

**Short Communication**

**Light-harvesting Complex and how it Affect Growth of *Arabidopsis thaliana* plants**

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**ABSTRACT**

Light-harvesting complexes (LHCs) control light-dependent energy transfer in photosystem II (PSII). In order to find out if defective LHCs affect plant growth, light-related parameters were compared between a *chlorinal-1* mutant (*chl-1*; defective LHCs) and wild-type (WT) plants of *Arabidopsis thaliana*. The aim of this study was to assess the effects of LHCs on light-related parameters on the growth of *Arabidopsis* plants. A JUNIORPAM fluorometer was used to measure the parameters such as coefficients of photochemical fluorescence quenching (qp and ql); parameters of non-photochemical quenching (qn and NPQ), the yield of non-regulated energy dissipation of PSII [Y(NO)], the value of the efficient quantum yield of PSII {Y(II)}, and yield of regulated energy dissipation of PSII {Y(NPQ)}. The *chl-1* mutant showed similar coefficient of photochemical quenching to the WT plants. On the other hand, a non-photochemical quenching, an efficient quantum yield of PSII, and yield of regulated energy dissipation of PSII significantly declined in *chl-1* mutant compared with the WT plants. The *chl-1* mutant plants exhibited the value of decreased growth and smaller size of leaf compared with that of WT plants. The percentage of the area,

length and width of the leaf of the mutant declined when compared with that of WT plants. These results suggest that defective LHCs regulated growth through affecting light-related parameters of the *chl-1* mutant of *Arabidopsis thaliana* plants

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## INTRODUCTION

*Arabidopsis thaliana* is widely used to understand molecular biology of various plant traits, involving flower growth and light sensing (Más, 2005). Glutathione (GSH) controls growth and development, stomatal movement, and yield of the *Arabidopsis thaliana* plants (Jahan et al., 2008; Jahan et al., 2014; Jahan et al., 2016) and corn plants (Munirah et al., 2015a). Different stimuli such as atmospheric pollutants, biotic and abiotic stress, hormones, and light-harvesting complexes (LHCs) affect GSH content of the *Arabidopsis thaliana* plants (Sánchez-Fernández et al., 1997; Okuma et al., 2011; Jahan et al., 2016). Antenna complexes in photosystems collect and channel the photons to power the carbon-fixing reactions (Caffarri et al., 2009; Ogawa et al., 2004; Barber, 2006).

External application of GSH increased light-related parameters such as chlorophyll (Chl) content, chlorophyll fluorescence, yield, photosynthesis of corn plants (Munirah et al., 2015a; Syuhada et al., 2014; Inani et al., 2015) and leaf numbers, Chl content and fluorescence of *Arabidopsis* plants (Jahan et al., 2016). On the other hand, it was shown genetically and chemically that deficient GSH levels in guard cells affected stomatal aperture of the *Arabidopsis* plants (Jahan et al., 2016; Okuma et al., 2011). This result might limit photosynthetic activity, growth, water loss and productivity of plants (Syuhada et al., 2016; Jahan et al., 2016; Khairi et al., 2017). Recent results indicate that chlorophyll content is correlated with light and gas exchange parameters

of corn plants (Munirah et al., 2015a and b; Syuhada et al., 2014). The LHCs regulate the light reaction in photosystem to modulate the chloroplastic progress (Krol et al., 1995) and LHCs regulate physiological functions of plants (Jahan & Hasan, 2017). The *chl-1* mutant with defective LHCs in photosystem II (PSII) accepts limited photosynthetic light core complex (Ogawa et al., 2004). Therefore, the mutation of LHCs in the *chl-1* mutant had resulted in reduced leaf development and lower accumulation of GSH compared with the wild-type *Arabidopsis* plants (Jahan et al., 2016; Jahan et al., 2011). Ogawa et al. (2004) stated that GSH biosynthesis affected leaf development of *Arabidopsis* plants. The effects of GSH on Chl content, photosynthesis rate, and yield of corn plants have been documented (Munirah et al., 2015a; Syuhada and Jahan, 2016). The *chl-1* mutant showed the presence of defective LHCs in the photosystem core complexes in plants (Takabayashi et al., 2011).

In this short communication, the objective is to evaluate the function of Wild-type (WT) ecotype [Columbia (Col-0)] and *chl-1* mutant of *Arabidopsis thaliana* plants were collected from Ohio State University, USA and grown in plastic pots. A mixture of 30% peat soil and 70% vermiculite (Vermiculite, Malaysia) by volume were used in preparing a growing media. The light intensity of 80  $\mu\text{mol photon m}^{-2} \text{s}^{-1}$ , temperature of  $22 \pm 2^\circ\text{C}$ , and a day / night cycle of 16/ 8 h were maintained in the growth chamber (Jahan et al., 2016; Jahan et al., 2012). Treatments were laid out as

completely randomised design with five replications (five different plants) unless otherwise stated. Experiments were carried out from March 2016 to Oct 2016.

