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Characterization and evaluation of response to heat and chilling stress in exotic weeds using chlorophyll a fluorescence OJIP transient

Soo In Sohn, Yong Ho Lee[1], Sun Hee Hong[1], Chang Seok Kim[2], Myung Hyun Kim, Chae Sun Na[3] and Young Ju Oh[4,*]

National Institute of Agricultural Sciences, RDA, Jeonju 54874, Republic of Korea
[1]Plant Life & Environmental Science, Hankyong National University, Anseong 17579, Republic of Korea
[2]National Institute of Crop Science, RDA, Pyeongchang 5481, Republic of Korea
[3]Seed Conservation Research Division, Baekdudaegan National Arboretum, Bonghwa 36209, Republic of Korea
[4]Institute for Future Environmental Ecology Co., Ltd., Jeonju 54883, Republic of Korea

***Corresponding author**
Young Ju Oh
Tel. 063-211-7122
E-mail. cave50joo@gmail.com

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Abstract: The occurrence of exotic weeds and their influx into farmlands due to climate change poses many problems. Therefore, it is necessary to generate a prediction model for the occurrence pattern of these exotic weeds based on scientific evidence and devise prevention measures. The photosynthetic apparatus is known as the most temperature-sensitive component of a plant cell and its initial response to temperature stress is to inhibit the activation of photosystem II. This study investigated the potential of OJIP transients in assessing temperature stress in exotic weeds. The four exotic weeds currently flowing into Korean farmlands include *Amaranthus spinosus*, *Conyza bonariensis*, *Crassocephalum crepidioides*, and *Amaranthus viridis*. These weeds were treated at 5°C, 10°C, 15°C, 20°C, 25°C, 30°C, 35°C, and 40°C and the OJIP curves and JIP parameters were measured and analyzed. The results showed that heat and chilling stress affected the photosystem II (PSII) electron transport of *A. spinosus*, whereas *C. crepidioides* and *A. viridis* were more affected by high-temperature stress than by low-temperature stress. Lastly, *C. bonariensis* showed resistance to both high and low-temperature stress. The results of this study suggest that OJIP transients and JIP parameters can be used to analyze damage to the photosynthetic apparatus by temperature stress and that they can serve as sensitive indicators for the occurrence pattern of exotic weeds.

Keywords: OJIP, exotic weed, photosystem, heat stress, chilling stress

INTRODUCTION

According to a report by the Intergovernmental Panel on Climate Change, the average temperature of the earth is predicted to rise by 0.3°C every 10 years due to global warming (IPCC 2007), and the United Nations (UN) estimated that the average temperature will rise by 1.4–5.78% over the century (Tkemaladze and Makhashvili 2016). Global warming has a direct impact on plants by altering their photosynthetic nature as well as their growth and development periods, making it a problem across the globe due to its association with food production (Qaderi and Reid 2009). These climate changes also influence the occurrence pattern of exotic weeds and many studies have reported that exotic weeds adapt to climate changes faster and become dominant species over the native plants (Bradley *et al.* 2010). Harmful exotic weeds not only deteriorate the production and quality of crops but also destroy the

plant ecosystem and even pose threat to humans by allergic reactions to exotic weed pollen (Cariñanos and Casares-Porcel 2011). Therefore, it is necessary to predict the occurrence pattern of exotic weeds according to climate changes and devise prevention and/or control methods.

The photosynthetic apparatus is the most temperature-sensitive component of a plant cell and the initial response to temperature stress is the inhibition of the activation of photosystem II (PSII) (Salvucci and Crafts-Brandner 2004). Many studies have been conducted and the analysis of chlorophyll fluorescence has been commonly used in studying plant responses to temperature stress conditions (Ibaraki and Marakami 2006; Haque *et al.* 2014). The fluorescence emission upon light illumination is not continuous, but a rapid increase is followed by a decrease to a steady level (Kautsky and Hirsch 1931). When the fluorescence value during the rapid rise is plotted on a logarithmic scale across time, different phases become evident (Strasser and Govindjee 1992). Based on the incidence of these different phases, Strasser *et al.* (2000) described and developed concepts for the changes in the rise kinetics and amplitudes of these phases under stress conditions. A formula to calculate a series of parameters based on these concepts was devised, which is now known as the JIP test.

The typical fast chlorophyll fluorescence increase kinetics display a series of phases, spanning an increase from the initial (F_O) fluorescence value to the maximal (F_M) fluorescence value. The series are step O (20 μs, all reaction centers (RCs) open), J (~2 ms), I (~30 ms), and P (equal to F_M when all RCs are closed) (Strasser 1995). Other steps occur under specific conditions other than these O-J-I-P steps, which are the L-step (reflecting the energetic connectivity of the PSII units), the K-step (reflecting the inactivation of the oxygen-evolving complex; OEC), and the H-step and G-step, which occur in coral or foraminifers (Tsimilli-Michael *et al.* 1999; Strasser *et al.* 2004). For example, the K-step is a rapid additional step that occurs between 200−300 μs when the sample is under heat or drought stress. Nitrogen deficiency has also been shown to induce the K-step, H-step, and G-step (Strasser *et al.* 2004). In contrast, one or two steps of the O-J-I-P steps disappear under some stress conditions. In PSII-herbicide treated samples, the J-step quickly increased to the P level and the I and P phases disappeared. This resulted from the action of the herbicide to block the delivery of electrons from the primary quinone electron acceptor of PSII (Q_A^-) to the secondary quinone electron acceptor of PSII (Q_B^-), causing it to accumulate in the Q_A of the PSII RCs (Strasser *et al.* 2004; Tóth *et al.* 2005a; Chen *et al.* 2007). Thus, the OJIP curve can be analyzed to obtain various quantitative data such as the change in energy flow and reduction in the electron acceptance by PSII under environmental stress (Strasser *et al.* 2000). However, it is yet unclear whether OJIP transients can be used as indicators of temperature stress for exotic weeds and how the photosynthetic physiological responses change for these exotic weeds under temperature stress conditions.

Thus, this study aimed to investigate how different temperatures affected the electron transport system of the photosystem using OJIP transients in an attempt to predict the occurrence pattern of exotic weeds *Amaranthus spinosus*, *Conyza bonariensis*, *Crassocephalum crepidioides*, and *Amaranthus viridis* currently flowing into the farmlands due to climate changes.

