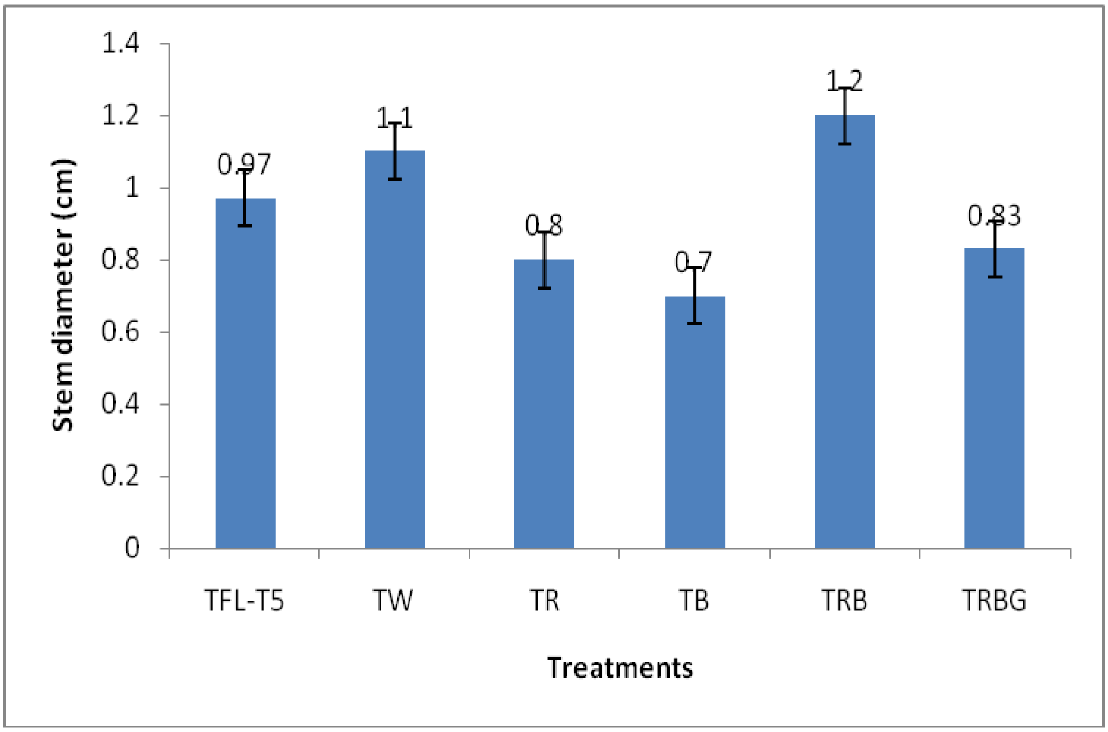

Figure 5. Stem diameter of pakchoi at harvest influenced by different LED-light spectral ratios

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

4.2.2 Leaf base breadth (cm)

Leaf base breadth of pakchoi at harvest differed significantly by the effect of different LED-light spectral ratios (Figure 6 and Appendix VIII). Results indicated that the maximum leaf base breadth (1.92 cm) was found from the treatment T_{RB} (Red and Blue - 4:1) that was significantly similar to control treatment T_{FL-T5} (Fluorescent light - Full). Again, the minimum leaf base breadth (1.36 cm) was recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) that was significantly same to T_w (White - Full). The result of the present study suggested that lighting effect of Red and Blue - 4:1 ratio was best for leaf base breadth of pakchoi compared to other treatments including control which indicated that for leaf base breadth, the treatment T_{RB} (Red and Blue -

4:1) is the best choice; because leaf base breadth is a yield contributing character that influence yield of pakchoi significantly.