### Measurement of Parameters

A JUNIORPAM fluorometer (Walz, Germany) was used to measure the coefficients of photochemical fluorescence quenching [(qp and ql]; The qp is more consistent with separated light-harvesting antennae of photosystems while ql combined light-harvesting antenna to absorb photon from many reaction centres (Kramer et al., 2004)], parameters of non-photochemical quenching (qn and NPQ), the yield of non-regulated energy dissipation of PSII [Y(NO)], the value of the efficient quantum yield of PSII {Y(II)}, and yield of regulated energy dissipation of PSII {Y(NPQ)} in 5-6-week old leaves of both plants. The qn is non-photochemical quenching coefficient, whereas NPO is an alternative calculation of qn related with the number of quenching centres in the light-harvesting antenna. Data was recorded at mid-day consistently. A CI-202 portable leaf area meter (CID Bioscience, USA) was used to measure the area, length and width of leaves of both plants. The percentages of these leaf parameters of the *chl-1* mutant plants against WT plants were computed. Plants were grown at different times to determine different parameters throughout the experimental time. Plants were grown under identical conditions where different planting times did not affect the growth of *Arabidopsis*

plants. Five plants were randomly selected as replicas.

### Statistical Analysis and Accession Number

Student's t-test was used to evaluate the significance difference between mean values at  $p < 0.05$  using MS Excel software (Microsoft Corporation). The *Arabidopsis* Genome Initiative numbers for the genes discussed in this article was *CHI-1*, At1g44446.

### RESULTS

Figure 1 shows the effect of defective LHCs on light-dependent parameters of the *Arabidopsis thaliana* plants. The coefficients of photochemical fluorescence quenching (qp and ql) were found to be similar in both plants (Figure 1[a]). The parameters of non-photochemical quenching (qn and NPQ) declined in *chl-1* mutant plants compared with that of WT plants (Figure 1[b]). The qn and NPQ were 0.122 and 0.079 respectively in WT and 0.038 and 0.02 respectively in *chl-1* mutant plants. The reduction of non-photochemical quenching in the *chl-1* plants indicated that the mutant plant was more susceptible to the photoinhibition due to the defective LHCs. In addition, the yield of non-regulated energy dissipation of PSII [Y(NO)] was 0.394 in the *chl-1* mutants which is higher than that of 0.251 in the WT plants (Figure 1[c]). The value of the efficient quantum yield of PSII {Y(II)} and yield of regulated energy dissipation of PSII {Y(NPQ)} decreased in

the *chl-1* mutants compared with the WT plants (Figure 1[c]). The PSII{Y(II)} and PSII{Y(NPQ)} were 0.631 and 0.117 in WT and 0.561 and 0.045 in *chl-1* mutant plants respectively. Reduction of Y(II) in the *chl-1* mutant may be caused by a decrease of

maximum quantum yield in PSII. Picture in Figure 1[d] shows the dwarf morphological characters of *chl-1* mutants relative to that of wild-type plants. Leaves of mutant plants show lighter colour than that of WT *Arabidopsis* plants.

