I. INTRODUCTION

Rice plant (2n=24) belongs to family *Poaceae*. There are more than 20 species of rice (*Oryza* genus), but two species, *Oryza sativa* L.*;* originated in the humid tropics of South and Southeast Asia, and *Oryzaglaberrima*L.*;* originated and cultivated in the Niger basin in Africa **(Chaudhary, 2003).**

Rice (*Oryza sativa* L.) is one of the agronomically and nutritionally most important cereal crops. It is a major source of food for more than 2.7 billion people and is planted on about one-tenth of the earth's arable land (more than 150 million ha, producing almost 600 million tons annually) **(Maclean *et al*., 2002;Chaudhary, 2003; El-Kady and Abd Allah, 2004;khush 2005).**

Oryza sativa L. is the smallest genome of the cereals family, consisting of just 430Mb across 12 chromosomes. It is renowned for being easy to genetic modifications, and is a model genome for cereal biology.

In Egypt, rice is considered as one of the most important cereal crops, since it contributes about 20% of the total cereal consumption and also as cash export crop. Annually, more than one million and halffeddans are cultivated with rice, producing 6-7 million tons of paddy with an average of 4 tons/fed.,which is considered as one of the highest per unit area average in the world. This production is covers the local consumption, in addition to some amount for exportation. However, in the near future, this production will not be sufficient because of high rate of population increase, which is expected to be 100 million individuals by the year 2025. Great efforts have to be done to increase the productivity of rice, keeping in mind the limitations of cultivated area and irrigation water (**RRTC, 2008**).

Plant development is strongly influenced by the environment. Of all of the environmental factors, light arguably has the most formative influence on the life history of a plant. Cues from the light environment are involved in the regulation of seed germination, establishment of seedlings, determination of growth habit, and the transition to flowering. Plants have evolved an extensive collection of photoreceptors to perceive signals about their light environment. Red (R) and far-red (FR) lights are important environmental signals in the regulation of plant development, with major roles in seedling deetiolation, neighbor detection, and photoperiodism**(Smith, 1994).**

The plant R/FR photoreceptors, the phytochromes, are soluble chromoproteins consisting of a 120-130 kDapoprotein and a linear tetrapyrrolechromophore. A small gene family (*PHY*) encoding the phytochromeapoproteins (chromoproteins), functions in the absorption of R and FR lights and the transduction of intracellular signals during light-regulated plant development **(Sharrock and Clack, 2002).**Phytochromes are signal-transducing photoreceptors that interchange between inactive and active forms in response to different wavelengths of light. This conversion is used to synchronize plant development to the exigencies of the light environment. Light signal provide information of crucial ecological value at many stages in development,seed germination and seedling establishment, the proper development of the photosynthetic machinery, the architecture of the vegetative plant, the timing of flowering, tuberization and bud dormancy, the responses to neighbour competition, and the allocation of resources to root, stem, leaf, reproductive or storage structures are all potentially controlled by the perception of environmental light signals by the phytochromes. It is now possible to begin to trace the biological significance of photoperception from the molecular mechanisms to the life of plants in their natural environment. (**Smith, 2000)**

Phytochromes also provide plants with temporal signals that entrain the phases of the biological clock, and others to ensure that crucial developmental steps initiated at appropriate points in the life cycle. Endogenous circadian rhythms synchronize development to the changing seasons, as exemplified in the photoperiodic control of flowering and dormancy. Even when employed as simple light detectors such as in the stimulation of seed germination or the conversion of the etiolated seedling to photosynthetic competence, the phytochromes may be thought of as timing agents. Germination of the seed and maturation of the developing seedling, both dependent upon limited stored reserves, are probably the most vulnerable stages of the life cycle, and getting the timing right must be vital to longterm survival. In these processes, the phytochromes do not work alone; the cryptochromes are often responsible for initiating germination and they have important roles in de-etiolation. One implication of this may be that getting the timing right is so crucial to survival that reliance on one set of environmental light signals alone is insufficient to guarantee success under all conditions.

RNA interference (RNAi) is a powerful reverse genetic tool to study gene function by the interference with the gene activity**(Shokry*et al*., 2006)**.

Microprojectile bom*bar*dment, also called the biolistic method or the particle gun method has been used in many laboratories to deliver transgene. The ability to deliver foreign DNA into regenerable cells, tissues or organs appears to provide the best method for achieving genotype independent transformation bypass Agrobacterium host specificity.

To construct transformation vectors that produce RNAs capable for duplex formation, gene-specific sequences (*PHYB* gene) in the sense and antisense orientations were linked and placed under the control of a strong viral promoter (35S).

The ultimate goal of rice investigation is to study the effect of knocking down the *PhyB* gene on some yield-related traits.

The objectives of this investigation were:

1. Developing a plasmid construct carrying the *PhyB-RNAi* cassette.
2. Transforming Giza 177 rice variety with the construct carrying the *PhyB-RNAi* cassette using biolistic gun.
3. Characterizing and analyzing transgenic plants for the expression of the *PhytochromeB* gene.

4- Study the differences in yield-related traits between wild type plants and *PhyB*-RNAi lines.

ABSTRACT

Hussien Fotouh Alameldin, Molecular genetic studies on some yield-related traits in rice using RNAi, Unpublished Ph.D. Thesis, Department of GeneticsFaculty of Agriculture, Ain Shams University. 2012.

This study aimed to evaluate some yield-related traits caused by knocking-down the expression of *phytochromeB* Gene in Egyptian rice (GIZA177). This was achieved by introducing plant an expression vector pPhy/RNAi/bar which included sense, loop and antisense strands, respectively, integrated into the genome of the cultivar Giza177 using mature embryo calli as a target tissue and microprojectile bombardment as a transformation device. The *PhyB-RNAi*cassette was driven by the *35S* promoter upstream to down regulate the expression of the gene. We succeeded to knock-down *PhyB* expression up to 5 fold change in PhyB-RNAi plants than to wild type Giza177 plant. This was confirmed by northern blot, reverse transcriptase PCR and quantitative real time PCR. Changes in some yield-related traits were detected in PhyB-RNAi transgenics, plant height after 30 days and flag leaf width were increased and the number of days to heading was decreased.

Key Words:

Rice, *Oryza Sativa*, RNAi, Bombardment, PhytochromeB.

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