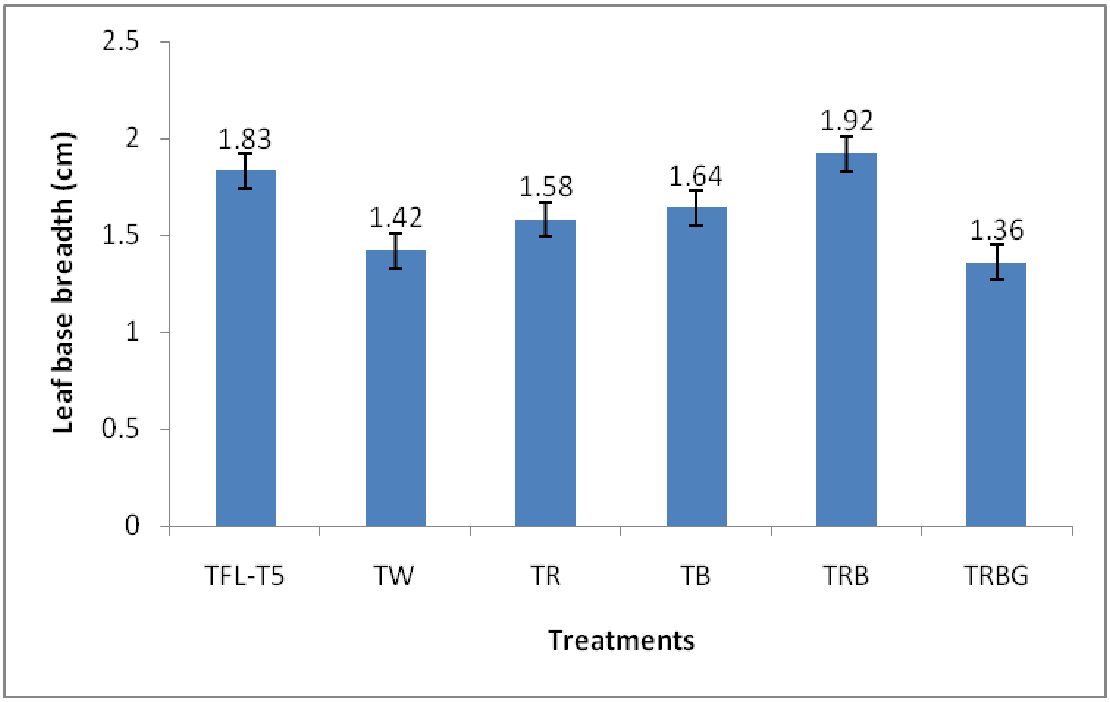

Figure 6. Leaf base breadth of pakchoi at harvest influenced by different LED-light spectral ratios

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

4.2.3 Whole plant diameter plant^{-1} (cm)

Whole plant diameter (cm) of pakchoi at harvest differed significantly by the effect of different LED-light spectral ratios (Figure 7 and Appendix VIII). Results indicated that the maximum average whole plant diameter plant^{-1} (8.24 cm) was found from the treatment T_{RB} (Red and Blue - 4:1) that was significantly different to other treatments followed by T_{FL-T5} (Fluorescent light - Full). Again, the minimum whole plant diameter plant^{-1} (6.08 cm) was recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) that was significantly same to T_w (White - Full). The result of the present study suggested that lighting effect of Red and Blue - 4:1 ratio was best for whole plant diameter plant^{-1} of pakchoi compared to other treatments including control which indicated that for whole plant diameter plant^{-1}, the treatment T_{RB} (Red and Blue - 4:1) is the best choice.

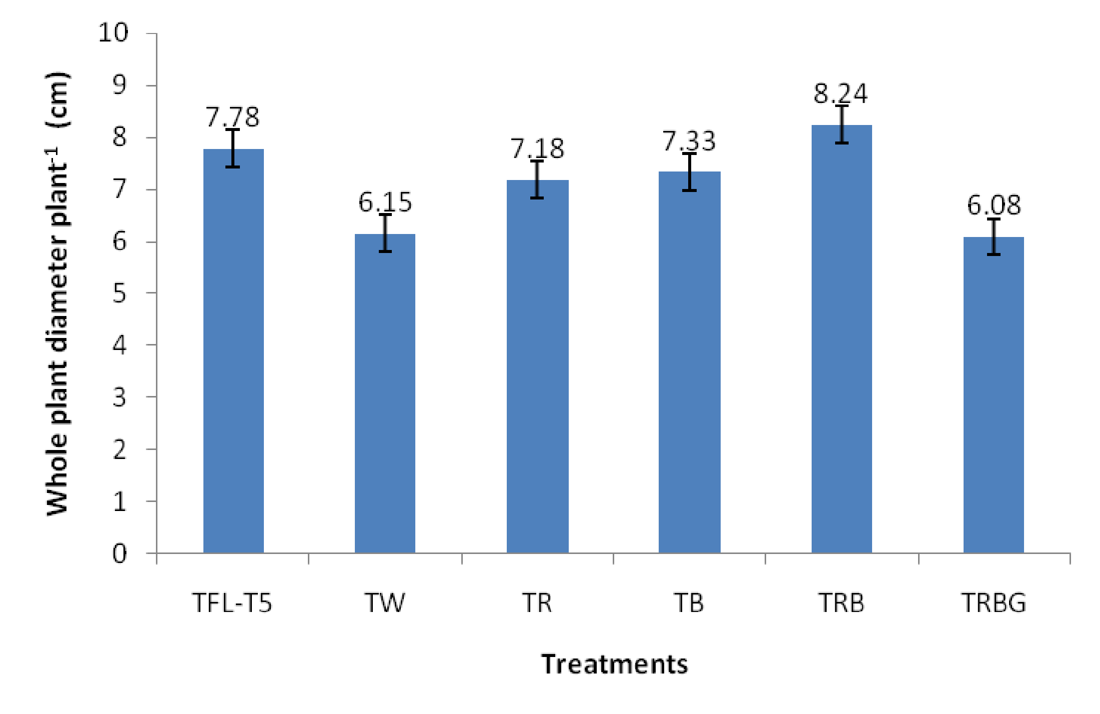

Figure 7. Whole plant diameter plant^{-1} of pakchoi at harvest influenced by different LED-light spectral ratios