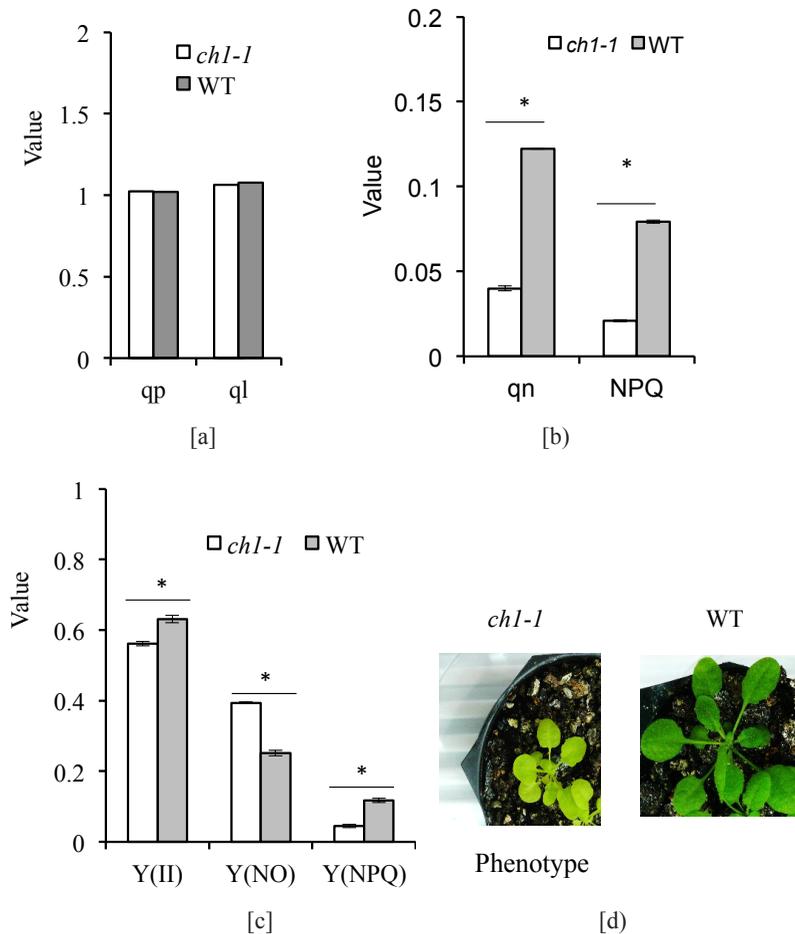


Figure 1. Different light related parameters in wild type and *chlorinal 1 (chl-1)* mutant plants of *Arabidopsis thaliana*. [a] co-efficient of photochemical fluorescence quenching (qp and ql) in WT (closed bars) and *chl-1* plants (open bars), [b] parameters of non-photochemical quenching (qn and NPQ) in WT (closed bars) and *chl-1* plants (open bars), [c] the yield of non-regulated energy dissipation of PSII [Y(NO)], the value of the efficient quantum yield of PSII {Y(II)} and yield of regulated energy dissipation of PSII {Y(NPQ)} in WT (closed bars) and *chl-1* plants (open bars), and [d] external structure, colour of leaf and leaf development between WT and *chl-1* mutant plants. Error bars represent the standard error (n=5). The asterisk indicates significant (P values  $\leq 0.05$ ) differences between plants by the horizontal line

The size and percentage of the different parameters of the leaves of both plants are shown in Table 1. The length, width, and area of the leaf of *chl-1* mutants were 1.34 cm, 1.1 cm, and 0.91 cm<sup>2</sup> respectively, significantly lower than the respective figures of 2.23 cm, 1.46 cm and 2.45 cm<sup>2</sup> of WT plants (Table 1). When the percentages of reduction of parameters of the leaf of the mutant was computed, it was found that area of leaf declined to 63% of WT. The trends were also found in length and width, 40 and 25%, respectively (Table 1). Thus, it is suggested that defective LHCs affected parameters of the leaf to suppress the growth of *chl-1* mutants of *Arabidopsis* plants.

## DISCUSSION

This study shows that defective LHCs significantly decreased qn, NPQ, Y(NO), and Y(NPQ) in the *chl-1* mutants compared with those of the WT plants (Figure 1). This might be related to the antenna function during photoinhibition. Light-dependent reaction boosted photosynthesis rate (Busch et al., 2009) and leaf development in *Arabidopsis* plants (Jahan et al., 2014; Owaga et al., 2004). Plants regulate photosynthesis process by adapting

photochemical function in the antenna complexes of photosystems (Jansson et al., 1997) to perform photosynthesis efficiently (Busch et al., 2009), which might increase the growth of *Arabidopsis* plants (Jahan et al., 2014; Table 1). Chlorophyll content and chlorophyll fluorescence are linked to the GSH content that influences the growth and yield of *Arabidopsis* plants (Jahan et al., 2014; Jahan et al., 2016). The *chl-1* mutants accumulate significantly lower amount of chlorophyll and GSH levels than those of WT *Arabidopsis* plants (Jahan et al., 2016; Jahan et al., 2011). Therefore, guard cells of the *chl-1* mutants showed higher sensitivity to abscisic acid (ABA) activity compared with the WT plants (Jahan et al., 2008; Jahan et al., 2014; Okuma et al., 2011) that limits photosynthesis rate in *chl-1* mutants. The above findings indicate that mutation of LHCs affected light-related parameters and photosynthesis rate of the plants. The mutation of LHCs reduced the gaseous movement through the smaller stomatal opening of the guard cells of the *Arabidopsis* plants (Jahan et al., 2016) and affected the growth and phenotype of the *chl-1* plants (Figure 1[d] and Table 1).

