Pertanika J. Trop. Agric. Sci. 40 (4): 621 – 628 (2017)

TROPICAL AGRICULTURAL SCIENCE

Journal homepage: http://www.pertanika.upm.edu.my/

Short-term Heat Exposure Effect on *PSII* Efficiency and Growth of Rice (*Oryza sativa* L.)

Dongsansuk, A.[1]*, Theerakulpisut, P.[2] and Pongdontri, P.[3]

[1]*Department of Plant science and Agricultural resources, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand*
[2]*Department of Biology, Faculty of Science, Khon Kaen University, Khon Kaen, Thailand*
[3]*Department of Biochemistry, Faculty of Science, Khon Kaen University, Khon Kaen, Thailand*

ABSTRACT

An increase in temperature is predicted to have a negative effect on plant growth and development, thereby leading to its loss of yield. Higher air temperature has damaging effects on plant physiology, including its photosynthetic mechanism. Therefore, this study aims to investigate the effects of short-term high temperatures on photosynthesis at PSII, and the growth of rice cultivars at the heading stage. The experiment was carried out with two rice cultivars, namely, PT60 and Dular under 45ºC for 30 minutes. The results showed that panicles and leaf tips of rice cv. PT60 had chlorophyll bleaching, and at day 5 after heat treatment (DAH), the symptoms were very severe. On the other hand, all plant parts of cv. Dular remained green at 5th DAH. Fluorescence parameters in cv. Dular remained unchanged after heat treatment. In contrast, cv. PT60 exhibited a significant decrease in all fluorescence parameters. Also, the electron transport rate in cv. PT60 drastically declined after exposure to heat. The growth of cv. PT60 was inhibited by heat stress as indicated by a slight reduction in plant height, whereas cv. Dular continued growing after heat exposure. Therefore, the cultivar PT60 was susceptible to heat, and the cultivar Dular seemed to be tolerant to heat during the heading stage.

Keywords: High temperature stress, fluorescence parameter, height, rice

ARTICLE INFO

Article history:
Received: 26 September 2016
Accepted: 06 June 2017

E-mail addresses:
Danoma@kku.ac.th (Dongsansuk, A.),
piythe@kku.ac.th (Theerakulpisut, P.),
paweena@kku.ac.th (Pongdontri, P.)
* Corresponding author

ISSN: 1511-3701

INTRODUCTION

Rice (*Oryza sativa* L.) is one of the main food staple for people worldwide. However, climate change has led to an increase in global temperatures causing a reduction in yields (Baker et al., 1992). An increase in air temperature of 1^{o}C causes a 10% reduction in rice yield (Peng et al., 2004). A study showed high temperatures have an effect on phenology, physiology, cellular and molecular response of rice plants (Wahid et al., 2007). Prasad et al. (2011) suggested that high temperatures at night induced the reduction of chlorophyll content to 8% and the photosynthetic rate to 22%. When the temperature is increased by approximately 10–15°C above the normal growth temperature, photosynthetic pigments change, reducing the rate of photosynthesis . Additionally, the reduction in photosynthesis as a result of exposure to high temperatures leads to photoinhibition of *PSII* and thylakoid disorganisation. Photosystem II is very sensitive to high temperatures compared with other chloroplast functions (Martinazzo et al., 2012; Ambavaram et al., 2014). Thus, high temperatures damage the organelle membranes, leading to the loss of their functions. As a result, it affects cellular permeability and the photosystem embedded in the thylakoid membrane (Martinazzo et al., 2012). High temperatures cause Mn-stabilising proteins in the reaction centre of *PSII* to break down, which damage its D1 and D2 proteins (Wahid et al., 2007). Chlorophyll a fluorescence changing and significantly decreased *Fv/Fm* is an indicator of *PSII* efficiency (Brestic & Zivcak, 2013). Stress from high temperatures affects plant growth and development. Additionally, high temperatures can have a greater effect on the reproductive stage in rice plants by decreasing pollination, increasing sterility and decreasing the pollen viability percentage, thereby decreasing yield (Poli et al., 2013). In this study, the effects of short-term heat exposure on growth and photochemical efficiency of *PSII* were investigated in two rice cultivars differing in responses to heat stress to understand heat tolerance mechanisms of rice.