$T_{FL\text{-}T5}$ = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

4.2.4 Fresh weight plant^{-1} (g)

Significant variation was recorded on fresh weight plant^{-1} of pakchoi as affected by different LED-light spectral ratios (Table 1 and Appendix VIII). Results revealed that the highest fresh weight plant^{-1} (336.67 g) was achieved from the treatment T_{RB} (Red and Blue - 4:1) which was significantly different from other treatments followed by $T_{FL\text{-}T5}$ (Fluorescent light - Full), T_R (Red - Full) and T_B (Blue - Full). The lowest fresh weight plant^{-1} (120.00 g) was recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was significantly same with T_W (White - Full). In general, green light is known to deeply penetrate the plant canopy compared to blue and red affecting the growth and synthesis of bioactive compounds via cryptochrome-dependent/independent processes (Sun *et al*., 1998). When green light is supplemental for red and blue LED combination in controlled environmental agriculture, the effects on plant physiology are reported to be dependent on its proportions. Green light at a proportion in the red-blue light environment, has been reported to reverse blue- or red-light-induced responses (i.e., stem growth rate inhibition, chloroplast gene expression, stomatal opening, phytochemical accumulation) (Zhang and Folta, 2012), and negatively influence the quality of microgreen products (Dou *et al*., 2019). The result of the present study was in agreement with the findings of Yan *et al*. (2020) and reported that red plus blue light-emitting diodes (LEDs) increased leaf fresh and dry weights of green leaf lettuce by 28% and 34%, respectively. Yan *et al*. (2020) also found similar result with the present study and found that red plus blue LEDs increased fresh biomass of hydroponic lettuce. Lobiuc *et al*. (2017) also found similar result with the present study.

4.2.5 Total yield (kg)

Statistically significant difference among the treatment was found on total yield of pakchoi in the experiment area per treatment as influenced by different LED-light spectral ratios (Table 1 and Appendix VIII). Results exhibited that the highest total yield (1.29 kg) was found from the treatment T_{RB} (Red and Blue - 4:1) which was significantly similar with control treatment $T_{FL\text{-}T5}$ (Fluorescent light - Full). Reversely, the lowest total yield (0.18 kg) was recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was significantly different from other treatments. Specht *et al*. (2014) reported that vertical farming facilitates the production of high value crops with higher yield than obtained from conventional farming. In such a case, LED lights are more effective with high energy use efficiency and durability than traditional light sources like fluorescent lamps (Specht *et al*., 2014). A blue and red background (B16R84) with 24% GL from green fluorescent lamp (B15G24R61) has been shown to increase lettuce yield (Kim *et al*., 2004), whereas in another study, an increase of up to 30% green LED light does not influence the dry mass in the same cultivar (Snowden *et al*., 2016). Piovene *et al*. (2015) found that the plants expressed increased biomass and fruit yield of basil and strawberry when treated with LED with highest energy use efficiency than traditional fluorescent lamps and spectral red: blue ratio of 0.7 was essential for proper plant growth and improved nutraceutical properties. It was reported that by Wong (2020) that lighting quality and quantity can be manipulated to improve yield and phytonutrient contents of leafy greens.

Table 1. Yield contributing parameters and yield of pakchoi at harvest influenced by different LED-light spectral ratios

Treatment	Yield contributing parameters and yield	
	Fresh weight plant^{-1} (g)	Total yield (kg)
T_{FL-T5}	276.67 b	1.04 ab
T_W	164.00 c	0.49 c
T_R	236.67 b	0.78 b
T_B	256.67 b	0.92 b
T_{RB}	336.67 a	1.29 a
T_{RBG}	120.00 c	0.18 d
SE(±)	26.813	0.119
$LSD_{0.05}$	59.743	0.265
CV(%)	8.17	8.59

In a column means having similar letter(s) arc statistically identical and those having dissimilar letter(s) differ significantly as per 0.05 level of probability