MATERIALS AND METHODS

1. Plant materials and temperature treatment

Amaranthus spinosus, *Conyza bonariensis*, *Crassocephalum crepidioides*, and *Amaranthus viridis* seeds were sown on vermiculite and germinated in a 25°C incubator for four weeks. These plants were used for chlorophyll a fluorescence OJIP transient analysis. The exotic weed seeds were provided by the Korea University Wild Resource Plant Seed Bank.

The effect of temperature change on the OJIP transients of each exotic weed was analyzed. Each exotic weed plant was treated in a dark incubator for 30 minutes at 5°C, 10°C, 15°C, 20°C, 25°C, 30°C, 35°C, and 40°C and chlorophyll fluorescence measurements were performed. Each treatment was carried out in three replicates. The control group was treated at 25°C.

2. Chlorophyll a fluorescence measurements

Chlorophyll a fluorescence was measured on a leaf using a hand fluorometer (FluorPEN FP 100; Photon System Instrument, Czech Republic). The leaves were exposed to a pulse of saturating light at a wavelength of 650 nm and 2700 μmol m^{-2} s^{-1} intensity for five seconds after becoming dark-adapted. The fluorescence transients were recorded at a 12-bit resolution between 10 μs and 5 s (Strasser *et al.* 2000). FluorPen software (PSI, CZ) was used to load the full fluorescence transients and calculate the O-J-I-P parameters from the variable fluorescence values at F50

Table 1. Short description of the chlorophyll fluorescence parameters used in the study according to the OJIP test

Abbreviation	Mathematical expressions	Description
Extracted and technical fluorescence parameters		
F_0	F_0	Initial fluorescence at 50 μs from dark-adapted tissue
F_K	F_K	Fluorescence intensity at 300 μs from dark-adapted tissue
F_J	F_J	Fluorescence intensity at 2 ms from dark-adapted tissue
F_I	F_I	Fluorescence intensity at 30 ms from dark-adapted tissue
F_M	F_M	Maximum fluorescence at 500 ms from dark-adapted tissue
Fluorescence parameters derived from the extracted data		
Fv/Fm	$(F_M-F_0)/FM$	Maximum yield of primary photochemistry
V_J	$(F_J-F_0)/(F_M-F_0)$	Relative variable fluorescence at the J-step
V_I	$(F_I-F_0)/(F_M-F_0)$	Relative variable fluorescence at the I-step
M_0	$4\cdot(F_K-F_0)/(F_M-F_0)$	Slope at the beginning of the transient Fo → Fm, maximal fractional rate of photochemistry
Quantum efficiency or flux ratios		
$\psi_0(ET_0/TR_0)$	$1-V_J$	Probability of a trapped exciton moving an electron beyond φ_{P0} - maximum quantum yield of primary photochemistry (Q_A^-)
$\varphi_{E0}(ER_0/ABS)$	$(1-F_0/F_M)\cdot\psi_0$	Probability of an absorbed exciton moving an electron beyond Q_A^-
$\varphi_{P0}(TR_0/ABS)$	$1-F_0/F_M = F_V/F_M$	Maximum yield of primary photochemistry, equal to Fv/Fm
$\delta_{R0}(RE_0/E_{T0})$	$(1-V_I)(1-V_J)$	Efficiency with which an electron can move from the reduced intersystem electron acceptors to the photosystem I(PSI) end electron acceptors
$\varphi_{R0}(RE_0/ABS)$	$\varphi_{P0}\cdot\psi_0\cdot\delta_{R0}$	Quantum yield of electron transport from Q_A^- to the PSI end electron acceptors
$\rho_{R0}(RE_0/TR_0)$	$\psi_0\cdot\delta_{R0}$	Efficiency with which a trapped exciton can move an electron into the electron transport chain from Q_A^- to the PSI end electron acceptors
Density of reaction centers		
RC/CS	$\varphi_{P0}\cdot(V_J/M_0)\cdot ABS/CS$	Density of active reaction centers per cross section
Vitality indexes		
PI_{abs}	$(RC/ABS)\cdot[\varphi_{P0}/(1-\varphi_{P0})]\cdot[\psi_0/(1-\psi_0)]$	Performance index (PI) on an absorption basis
Specific fluxes or specific activities per reaction center (RC)		
ABS/RC	$M_0\cdot(1/V_J)\cdot(1/\varphi_{P0})$	Absorption flux per RC
TR_0/RC	$M_0\cdot(1/V_J)$	Trapped energy flux per RC (at t=0)
ET_0/RC	$M_0\cdot(1/V_J)\cdot\psi_0$	Electron transport flux per RC (at t=0)
DI_0/RC	$(ABS/RC)-(TR_0/RC)$	Dissipated energy flux per RC (at t=0)
RE_0/RC	$(RE_0/ET_0)\cdot(ET_0/RC)$	Reduction of end acceptors at PSI electron acceptor side per RC (at t=0)
Phenomenological fluxes or phenomenological activities per excited cross section (CS)		
ABS/CS	F_M	Absorption flux of photons per cross section
TR_0/CS	$\varphi_{P0}\cdot(ABS/CS)$	Trapping of electrons per cross section
ET_0/CS	$\varphi_{P0}\cdot\psi_0\cdot(ABS/CS)$	Electron flux per cross section
DI_0/CS	$(ABS/CS)-(TR_0/CS)$	Energy dissipation per cross section

μs, F100 μs, F300 μs, F2 ms, and F30 ms according to the OJIP test formula (Strasser and Govindjee 1992; Strasser and Tsimili-Michael 2001). The expressed specific fluxes (ABS/RC; TR_0/RC; DI_0/RC; ET_0/RC) per reaction center and phenomenological flux (ABS/CS; TR_0/CS; DI_0/CS; ET_0/CS) per cross-section were derived from

the theory of the biomembrane energy flux and was calculated from the O-J-I-P test (Sironval *et al.* 1981; Strasser and Tsimili-Michael 2001). Absorbance (ABS) refers to the absorption of the photons by the chlorophyll molecule within the antenna complex. Part of the absorbed energy is trapped (TR_0) by the reaction center of PSII (P680) while the rest is lost in the form of heat or fluorescence (DI_0). Part of the trapped energy is changed to redox energy by electron transport (ET_0) through Q_A and Q_B (Strasser *et al.* 2000). The JIP parameters were determined according to the formula shown in Table 1.