MATERIALS AND METHODS

Plant materials and stress conditions

Seeds of two rice cvs. Dular and PT60 were surface sterilised with 70% C_2H_5OH (ethanol) for 1 minute, followed by 5% NaOCl (sodium hypochlorite) for 5 minutes to protect against fungal invasiveness, and then rinsed with distilled water three times. Sterilised seeds were soaked with distilled water for 24 hours and placed on wet paper until the emergence of the shoot and radical. Seedlings were transferred to pots which contained plain soil and grown in a greenhouse at the Faculty of Agriculture, Khon Kaen University, Thailand. At the heading stage, rice plants were exposed to temperatures between 25°C and 45°C in a temperature-controlled chamber. After 30-minute exposure at 45°C, the temperature was cooled down to 25°C and then the rice plants were returned to the greenhouse. The *PSII* efficiency and growth measurements were performed at 0 (before heat exposure), 1, 2, 3, 4 and 5 days after heat exposure (DAH). Flag leaf of the same plant was used to measure *PSII* efficiency.

Determination of *PSII* efficiency

Chlorophyll fluorescence parameters were investigated before and after the plants were treated with high temperatures until the rice plants showed severe symptoms. A PAM fluorometer (Mini-PAM, Walz, Effeltrich, Germany) was used for chlorophyll fluorescence measurements. Fluorescence parameters were investigated from 8.00 to 11.00 am: basic fluorescence in the dark-adapted state (*Fo*), steady-state fluorescence under natural irradiation (*Fs*), the maximal fluorescence in the dark-adapted state (*Fm*), and the maximal fluorescence under natural irradiation (*Fm*). The maximal quantum yield of *PSII* photochemistry (*Fv/Fm*) and effective quantum yield of *PSII* photochemistry (*ΔF/Fm*) were calculated based on Schreiber (2004).

Determination of plant growth

The height of rice cvs. Dular and PT60 were measured before and after treatment with high temperatures until the rice plants showed severe symptoms. The height of rice plants was measured from the above ground surface to the tip of the flag leaf.

Statistical analysis

The experiment was a completely randomised design (CRD) with four replications. Difference of photosynthetic and growth data between the two rice cultivars were statistically analysed by using t-test method with SPSS window version 11.0.

RESULTS AND DISCUSSIONS

Plant characteristics exposed to heat were examined after a 30-min heat treatment (45^{o}C). Two previous studies have reported that rice cv. Dular showed heat tolerance during the reproductive stage (Tenorio et al., 2013; Manigbas et al., 2014). Figure 1 shows heat tolerance in all plant parts, and rice cv. Dular remained green after short-term heat treatment (45^{o}C, 30 min). In contrast, heat stress symptoms were exhibited in rice cv. PT60. Chlorophyll bleaching was found in some parts of the panicles and some leaf tips of rice cv. PT60 after a 30-min heat treatment. Additionally, rice cv. PT60 showed leaf wilting and drying symptoms (Figure 1). Moreover, severe symptoms developed after 5 DAH in cv. PT60. In contrast to cv. PT60, cv. Dular did not show any symptoms.

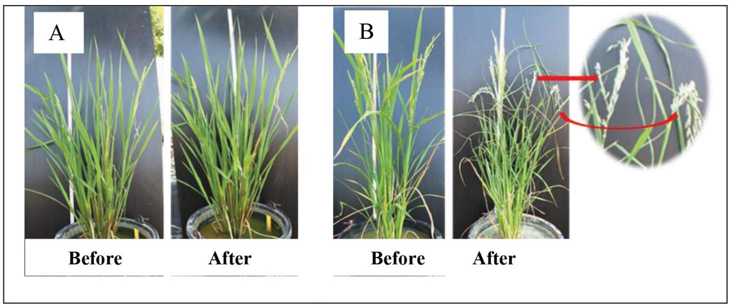

Figure 1. The photographic view of rice cvs. Dular (A) and PT60 (B) exposed to high temperature at 45^{o}C for 30 min

In general, the reproductive stage of rice was affected by elevated temperatures. Booting and flowering are the most heat-sensitive stages of rice growth (Tenerio et al., 2013). Plant physiological processes such as photosynthesis are the most sensitive to high temperature stress (Yin et al., 2010). An air temperature higher than 35°C reduced rice photosynthesis by 50% (Restrepo-Diaz & Garces-Vanron, 2013). High temperature stress influences rubisco activity, leaf chlorophyll content, maximum quantum yield of *PSII* efficiency (*Fv/Fm*), effective quantum yield of *PSII* efficiency (*ΔF/Fm'*) and non-photochemical quenching in rice plants (Cao et al., 2009; Yin et al., 2010). Chlorophyll fluorescence is light re-emitted by chlorophyll molecules during the return from an excited state to a ground state. This is useful for determining photosynthetic performance and as a non-invasive tool for studying plant responses to environmental stress (Sayed, 2003; Baker, 2008; Murchie and Lawson, 2013), including low and high temperature stress.