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

4.3 Quality parameters

4.3.1 SPAD value

SPAD value of pakchoi showed statistically significant variation as affected by different LED-light spectral ratios (Table 2 and Appendix IX). Results revealed that the highest SPAD value (42.04) was found from the treatment T_B (Blue - Full) which was significantly similar with T_{FL-T5} (Fluorescent light - Full), T_R (Red - Full), T_{RB} (Red and Blue - 4:1) and T_{RBG} (Red, Blue with Green - 4:1:1) whereas the lowest SPAD value (34.60) was recorded from the treatment T_W (White - Full). This result indicated that the blue light (BL) is responsible for higher chlorophyll content. BL influences photosynthetic activity by inducing stomatal opening (Zeiger *et al*., 2002) and affecting chloroplast movement within the cell (Kasahara *et al*., 2002) in the short term while increasing stomata number and leaf thickness in the long term (Hogewoning *et al*., 2010; Wang *et al*., 2016). BL is also known to increase the chlorophyll content (Hogewoning *et al*., 2010; Johkan *et al*., 2010; Matsuda *et al*., 2007). A greater fraction of BL is associated with the development of "sun-type" leaf characterized by a high leaf thickness and photosynthetic capacity (Hogewoning *et al*., 2010; Matsuda *et al*., 2007).

4.3.2 Vitamin-C content (mg/100g)

Different LED-light spectral ratios showed significant influence on vitamin-C content of pakchoi (Table 2 and Appendix IX). The highest vitamin-C content (41.81 mg/100g) was recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1) which was statistically similar with control treatment T_{FL-T5} (Fluorescent light - Full), T_W (White - Full) andT_{RB} (Red and Blue - 4:1) whereas the lowest vitamin-C content (38.24mg/100g) was found from the treatment T_R (Red - Full). The result obtained from the present study on vitamin-C content was similar with the findings of Huang *et al*. (2021).

4.3.3 Brix content (°Brix)

Brix content of pakchoi varied significantly due to different LED-light spectral ratios (Table 2 and Appendix IX). The maximum total soluble solids (9.24 °Brix) was found in T_{RB} (Red and Blue - 4:1) which was significantly differed to other treatments followed by T_{FL-T5} (Fluorescent light - Full) whereas minimum total soluble solids (5.16 °Brix) was recorded from T_B (Blue - Full) that was significantly similar to T_R (Red - Full).

4.3.4 Moisture content (%)

Significant variation was found for moisture content percentages of pakchoi due to different LED-light spectral ratios (Table 2 and Appendix IX). The highest moisture content (94.12%) was observed in T_B (Blue - Full) that was statistically similar to T_R (Red - Full) treatment whereas the lowest moisture content (88.72%) was in T_{FL-T5} (Fluorescent light - Full) that was statistically similar (88.72%) to T_{RB} (Red and Blue - 4:1).

Table 2. Quality parameters of pakchoi at harvest influenced by different LED-light spectral ratios

Treatment	Quality parameters			
	SPAD value	Vitamin-C content (mg/100g)	Brix value (°Bx)	Moisture content (%)
T_{FL-T5}	36.45 ab	40.67 ab	8.12 b	88.72 d
T_W	34.60 b	40.43 ab	6.92 c	92.68 c
T_R	38.82 ab	38.24 c	5.27 d	93.96 a
T_B	42.04 a	39.81 b	5.16 d	94.12 a
T_{RB}	41.30 ab	41.00 ab	9.24 a	88.94 d
T_{RBG}	38.68 ab	41.81 a	6.48 c	93.04 b
SE(±)	3.064	0.650	0.211	0.286
$LSD_{0.05}$	6.826	1.449	0.463	0.344
CV(%)	9.71	3.97	3.14	5.72

In a column means having similar letter(s) are statistically identical and those having dissimilar letter(s) differ significantly as per 0.05 level of probability

T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1, T_{RBG} = Red, Blue with Green - 4:1:1

CHAPTER V

SUMMARY AND CONCLUSION

The experiment was conducted at the roof top (indoor structure) of Dr. M. Wazed Mia Central Laboratory, Sher-e-Bangla Agricultural University, Dhaka during the period from November 2020 to December 2020 to study the improvement of plant growth, nutrient and energy use efficiency of Pakchoi under different LED-light spectrum in vertical grow-house. The experiment consisted of one factor regarding the treatments of T_{FL-T5} = Fluorescent light (control) - Full, T_W = White - Full, T_R = Red - Full, T_B = Blue - Full, T_{RB} = Red and Blue - 4:1 and T_{RBG} = Red, Blue with Green - 4:1:1. Photosynthetic Photon Flux Density (PPFD) of different LED light was maintained between 100-120 μmol/m^2/s and duration was 16 hrs per day. Six treatments of the experiment which were laid out in a Randomized Complete Block Design (RCBD) with three replications. Data on different growth, yield contributing parameters and yield parameters and quality parameters were recorded and statistically analyzed using MSTAT-C computer package program. Different treatments showed significant influence on most of the growth, yield contributing parameters and yield and quality parameters of pakchoi.