3. Statistical analysis

The parameters of the JIP test were log-transformed (different temperature treatment/control (25°C)) for evaluation of the relative changes. The performance index PI_{abs} were subjected to an analysis of variance (ANOVA). When the F-test was significant, a comparison of the means was performed with Dunnett's test. Statistical analyses were performed using SAS 9.4 (SAS Institute Inc., Cary, NC, USA).

RESULTS

1. OJIP curve

A. spinosus had a relatively higher F_O at 5°C and 40°C temperature conditions compared to other temperatures and showed low fluorescence density F_I and F_P at the I and P stages (Fig. 1a). The results showed that the OJIP curves at temperatures of 15°C, 20°C, 30°C, and 35°C were not significantly different from those at 25°C. For *Conyza bonariensis*, the F_P values showed a tendency to slightly decrease with higher temperature treatments, but fluorescence density was not significantly different at each stage under all temperature treatment conditions (Fig. 1b). *C. crepidioides* also showed a higher F_O at 40°C heat stress conditions like that of *A. spinosus* and the F_I and F_P fluorescence density at the I and P stages showed a relatively rapid decrease compared to other temperature treatments. In contrast, the fluorescence density at the J, I, and P stages was higher at 5°C cold stress conditions compared to that at 25°C (Fig. 1c). The results for *A. viridis* showed a higher F_O at 40°C compared to the other temperatures, and the F_I

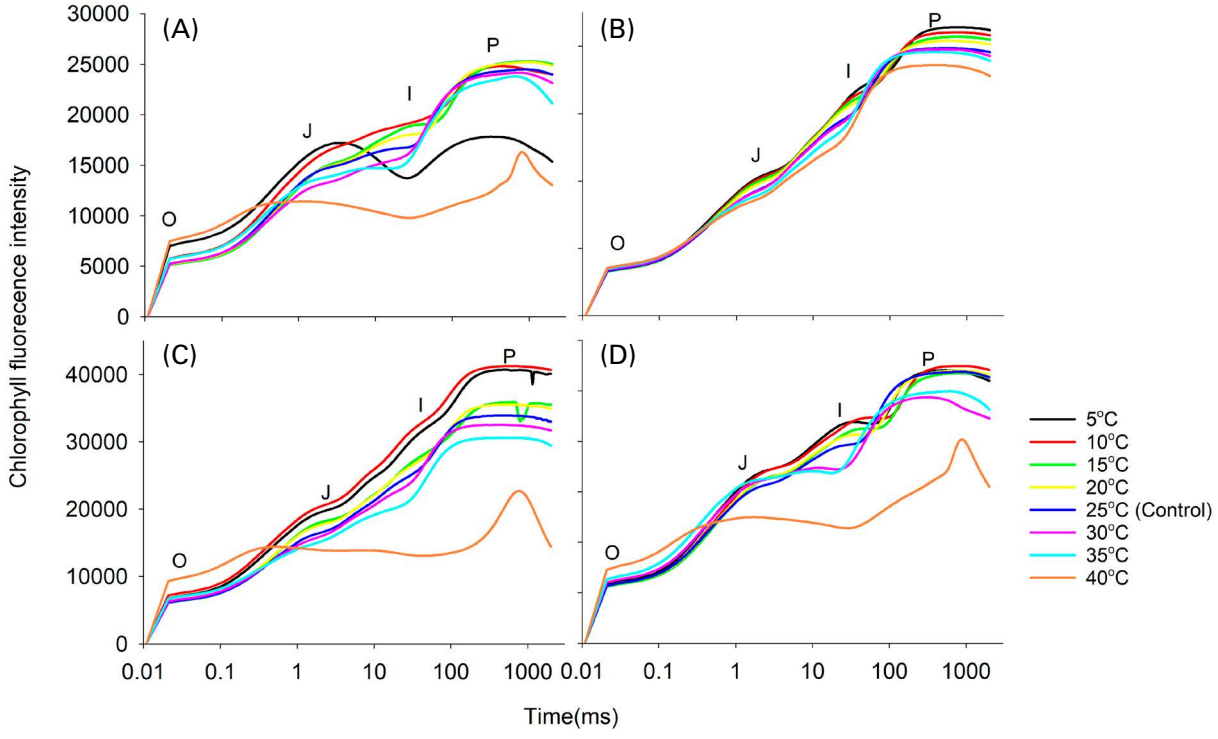

Fig. 1. Chlorophyll a fluorescence OJIP transient curves from 4-week leaves of *Amaranthus spinosus* (A), *Conyza bonariensis* (B), *Crassocephalum crepidioides* (C), and *Amaranthus viridis* (D) under different temperatures. O, K, J, I, and P in the transient curves indicate minimal fluorescence intensity when all PSII reaction centers are open, the intensity at 300–400 μs, the intensity at 2 ms, the intensity at 30 ms, and the maximal intensity when all PSII RCs are closed, respectively.