In the present study, chlorophyll fluorescence parameters including *Fv/Fm, ΔF/Fm', Fo, Fm, Fs and Fm',* were measured before and after exposure to heat treatment in rice cvs. Dular and PT60. Figures 2 and 3 show that all chlorophyll fluorescence parameters in rice cv. Dular remained unchanged after treatment with high temperature stress. Nevertheless, rice cv. PT60 showed a significant decline in all fluorescence parameters. The value of *Fv/Fm* in cv. Dular was in the range of 0.73–0.81. However, rice cv. PT60 exhibited *Fv/Fm* of 0.8 before heat treatment, but after heat treatment, the *Fv/Fm* values were drastically decreased within the range of 0.00–0.15. *Fv/Fm* is useful for estimating the maximum quantum yield of *PSII* photochemistry. *Fv/Fm* value of ~0.83 is markedly considered for non-stressed leaves (Baker, 2008; Murchie and Lawson, 2013). Thus, we suggested that the leaves of rice cv. PT60 were severely affected by heat exposure at 45°C for 30 min.

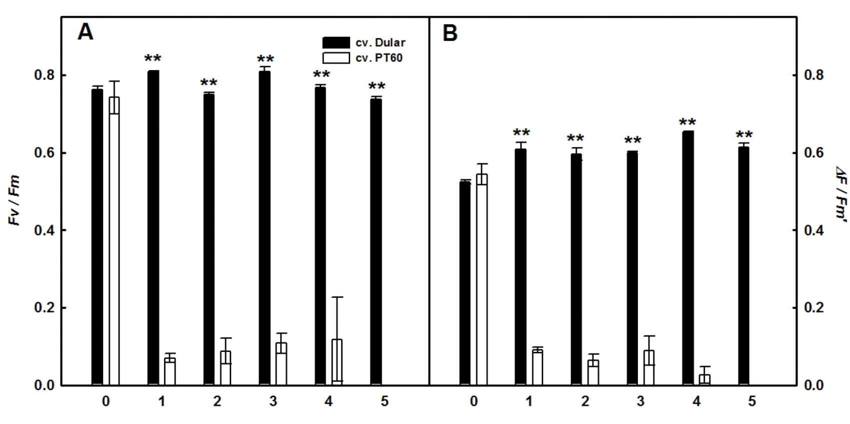

Figure 2. Fv/Fm (A) and *ΔF/Fm* (B) in rice cvs. Dular and PT60 at 0, 1, 2, 3, 4 and 5 DAH. The values are means ± SE (n=4). ** represents the means significantly different at $p \leq 0.05$