Regarding growth parameters, the highest plant height at 7, 14, 21, 28 and 35 DAT, (24.41, 15.50, 19.85, 21.75 and 24.41 cm, respectively), number of leaves $plant^{-1}$ (7.00, 9.00, 10.33, 12.33 and 14.33, respectively) and leaf breadth (2.27, 6.97, 7.77, 8.01 and 9.10 cm, respectively) were found from the treatment T_{RB} (Red and Blue - 4:1) but the highest leaf length at 7 and 14 DAT (4.17 and 15.31 cm, respectively) was found from the treatment T_{RB} (Red and Blue - 4:1) while at 21, 28 and 35 DAT, it was highest (17.11, 17.71 and 18.23 cm, respectively) from the treatment T_W (White - Full). On the other hand, at 7, 14, 21, 28 and 35 DAT, the lowest plant height (3.39, 10.07, 14.41, 15.81 and 17.68 cm, respectively), number of leaves $plant^{-1}$ (34.00, 5.33, 6.67, 8.33 and 9.00, respectively), leaf length (1.56, 13.19, 15.38, 15.98 and 16.50 cm, respectively) and leaf breadth (0.83, 3.21, 3.70, 3.85 and 4.22 cm, respectively) were found from the treatment T_R (Red - Full).

Regarding yield contributing and yield parameters, the highest stem diameter (1.20 cm), leaf base breadth (1.92 cm), whole plant diameter plant^{-1} (8.24 cm), fresh weight plant^{-1} (336.67 g) and total yield (1.29 kg) per replication (5 plants) were found from the treatment T_{RB} (Red and Blue - 4:1) whereas the lowest stem diameter (0.70 cm) was recorded from the treatment T_B (Blue - Full) but lowest leaf base breadth (1.36 cm), fresh weight plant^{-1} (120.00 g) and total yield (0.18 kg) per replication (5 plants) were recorded from the treatment T_{RBG} (Red, Blue with Green - 4:1:1).

In respect of quality parameters, the treatment T_B (Blue - Full) gave the highest SPAD value (42.04) whereas T_W (White - Full) showed lowest SPAD value (34.60). Similarly, the treatment T_{RBG} (Red, Blue with Green - 4:1:1) showed the highest vitamin-C content (41.81 mg/100g) whereas T_R (Red - Full) gave the lowest vitamin-C content (38.24mg/100g). Again, the treatment T_{RB} (Red and Blue - 4:1) gave the maximum total soluble solids content (9.24 °Brix) whereas the treatment T_B (Blue - Full) showed minimum total soluble solids content (5.16 °Brix). Similarly, the maximum moisture content (94.12%) was observed in T_B (Blue - Full) and the minimum moisture content (88.72%) was in $T_{FL\text{-}T5}$ (Fluorescent light - Full).

Conclusion

From the above results, it can be concluded that among the treatments of different LED-light spectrum in vertical farming, the treatment T_{RB} (Red and Blue - 4:1) had best significant positive effect on growth, yield contributing parameters and yield and quality parameters of pakchoi and resulted highest fresh weight plant^{-1} (336.67 g) and total yield per replication (5 plants) (1.29 kg) compared to all other treatments. And the highest photosynthetic performance was found in treatment T_B (Blue - Full) where SPAD value was highest (42.04).All of these were my objectives that i found .So, the treatment

T_{RB} (Red and Blue - 4:1) can be considered as the best treatment followed by control treatment $T_{FL\text{-}T5}$ (Fluorescent light - Full) among all the treatments.

Recommendation

Considering this situation from the present study, further studies in the following areas may be suggested:

1. Some other LED-light spectrum treatments may be used in future study.
2. Another variety of pakchoi and/or other vegetables need to be considered before final recommendation.