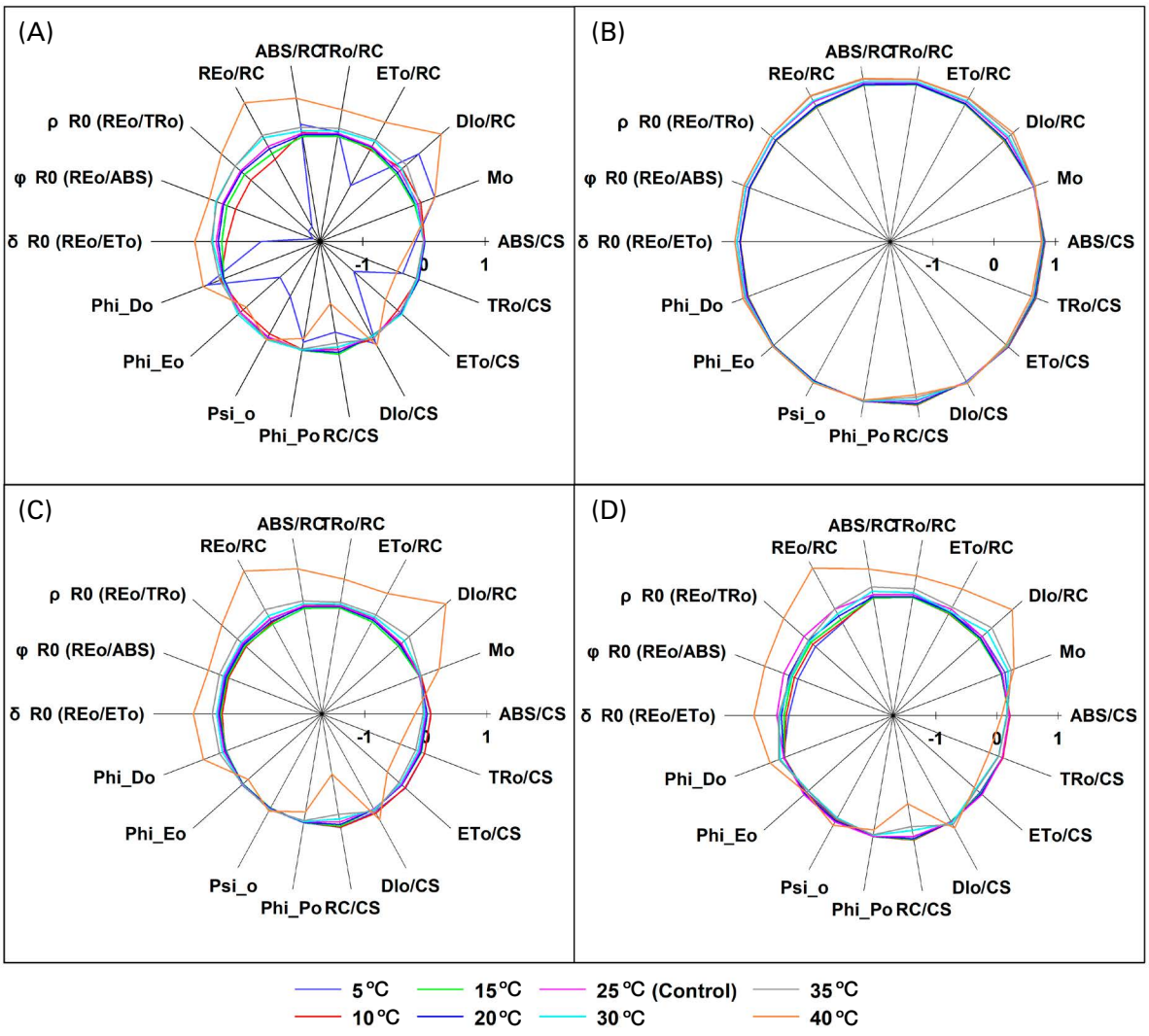

Fig. 2. Spider plots of JIP parameters deduced from chlorophyll a fluorescence OJIP transient curves of 4-week leaves of exotic weeds under different temperatures. Extracted and technical fluorescence parameters from *Amaranthus spinosus* (A), *Conyza bonariensis* (B), *Crassocephalum crepidioides* (C), and *Amaranthus viridis* (D). For each parameter, the value at 25°C was set as 1.0 and log-transformed.

and F_P fluorescence density rapidly decreased at the I and P stages. The fluorescence density for each stage of the 5°C and 10°C cold stress conditions was similar to the fluorescence density at each stage at 25°C. Treatment at 30°C and 35°C showed lower fluorescence densities at the I and P stages compared to the 25°C treatment (Fig. 1c).

2. Change in chlorophyll fluorescence parameter

The fluorescence parameters were calculated from the OJIP analysis of exotic weeds (Fig. 2). Most of the parameters for *A. spinosus* at treatments of 5°C and 40°C were different from those at 25°C. Under 5°C temperature conditions, the PSII parameters φ_{EO}, ψ_O, and φ_{PO} and all phenomenological fluxes per CS including ET_O/CS, TR_O/CS, and ABS/CS, except for DI_O/CS, were found to be lower compared to those at 25°C. The photosystem I (PSI) parameters including RE_O/RC, ρ_{RO}, φ_{RO}, and δ_{RO}, and the specific fluxes ET_O/RC were lower than those at 25°C. In contrast, M_O, the specific fluxes, PSI parameters, and ψ_O of PSII were higher at 40°C compared to those at 25°C. However, φ_{EO} and φ_{PO} of the PSII parameters and all phenomenological fluxes per CS including ETo/CS, TRo/CS, and ABS/CS, except for DI_O/CS, were lower compared to those at 25°C. For *C. bonariensis*, the PSI parameter values