Table 1  
The area, length and width of the leaf of *chl-1* and WT plants

Type	Area (cm <sup>2</sup> )	Length (cm)	Width (cm)
<i>chl-1</i>	0.91 <sup>b</sup> ± 0.01	1.34 <sup>b</sup> ± 0.017	1.1 <sup>b</sup> ± 0.005
WT	2.45 <sup>a</sup> ± 0.03	2.23 <sup>a</sup> ± 0.01	1.46 <sup>a</sup> ± 0.012
Reduction (%) in <i>chl-1</i> against WT plants	62.8	39.9	24.6

Means + standard errors with different letters within a column were significantly different at p ≤ 0.05 by t-test

In the photosynthesis process, the light energy is converted into chemical energy (Barber, 2006). The *chl-1* mutation could cause a lower light-induced efficiency of energy in PSII due to the reduced non-photochemical quenching (qn and NPQ) in the *chl-1* plants than that of WT plants (Figure 1[b]). Different factors [including biochemical alleviation] affect plant growth through disturbing photosynthetic parameters of rice plants (Khairi et al., 2015; Hisyam et al., 2017). The NPQ is a prominent prophylactic protection strategy for the light reaction in the photosynthetic electron pathway. In the light-harvesting complexes, NPQ scatters additional excitation energy by using xanthophylls and the absorbance of the cross-section of the photosystems (Bailey et al., 2005). The NPQ and photosynthesis showed a positive correlation in plants (Schubert et al., 2006). The yield of non-regulated energy dissipation of PSII in the *chl-1* mutants was higher than that of the WT plants (Figure 1[c]) indicating that PSII used a smaller amount of light energy due to the mutation of LHCs in the *chl-1* mutant plants. This mutation confirms the higher energy fraction and photo inactivation of PSII dissipated as heat and fluorescence indicating instability despite the presence of environmental stresses (Busch et al., 2029). The mutation

of LHCs might affect the photosynthesis activity (Müller et al., 2004) and movement of guard cells of *Arabidopsis* plants (Jahan et al., 2016; Jahan et al., 2014), in which they are linked to the growth of the *chl-1* plants (Figure 1[d] and Table 1).

Previous studies have shown that GSH biosynthesis regulated the growth and flowering of the *chl-1* mutant plants (Jahan et al., 2014; Ogawa et al., 2004), which confirms the finding of this study that defective LHCs affect the growth of *chl-1* mutants (Figure 1[d] and Table 1). Deficiency of GSH increased stomatal closure of the guard cells (Okuma et al., 2011) and energy reaction in leaves of *Arabidopsis* plants (Owaga et al., 2004) that may affect photosynthesis rate and growth of the mutant plants. Moreover, Jahan et al. (2014) confirmed that the impairment of the growth of the *chl-1* mutant compared with the WT plants was due to the mutation of LHCs in plants but the authors did not discuss photo parameters in their studies. This study confirms that defective LHCs in *chl-1* mutant plants regulate light related parameters (Figure 2), which might reduce the photosynthesis rate. In conclusion, defective LHCs impair light-related parameters and GSH biosynthesis to affect the growth of the *chl-1* mutant plants (Figure 2).

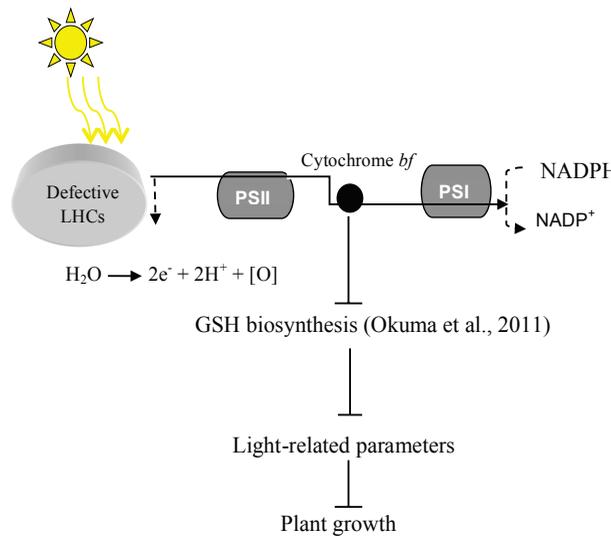


Figure 2. A schematic diagram shows mutation of LHCs affects light-related parameters and GSH biosynthesis in the *chl-1* mutant plants of *Arabidopsis thaliana*. The arrow indicates energy flow in PSII and broken arrow indicates light reaction in the *chl-1* mutant. Reverse T-bar indicates suppressive pathways. Defective light-harvesting complexes affect light-related parameters [(Chl content, Chl fluorescence, qn and NPQ, Y(NO), Y(II), Y(NPQ)] regulate GSH biosynthesis in cells of leaves to modify growth of the *Arabidopsis* plants

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