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APPENDICES

Appendix I. Agro-Ecological Zone of Bangladesh showing the experimental location

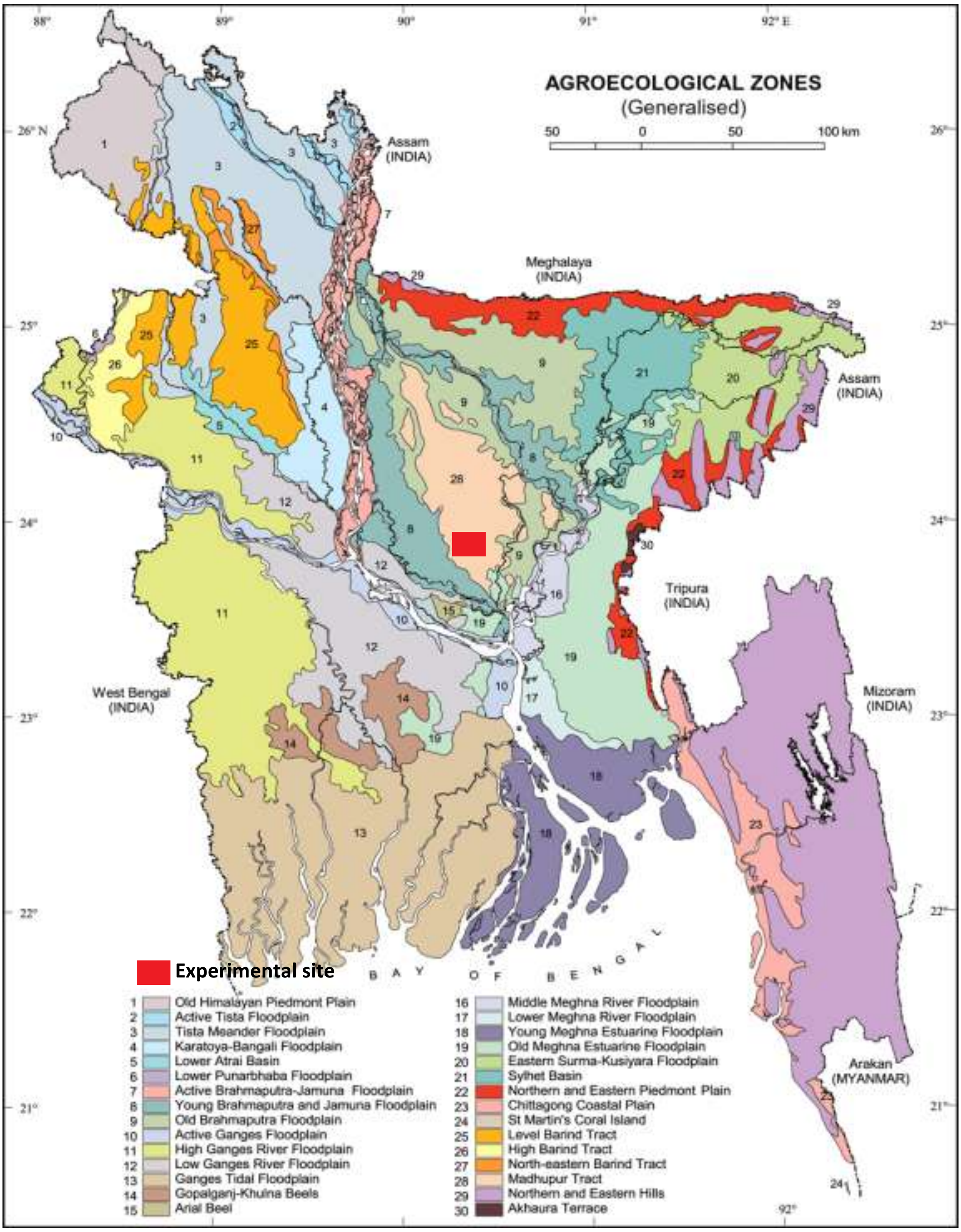

Figure 10. Experimental site

Appendix II. Monthly average temperature, relative humidity and total rainfall and sunshine of the experimental site during the period from November, 2020 to December, 2020.

Month	Air temperature (°c)		Relative humidity (%)	Rainfall (mm) (total)	Sunshine (hr)
	Maximum	Minimum			
November, 2020	31	18.0	99	227	5.8
December, 2020	32.4	16.3	69	0	7.9

Source: Bangladesh Meteorological Department (Climate and Weather Division), Agargoan, Dhaka – 1212