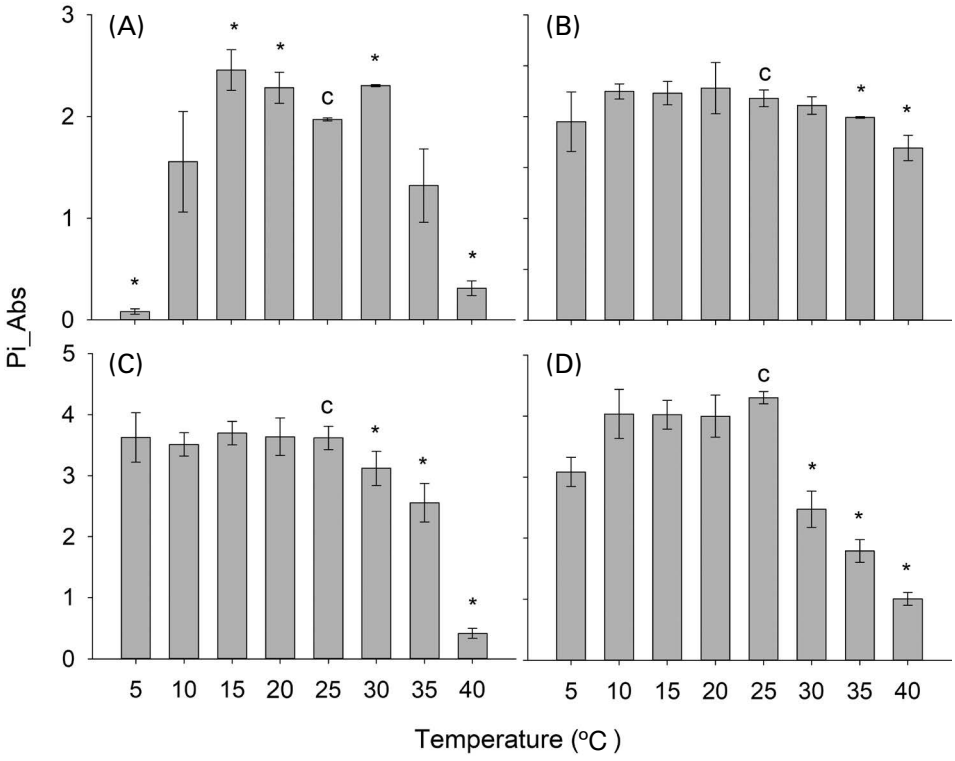

Fig. 3. Performance index (PI_{Abs}) in the exotic weeds exposed to 5°C, 10°C, 15°C, 20°C, 25°C, 30°C, 35°C, and 40°C for 30 min. *Amaranthus spinosus* (A), *Conyza bonariensis* (B), *Crassocephalum crepidioides* (C), and *Amaranthus viridis* (D). Asterisks indicate significant differences from the controls (25°C) at $p<0.05$ by Dunnett's multiple comparison test.

of RE_O/RC, ρ_{RO}, φ_{RO}, and δ_{RO} were relatively lower at 5°C, 10°C, 15°C and 20°C compared to those at 25°C, whereas these values were 1.3-, 1.1-, 1.1- and 1.1-fold higher, respectively, at 40°C compared to 25°C. *C. crepidioides* showed no significant difference in the parameters between treatments at 5°C and 25°C, whereas M_O, the specific fluxes, and PSI parameters were higher at 40°C treatment compared to those at 25°C. However, the PSII parameters φ_{EO} and φ_{PO}, as well as phenomenological fluxes were lower compared to those at 25°C. For *A. viridis*, the PSI parameters at 5°C, 10°C, 15°C, and 20°C were lower compared to those at 25°C. M_O, the specific fluxes, and PSI parameters were higher at 40°C compared to those at 25°C, yet φ_{EO} and φ_{PO} of the PSII parameters and all phenomenological fluxes including ET_O/CS, TR_O/CS, and ABS/CS, except for DI_O/CS, were higher.

3. Comparative analysis of plant vitality

The PI_{abs}, indicating the vitality of the plant, was lower in *A. spinosus* at 5°C and 40°C temperature treatments compared to the other temperatures (Fig. 3). The PI_{abs} for *C. bonariensi* decreased as the temperature increased. In *C. crepidioides*, the PI_{abs} was significantly lower at 40°C treatment conditions compared to 25°C. The PI_{abs} for *A. viridis* decreased as the temperature increased, similar to *C. bonariensis*.

DISCUSSION

1. OJIP curve

A. spinosus at 5°C and 10°C temperature conditions showed a greatly increased fluorescence intensity at 2 ms compared to that at 25°C. This was likely the result of the accumulation of a reduced Q_A^- pool, which led to a decrease in the delivery of electrons across Q_A^- (Strasses *et al.* 1995; Lazár and Ilík 1997; Haldimann and Strasser 1999). The same observation was made in two genotypes of soybean, PAN809 and Sonop, where the dark chilled samples had greatly increased fluorescence intensity com-

pared to the non-treated controls at 2 ms (J-peak) (Strauss *et al.* 2006). In this study, *A. spinosus* treated at 40°C heat stress conditions had a higher F_O value compared to that at other temperatures, and the K peak was found at 300 μs in the OJIP curve. Also, fluorescence densities F_J, F_I, and F_P decreased at the J, I, and P stages. F_O refers to the fluorescence emission when all primary quinone-type acceptors (Q_A) of the reaction center are in the oxidized state (Rodríguez *et al.* 2015). A significant increase in F_O under high-temperature conditions was the result of a partial dissociation of the outer antenna from the rest of the PSII or a shift in the equilibrium between the electron acceptors Q_A and Q_B and thus, an increase in the reverse transfer of electrons from Q_B to Q_A. In this state, Q_A was partially reduced in the dark state and the O-step no longer indicated the F_O (Srivastava *et al.* 1997). At the high-temperature (40°C) treatment of *A. sponisus*, F_K was increased and the I-step disappeared, which seemed to be the result of an inhibition of electron transfer within PSII to the secondary electron donor due to the inactivation of the oxygen-evolving complex (OEC) (Guisse *et al.* 1995; Strasser 1997; Chen *et al.* 2016). The K-peak is known to arise from the accumulation of Q_A^- from the stable charge separation and the absence of an electron donor is known to result in the accumulation of $P680^+$ (Toth *et al.* 2007). Commonly, the presence of the K-peak is known as an important change in the OJIP rise kinetics under heat treatment but is also used as a heat indicator in predicting the plant response to high temperature (Srivastava and Strasse 1995; Lu and Chang 1999; Oukarroum *et al.* 2013). In wheat (*Triticum aestivum* L.), F_K was absent in leaves grown at 25°C but leaves exposed to a high temperature of 45°C showed decreases in the F_J, F_I, and F_P values with a distinct presence of F_K. It was also reported that F_K steadily increased as the temperature increased above 37.5°C (Lu and Zhang 2000; Mathur *et al.* 2011). Based on these previous results, it seems *A. spinosus* was affected by both chilling and heat stress. *C. bonariensis* showed no significant difference in fluorescence intensity under both chilling stress (5−10°C) or heat stress (35−40°C) conditions compared to 25°C, indicating it was more resistant to temperature stress conditions than the other exotic weeds. *C. crepidioides* seems to be more affected by heat stress compared to cold stress where the presence of the K-step, the missing I-step, and decreases in fluorescence intensity in the J and P steps indicated the inhibition of electron transfer as a result of damage to the donor side of PS II. Like *C. crepidioides*, *A. viridis* was found to have a K-step, a missing I-step, and the fluorescence intensity decreased at the J and P steps, which indicated it was also more affected by heat stress than chilling stress.