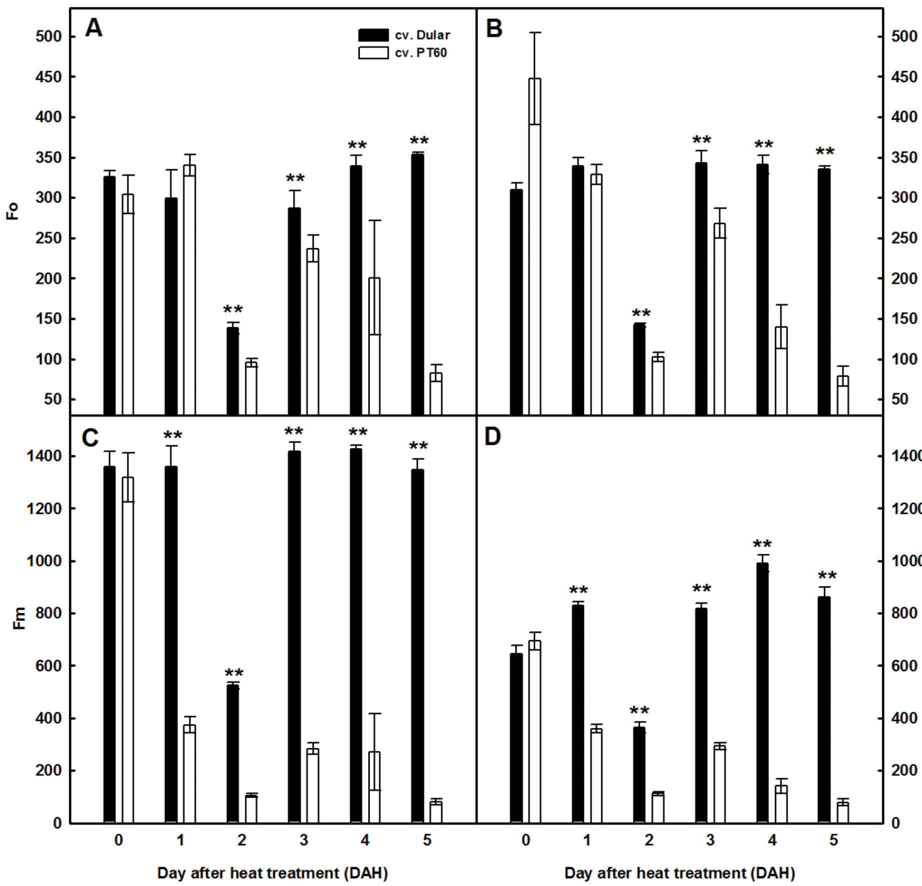

Figure 3. *Fo* (A), *Fs* (B), *Fm* (C) and *Fm* (D) in rice cvs. Dular and PT60 at 0, 1, 2, 3, 4 and 5 DAH. The values are means ± SE (n=4). ** represented the means significantly different at $p \leq 0.05$

Electron transport rate values of rice cv. PT60 markedly declined after heat treatment (Figure 4). On the other hand, cv. Dular exhibited an increasing trend in ETR values. At fifth day after heat treatment, the ETR value of cv. Dular was approximately 414.44 ± 6.33 µmol $m^{-2}s^{-1}$, in contrast to cv. PT60 in which ETR values fell to zero. The ETR can be used as an estimator of the relative photosynthetic electron transport rate (Schreiber et al., 2011) and is calculated using the *ΔF/Fm'* values, which reflect the noncyclic electron transport rate through *PSII* (Baker, 2008). This therefore suggests that the photosynthetic apparatus of cv. Dular is highly resistant to heat stress, and electron transport activities are able to completely recover 5 days after heat treatment. In contrast, rice cv. PT60 is highly sensitive to heat stress that the photosynthetic components were severely damaged, leading to a drastic reduction in *PSII* efficiency and electron transport activity. Additionally, the plant height of cv. PT60 was affected by heat stress, but cv. Dular still had an increase in height after heat treatment.

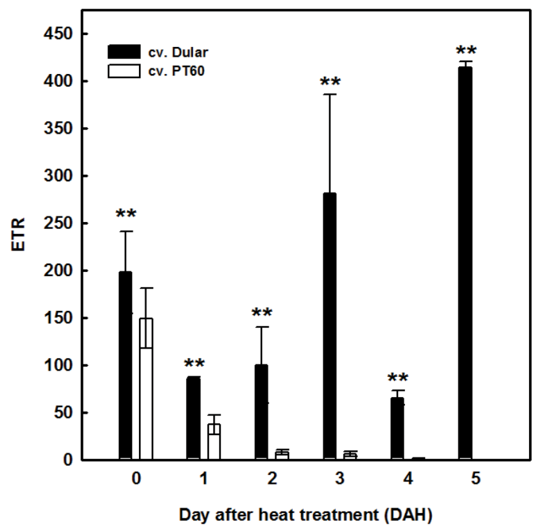

Figure 4. ETR in rice cvs. Dular and PT60 at 0, 1, 2, 3, 4 and 5 DAH. The values are means ± SE (n=4). ** represented the means significantly different at p≤0.05

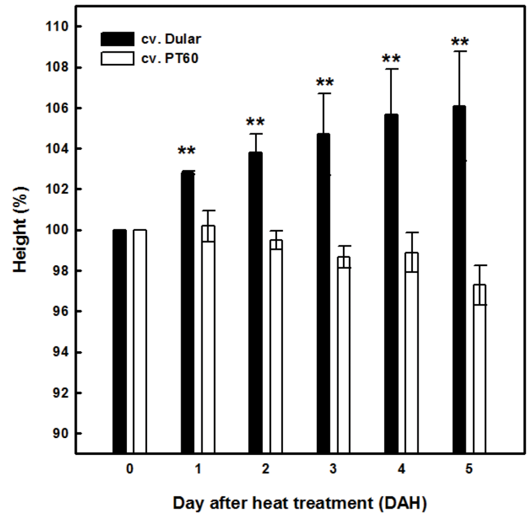

Figure 5. Height in rice cvs. Dular and PT60 before and after heat exposure. The values are means ± SE (n=4). ** represented the means significantly different at p≤0.05

CONCLUSIONS

This experiment showed that short-term high temperature stress during the heading stage affected leaf photosynthesis of *PSII* efficiency. PT60 was susceptible to high temperatures as indicated by the bleaching of chlorophyll, decrease in quantum yield of *PSII* efficiency and lack of growth, whereas the cultivar Dular was high temperature tolerant, as indicated by its green colour and high photosynthetic efficiency of *PSII*.

ACKNOWLEDGEMENT

The authors thank Agriculture Research Funding, Faculty of Agriculture, KKU and Salt Tolerance Rice Research Group, KKU for their research support. The rice seedlings were provided by Biotechnology research and development office, Thailand.

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