Appendix III. Layout of the experiment treatments

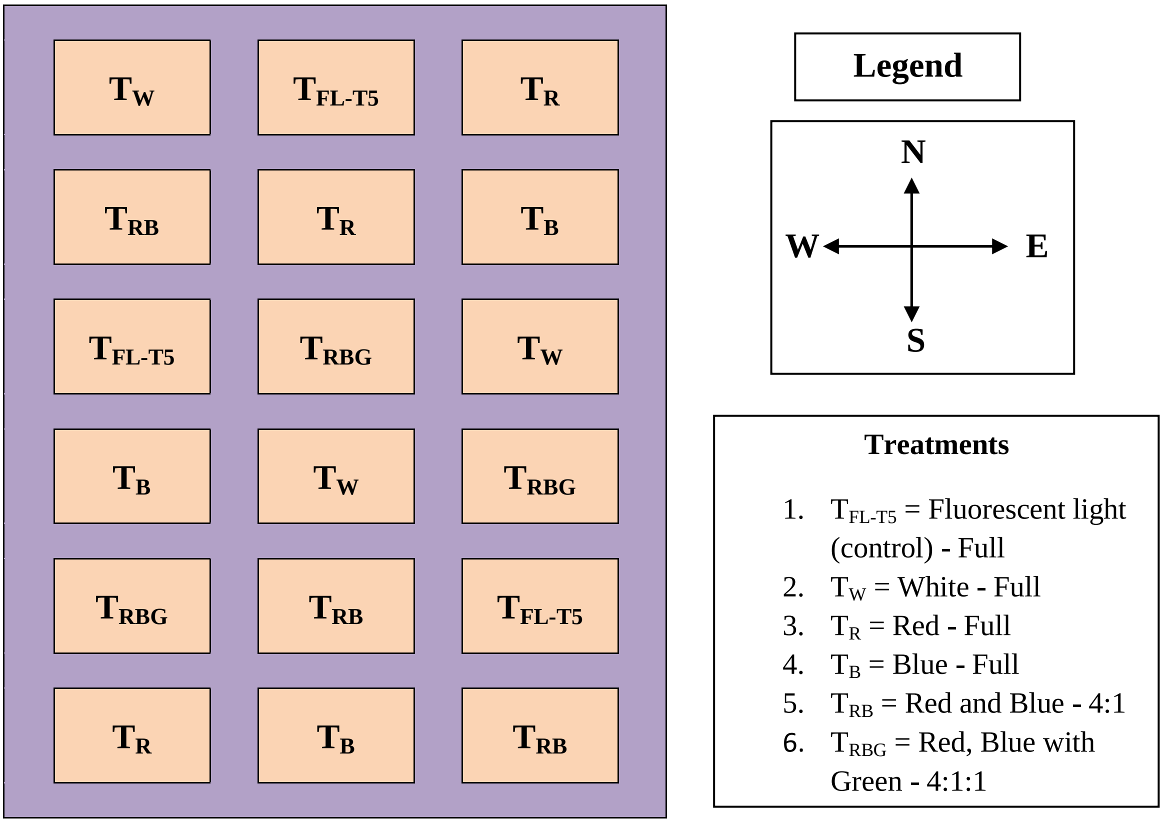

Figure 11. Layout of the experimental plot

Appendix IV. Mean square of plant height of pakchoi at different growth stages influenced by different LED-light spectral ratios

Source	DF	Mean square of plant height				
		7 DAT	14 DAT	21 DAT	28 DAT	35 DAT
Replication	2	0.125	2.434	4.134	4.136	3.489
Light	5	4.279**	16.388*	16.392*	18.542*	22.967*
Error	10	0.110	1.005	1.004	1.001	0.987

NS = Non-significant * = Significant at 5% level ** = Significant at 1% level

Appendix V. Mean square of number of leaves plant^{-1} of pakchoi at different growth stages influenced by different LED-light spectral ratios

Source	DF	Mean square of number of leaves plant^{-1}				
		7 DAT	14 DAT	21 DAT	28 DAT	35 DAT
Replication	2	0.056	1.052	5.388	11.167	10.170
Light	5	3.555**	7.658**	7.655**	8.233*	14.133*
Error	10	0.065	0.645	0.653	0.700	0.702

NS = Non-significant * = Significant at 5% level ** = Significant at 1% level

Appendix VI. Mean square of leaf length of pakchoi at different growth stages influenced by different LED-light spectral ratios

Source	DF	Mean square of leaf length				
		7 DAT	14 DAT	21 DAT	28 DAT	35 DAT
Replication	2	0.063	0.683	1.986	2.124	2.290
Light	5	4.111*	1.897^{NS}	1.186^{NS}	1.268^{NS}	1.268^{NS}
Error	10	0.116	1.444	2.162	2.257	2.257

NS = Non-significant * = Significant at 5% level ** = Significant at 1% level

Appendix VII. Mean square of leaf breadth of pakchoi at different growth stages influenced by different LED-light spectral ratios

Source	DF	Mean square of leaf breadth				
		7 DAT	14 DAT	21 DAT	28 DAT	35 DAT
Replication	2	0.003	6.057	5.707	5.736	11.106
Light	5	1.191**	8.376**	9.279*	9.603*	13.457*
Error	10	0.064	1.220	1.478	1.520	1.390