F_M represents the maximal fluorescence intensity, and a decrease in F_M and an increase in F_O indicate the inhibition of the delivery of electrons from OEC to the PQ pool (Havaux 1993; Yamane *et al.* 2000). When the F_M values at different temperature treatments were comparatively analyzed across the exotic weeds, the results showed that they were more sensitive to heat stress compared to chilling stress and the photochemical efficiency of PS II was found to be lowest at the 40°C treatments of *A. spinosus* and *C. crepidioides*.

2. Change in chlorophyll fluorescence parameter

M_O represents the initial fluorescence slope (Oh *et al.* 2014). Increases in M_O indicate a greater reduction of Q_A to Q_A^- and a net rate increase in RC closure (Zushi *et al.* 2012). *A. spinosus* showed a 2-fold higher M_O value at 5°C and 40°C compared to 25°C. *C. bonariensis* showed no significant differences at 5°C and 40°C compared to 25°C. *C. crepidioides* was 2.2-fold higher at 40°C, and 1.1-, 1.3-, 1.4-, and 1.6-fold higher at 5°C, 30°C, 35°C, and 40°C, respectively, compared to 25°C. Previous reports on heat stress treatments of pea (*Pisum sativum* L.) leaves, apple (*Malus domestica* Borkh.) and tomato (*Solanum lycopersicon* cv. Muni Carol) leaves, and fruit peel at temperatures higher than 40°C also showed an increase in M_O (Strasser *et al.* 2000; Chen and Cheng 2009; Zushi *et al.* 2012).

The JIP parameters and energy pipeline model of tomato leaves and fruit juice suggest several sensitive sites of PSII that respond to heat stress (Zushi *et al.* 2012). For example, decreases in ABS/CS_M of the phenomenological flux per CS_M indicate an increased density of the inactive reaction centers. Also, decreases in TR/CS_M and ET_O/CS_M indicate the transition of active RCs to inactive RCs, which represents a decrease in trapping efficiency and PSII activity. In other words, decreases in ABS/CS, TR_O/CS, ET_O/CS due to chilling and heat stress are indications that the light energy absorbed per leaf area, energy captured in PSII, and the energy transferred by electron transfer are all decreased. On the contrary, increases in DI_O/CS as temperature increases indicate an increase in energy not being used for electron transfer. In the case of *A. spinosus*, ABS/CS, TR_O/CS, and ET_O/CS at 5°C temperature conditions decreased 73%, 55%, and 10%, respectively, compared to the results at 25°C. At 40°C temperature conditions, the

values decreased 66%, 44%, and 50%, respectively, compared to those at 25°C. In contrast, DI_O/CS was found to increase by 133% and 143% at 5°C and 40°C temperature conditions, respectively, compared to that at 25°C. For *C. bonariensis*, the results showed relatively small changes in these parameters under both heat and chilling stress. *C. crepidioides* showed a decrease in the ABS/CS, TR_O/CS, and ET_O/CS values and an increase in DI_O/CS under 40°C heat stress, suggesting that it was more sensitive to heat stress compared to chilling stress. *A. viridis* showed a result similar to that of *C. crepidioides*.

Φ_{Po}, ψ_o, and φ_{Eo}, which represent the quantum efficiency of PSII, decreased under heat stress conditions in Chinese cabbage, which indirectly indicated an increase in inactive reaction centers and that most of the captured energy was not being used to transfer electrons when the plant was under high-temperature stress (Oh *et al.* 2014). The Fv/Fm value of dark-adapted leaves, Φ_{Po}, represents the maximum potential photosynthetic value of the leaf, and the healthy leaves of most plants commonly have a value of around 0.83 (Bjorkman and Demmig 1987; Johnson *et al.* 1993). The value will be lower when the plant has been exposed to stress. *A. spinosus* had a Φ_{Po} value of 0.58 at 5°C, 0.78 at 25°C, and 0.51 at 40°C treatment conditions. This suggests that *A. spinosus* was affected by both chilling and heat stress. *C. bonariensis* had a value of 0.84 at 5°C, 0.82 at 25°C, and 0.80 at 40°C treatment conditions. *C. crepidioides* had a value of 0.82 at 5°C, 0.81 at 25°C, and 0.56 at 40°C treatment conditions. Lastly, *A. viridis* had a value of 0.77 at 5°C, 0.78 at 25°C, and 0.61 at 40°C treatment conditions. Thus, these results suggest that *C. bonariensis* was resistant to both chilling and heat stress, *C. crepidioides* was resistant to chilling stress but sensitive to heat stress, and *A. viridis* was affected by both chilling and heat stress.

Specific fluxes per RC were found to increase with temperature across all exotic weeds. An increase in the average ABS/RC, TRo/RC, and DIo/RC per active RC was seen due to the inactivation of several RCs, which also suggests an increase in the total dissipation ratio to the active RCs as a result of the high dissipation of inactive RCs (Zushi *et al.* 2012).

The PSI and PSII responses to chilling stress and heat stress were different according to each exotic weed. It is widely accepted that PSII is the most thermally labile component of the photosynthetic apparatus, whereas PSI is relatively resistant to heat stress (Havaux *et al.* 1991; Apostolova and Dibrikova 2011). In accordance with this, most PSII parameters were lower under 40°C temperature treatment compared to those at 25°C, while most PSI parameters were relatively higher compared to 25°C for all the exotic weeds used in this study. PSI is known to be more sensitive to chilling stress compared to PSII (Zushi *et al.* 2012). The results of this study showed that for *C. bonariensis* and *A. viridis*, but not for *A. spinosus* and *C. crepidioides*, the PSII parameters under chilling stress treatment conditions were not different than those at 25°C, while the PSI parameters under chilling stress treatment conditions were lower compared to those at 25°C.

3. Performance index

Of the fluorescence parameters, the PI_{abs} indicates the vitality of the plant. *A. spinosus* had significantly lower PI_{abs} values at 5°C and 40°C temperature treatments compared to those at other temperatures. At a 5°C treatment temperature, the PI_{abs} decreased by 3% compared to that at 25°C, whereas at 40°C, the PI_{abs} decreased by 12% compared to the 25°C treatment. This suggests that *A. spinosus* was sensitive to damage by chilling stress and heat stress. The results for *C. bonariensis* showed no significant difference between the 5°C and 25°C treatments, but at 40°C treatment conditions, the PI_{abs} decreased by 81% compared to that at 25°C. This suggests that *C. bonariensis* was more sensitive to heat stress compared to chilling stress, but was the least sensitive to heat stress of all the exotic weeds studied. *C. crepidioides* showed significant differences between the 5°C and 25°C treatments, whereas, at 40°C treatment conditions, the PI_{abs} decreased by 11% compared to that at 25°C, suggesting that it was more sensitive to damage by heat stress. The results for *A. viridis* showed that at a treatment temperature of 5°C, the PI_{abs} decreased by 78%. At a 40°C treatment temperature, the PI_{abs} decreased by 26%, which suggests that *A. viridis* was more sensitive to heat stress than to chilling stress.