NS = Non-significant * = Significant at 5% level ** = Significant at 1% level

Appendix VIII. Mean square of yield contributing parameters and yield of pakchoi at harvest influenced by different LED-light spectral ratios

Source	DF	Mean square of yield contributing parameters and yield				
		Stem diameter (cm)	Leaf base breadth	Whole plant diameter plant^{-1} (cm)	Fresh weight plant^{-1} (g)	Total yield (kg)
Replication	2	0.015	0.008	1.107	30.401	0.032
Light	5	0.109**	0.086**	6.733*	18448.91*	0.478**
Error	10	0.022	0.014	0.204	78.402	0.021

NS = Non-significant * = Significant at 5% level ** = Significant at 1% level

Appendix IX. Mean square of quality parameters of pakchoi at harvest influenced by different LED-light spectral ratios

Source	DF	Mean square of quality parameters			
		SPAD value	Vitamin-C content (mg/100g)	Brix value (°Bx)	Moisture content (%)
Replication	2	4.091	0.069	0.397	2.384
Light	5	23.868*	4.448**	12.144**	275.36**
Error	10	14.078	0.634	0.632	2.137

NS = Non-significant * = Significant at 5% level ** = Significant at 1% level

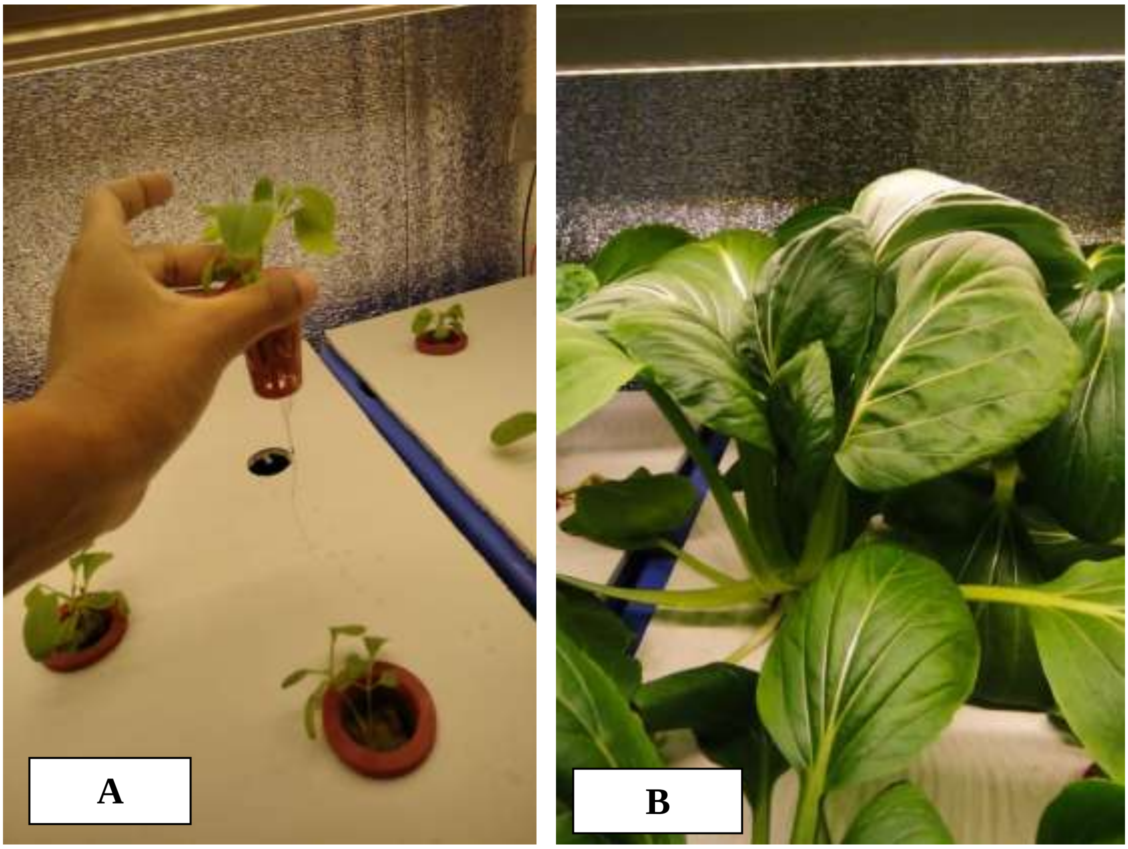

Plate 7. Plants at A = Early stage and B = Maturity stage

Plate 8. Plant growth under red + blue + green at 4:1:1 ration (L_1)

Plate 9. Plant growth under white – Full light (L_5)

Plate 10. Plant growth under Fluorescent light (control) – Full (L_6 - Worm)