This study used chlorophyll a fluorescence OJIP transients to investigate the effect of temperature stress on the photosynthetic apparatus of exotic weeds to predict the occurrence pattern of exotic weeds that are currently flowing into Korean farmlands based on climate changes. It also aimed to assess the potential of OJIP transients and the calculated JIP parameters for use as sensitive methods to detect damage to the photosynthetic apparatus caused by temperature stress. Differing results were found regarding how temperature stress conditions affected the photosynthetic apparatus in exotic weeds. The results of this study indicate that from the range of temperatures studied, chill-

ing stress conditions (5°C and 10°C) and heat stress conditions (40°C) had the greatest impact on the photosynthetic apparatus of the exotic weeds. All of the exotic weeds used in the study had lower PSII parameters at 40°C temperature treatment compared to those at 25°C, whereas PSI parameters were all relatively higher compared to those at 25°C. The results suggested that PSI was more sensitive to chilling stress than PSII.

OJIP transients were found to be an efficient and non-destructive tool to easily and quickly assess the occurrence pattern of exotic weeds by providing a description of the changes in the photosynthetic apparatus of exotic weeds upon temperature stress. This study was the first study to assess the potential of OJIP transients for use as a system to predict the occurrence pattern of exotic weeds that result from climate changes. This study looked at four exotic weed species and further studies conducted on other weed species using the same methods may be effective in predicting the occurrence pattern of exotic weeds due to climate change and devising prevention methods.

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REFERENCES

Apostolova EL and AG Dobrikova. 2011. Effect of high temperature and UV-A radiation on the photosystem II. pp. 577–591. In: Handbook of Plant and Crop Stress (Pessarakli M, ed.). CRC press. Boca Raton, London, New York.

Björkman O and B Demmig. 1987. Photon yield of O_2 evolution and chlorophyll fluorescence at 77 K among vascular plants of diverse origins. Planta 170:489–504.

Bradley BA, DS Wilcove and M Oppenheimer. 2010. Climate change increases risk of plant invasion in the Eastern United States. Biol. Invasions 12:1855–1872.

Cariñanos P and M Casares-Porcel. 2011. Urban green zones and related pollen allergy: A review. Some guidelines for designing spaces with low allergy impact. Landsc. Urban Plan. 101:205–214.

Chen LS and L Cheng. 2009. Photosystem 2 is more tolerant to high temperature in apple (*Malus domestica* Borkh.) leaves than in fruit peel. Photosynthetica 47:112–120.

Chen S, X Xu, X Dai, C Yang and S Qiang. 2007. Identification of tenuazonic acid as a novel type of natural photosystem II inhibitor binding in QB-site of *Chlamydomonas reinhardtii*. Biochim. Biophys. Acta 1767:306–318.

Chen S, J Yang, M Zhang, RJ Strasser and S Qiang. 2016. Classification and characteristics of heat tolerance in *Ageratina adenophora* populations using fast chlorophyll a fluorescence rise O-J-I-P. Environ. Exp. Bot. 122:126–140.

Guisse B, A Srivastava and RJ Strasser. 1995. The polyphasic rise of the chlorophyll a fluorescence (O-K-J-I-P) in heat stressed leaves. Arch. Sci. 48:147–160.

Haque MS, KH Kjaer, E Rosenqvist, DK Sharma and CO Ottosen. 2014. Heat stress and recovery of photosystem II efficiency in wheat (*Triticum aestivum* L.) cultivars acclimated to different growth temperatures. Environ. Exp. Bot. 99:1–8.

Havaux M, H Greppin and RJ Strasser. 1991. Functioning of photosystems I and II in pea leaves exposed to heat stress in the presence or absence of light. Planta 186:88–98.

Haldimann P and RJ Strasser. 1999. Effects of anaerobiosis as probed by the polyphasic chlorophyll a fluorescence rise kinetic in pea (*Pisum sativum* L.). Photosynth. Res. 62:67–83.

Ibaraki Y and J Murakami. 2007. Distribution of chlorophyll fluorescence parameter Fv/Fm within individual plants under various stress conditions. pp. 255–260. In: XXVII International Horticultural Congress-IHC2006: International Symposium on Advances in Environmental Control, Automation 761. Seoul, Korea.

Intergovernmental Panel on Climate Change (IPCC). 2007. Climate Change 2007: Mitigation of Climate Change, Contribution of Working Group III Contribution to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press. Cambridge, New York, USA.

Johnson GN, AJ Young, JD Scholes and P Horton. 1993. The dissipation of excess excitation energy in British plant species. Plant Cell Environ. 16:673–679.

Kautsky H and A Hirsch. 1931. Neue versuche zur kohlensäure-assimilation. Naturwissenschaften 19:964.

Kriedemann PE, RD Graham and JT Wiskich. 1985. Photosynthetic dysfunction and in vivo changes in chlorophyll a fluorescence from manganese-deficient wheat leaves. Aust. J. Agric. Res. 36:157–169.

Lazár D and P Ilík. 1997. High-temperature induced chlorophyll fluorescence changes in barley leaves: comparison of the critical temperatures determined from fluorescence induction and from fluorescence temperature curve. Plant Sci. 124:159–164.

Lu C and J Zhang. 1999. Effects of water stress on photosystem II photochemistry and its thermostability in wheat plants. J. Exp. Bot. 50:1199–1206.

Lu CM and JH Zhang. 2000. Heat-induced multiple effects on PS II in wheat plants. J. Plant Physiol. 156:259–265.

Mathur S, A Jajoo, P Mehta and S Bharti. 2011. Analysis of elevated temperature-induced inhibition of photosystem II using chlorophyll a fluorescence induction kinetics in wheat leaves (*Triticum aestivum*). Plant Biol. 13:1–6.

Oh S and SC Koh. 2013. Chlorophyll a fluorescence response to mercury stress in the freshwater microalga *Chlorella vulgaris*. J. Environ. Sci. Int. 22:705–715.

Oh S, KH Moon, IC Son, EY Song, YE Moon and SC Koh. 2014. Growth, photosynthesis and chlorophyll fluorescence of Chinese cabbage in response to high temperature. Korean J. Hortic. Sci. 32:318–329.

Qaderi MM and DM Reid. 2009. Crop responses to elevated carbon dioxide and temperature (chp1). In: Climate Change and Crops, Environmental Science and Engineering (Singh SN, ed.). Springer-Verlag Berlin Heidelberg.

Rhee KH, EP Morris, J Barber and W Kühlbrandt. 1998. Three-dimensional structure of the plant photosystem II reaction centre at 8 Å resolution. Nature 396:283–286.

Rodríguez VM, P Soengas, V Alonso-Villaverde, T Sotelo, ME Cartea and P Velasco. 2015. Effect of temperature stress on the early vegetative development of *Brassica oleracea* L. BMC Plant Biol. 15:145.

Salvucci ME and SJ Crafts-Brandner. 2004. Mechanism for deactivation of Rubisco under moderate heat stress. Physiol. Plant. 122:513–519.

Sironval C, RJ Strasser and M Brouers. 1981. Equivalence entre la theorie des flux et la theorie des relations entre proportions de pigments pour la description de la repartition de l'energie lumineuse absorbée par les membranes photoactives. Bulletin de l'Académia royale de Belgique 67:248–259.

Srivastava A, B Guissé, H Greppin and RJ Strasser. 1997. Regulation of antenna structure and transport in photosystem II of *Pisum sativum* under elevated temperature probed by fast polyphasic chlorophyll a fluorescence transient: OKJIP. Biochim. Biophys. Acta-Bioenerg. 1320:95–106.

Strasser BJ. 1995. Measuring fast fluorescence transients to address environmental questions: The JIP test. pp. 977–980. In: Photosynthesis: From Light to Biosphere (Mathis P, ed.). Kluwer Academic. Dordrecht, Netherlands.

Strasser BJ. 1997. Donor side capacity of photosystem II probed by chlorophyll a fluorescence transients. Photosynth. Res. 52:147–155.

Strasser R and Govindjee. 1992. On the O-J-I-P fluorescence transients in leaves and D1 mutants of *Chlamydomonas reinhardtii*. pp. 29–32. In: Research in Photosynthesis (Murata N, ed.). Kluwer Academic Publishers. Dordrecht, Netherlands.

Strasser RJ, A Srivastava and Govindjee. 1995. Polyphasic chlorophyll a fluorescence transient in plants and cyanobacteria. Photochem. Photobiol. 61:32–42.

Strasser RJ, A Srivastava and M Tsimilli-Michael. 2000. The fluorescence transient as a tool to characterize and screen photosynthetic samples. pp. 443–480. In: Probing Photosynthesis: Mechanisms, Regulation and Adaptation (Yunus M, U Pathre and P Mohanty, eds.). Taylor & Francis. London, UK.

Strasser RJ and M Tsimilli-Michael. 2001. Structure function relationship in the photosynthetic apparatus: a biophysical approach (chapter 16). pp. 271–303. In: Biophysical Processes in Living Systems (Saradhi PP, ed.). Science Publishers, Inc. Enfield (NH). New Hampshire, USA.

Strasser RJ, M Tsimilli-michael and A Srivastava. 2004. Analysis of the chlorophyll fluorescence transient. pp. 321–362. In: Chlorophyll a Fluorescence: a Signature of Photosynthesis. Advances in Photosynthesis and Respiration (Papageorgiou GC and Govindjee, eds.). Springer. Dordrecht, Netherlands.

Strauss AJ, GHJ Kruger, RJ Strasser and PDR van Heerden. 2006. Ranking of dark chilling tolerance in soybean genotypes probed by the chlorophyll a fluorescence transient O-J-I-P. Environ. Exp. Bot. 56:147–157.

Takahashi S and N Murata. 2008. How do environmental stresses accelerate photoinhibition? Trends Plant Sci. 13:178–182.

Tkemaladze GS and KA Makhhashvili. 2016. Climate changes and photosynthesis. Ann. Agrar. Sci. 14:119–126.

Tóth SZ, G Schansker, G Garab and RJ Strasser. 2007. Photosynthetic electron transport activity in heat-treated barley leaves: the role of internal alternative electron donors to photosystem II. Biochim. Biophys. Acta-Bioenerg. 1767:295–305.

Tóth SZ, G Schansker and RJ Strasser. 2005a. In intact leaves, the maximum fluorescence level (FM) is independent of the redox state of the plastoquinone pool: a DCMU-inhibition study. Biochim. Biophys. Acta-Bioenerg. 1708:275–282.

Tsimilli-Michael M, M Pêcheux and RJ Strasser. 1999. Light and heat stress adaptation of the symbionts of temperate and coral reef foraminifers probed in hospite by the chlorophyll a fluorescence kinetics. Zeitschrift für Naturforschung 54C:671–680.

Yan K, P Chen, H Shao, S Zhao, L Zhang, L Zhang, G Xu and J Sun. 2012. Responses of photosynthesis and photosystem II to higher temperature and salt stress in Sorghum. J. Agron. Crop Sci. 198:218–225.

Yoshioka M, S Uchida, H Mori, K Komayama, S Ohira, N Morita, T Nakanishi and Y Yamamoto. 2006. Quality control of photosystem II: Cleavage of reaction center D1 protein in spinach

thylakoids by FtsH protease under moderate heat stress. J. Biol. Chem. 281:21660–21669.

Zouni A, HT Witt, J Kern, P Fromme, N Krauss, W Saenger and P Orth. 2001. Crystal structure of photosystem II from *Synechococcus elongatus* at 3.8 Å resolution. Nature 409:739–743.

Zushi K, S Kajiwara and N Matsuzoe. 2012. Chlorophyll a fluorescence OJIP transient as a tool to characterize and evaluate response to heat and chilling stress in tomato leaf and fruit. Sci. Hortic. 148:39–46.

