Photosynthesis and activity of photosystem II in response to drought stress in Amur Grape (Vitis amurensis Rupr.)


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Abstract

The Amur Grape (Vitis amurensis Rupr.) cultivars ‘ShuangFeng’ and ‘ZuoShanyi’ were grown in shelter greenhouse under natural sunlight and subjected to drought. Sap flow rate, net photosynthetic rate (P\textsubscript{N}), and chlorophyll (Chl) fluorescence were measured on Amur Grape leaves subjected to different drought treatments. Significant decreases in P\textsubscript{N} were associated with increasing intercellular CO\textsubscript{2} concentration (C\textsubscript{i}), suggesting that the reduction in P\textsubscript{N} was caused by nonstomatal limitation. Analysis of OJIP transients according to the JIP-test protocol revealed that specific (per PSII reaction center) energy fluxes for light absorption, excitation energy trapping and electron transport have significantly changed. The appearance of a pronounced K-step and J-step in polyphasic rise of fluorescence transient suggested the oxygen-evolving complex and electron transport were inhibited. Drought stress has relatively little effect on the parameter maximal quantum yield of PSII photochemistry (F\textsubscript{v}/F\textsubscript{m}), but the performance index (PI\textsubscript{ABS}) is more sensitive in different drought treatment. There are cultivar differences in the response of PSII activity to drought, the photosynthetic apparatus of ‘ZuoShanyi’ cultivar is more resistant to drought than that of ‘ShuangFeng’, and JIP-test could be a useful indicator for evaluation and selection to drought tolerance.

Additional key words: Amur Grape; drought stress; JIP-test; net photosynthetic rate; photosystem II.

Introduction

Water is the most limiting resource in the arid and semiarid area, drought is one of the most limiting environmental factors for crop growth, so drought stress severely affects crop growth and reduces average yields (IPCC 2001, Chaves et al. 2003). With global warming, arid and semiarid area will be further expanded. The Amur Grape (Vitis amurensis Rupr.) as an economic crop is native to the Far East (China, Russia, Japan, Korea, and Siberia). It is very resistant to cold, but not tolerant to drought and sometimes need abundant watering.

Drought stress often results in stomatal closure, which is the initial cause for decrease of P\textsubscript{N} (Comin and Briantais 1991, Chaves 1991, Yordanov et al. 2000), but nonstomatal limitation may play a dominant role in decreased P\textsubscript{N} as stress develops. Nonstomatal limitation has been attributed to the reduced carboxylation efficiency or to the inhibited functional activity of PSI. Yu et al. (2009) thought that the decrease in P\textsubscript{N} might be associated with the increase in leaf to air vapour pressure deficit and the depression of effective quantum yield of PSII photochemistry (\Phi\textsubscript{PSII}) on grapevine. However, Vos and Oyarzun (1987) showed that the decrease in P\textsubscript{N} is related to stomatal limitation on potato.

Photosynthetic capacity and quantum yield show little or no change in low drought stress (Yordanov et al. 2003), but moderate and severe drought stress often inhibits activity of enzymes related to Calvin cycle and...
photochemistry (Maroco et al. 2002, Lawlor and Cornic 2002, Dias and Brüggemann 2007). Although plant have efficient mechanisms to protect photosynthetic apparatus from injury, for example, the over-excited energy can be consumed by Mehler reaction and photorespiration (Thomas and Stoddart 1980, Demmig-Adams and Adams 1992, Foyer and Noctor 2000, Foyer and Noctor 2003), severe drought stress still will result in damage of photosynthetic apparatus (Valladares and Pearcy 2002). Flexas et al. (2000) studied the change of F_m in drought stress on grape. Christen et al. (2007) thought that the JIP-test analysis may offer more information about photosynthetic processes and supplied a sensitive means to detect drought stress on mature grapevines.

Chl a fluorescence measurements represent a sensitive and reliable method to monitor and quantify the changes induced in the photosynthetic apparatus by drought stress (Van Heerden et al. 2007, Naumann et al. 2007, Oukarroum et al. 2007, Oukarroum et al. 2009). Although effects of drought stress on other plants have been extensively studied, growing ability of Amur grape on drought-prone areas has barely been studied. For this reason, two Amur Grape cultivars were compared: ‘ZuoShanyi’ and ‘ShuangFeng’ are main Amur grape cultivars in northeast area of China, they have shown a high productivity. Many farmers in the area use the two cultivars. In recent years, Amur grape have been planted gradually in arid regions, for example Mongolia, which decrease the probability for downy mildew infection. But in these regions Amur grape is often subjected to severe drought stress as well as other environmental constraints, such as high temperature and excessive light, which cause severe decrease of yield and fruit quality. Nondestructive methods to evaluate drought of Amur grape are still missing. Therefore, the purpose of this study was to analyze photosynthetic efficiency and Chl a fluorescence transients of Amur grape affected by drought stress, and to assess fluorescence monitoring as a possible tool to evaluate drought tolerance of Amur grape. In this frame, it can be used to adapt agricultural and forestry practices to cope with fluctuations of local environmental conditions as a result of predicted global climate changes.

Materials and methods

Plants: A rain shelter greenhouse experiment was conducted from May 2010 till August 2010, annual cutting seedlings of Amur Grape (V. amurensis Rupr.) cultivars ‘ShuangFeng’ and ‘ZuoShanyi’ were grown in plastic pots (290 mm in diameter and 180 mm high) under natural daylight conditions at the Institute of Special Wild Economic Animal and Plant Science. Jilin city, Jilin province was selected as experimental site. It is situated between 44°04′N and 126°05′E, annual precipitation is 679 mm, annual highest and lowest temperatures are 35°C and –30°C, respectively. When the plants were in direct sunlight they received a PPFD of 1,400–1,600 µmol m–2 s–1. Average night- and day-time temperatures in shelter greenhouse were 16°C and 30°C, and relative air humidity was 85% and 60%, respectively. Plant materials of two varieties were uniform and of the same size about 200 mm high. Prior to the start of the experiment, the cutting seedlings were taken out of sandy bed, the roots washed and individually transplanted in pots filled with matrix which was composed of deer manure, vermiculite, sand, and garden soil (1:1:1:4, v/v/v). All seedlings were irrigated daily. After 7 weeks of growth, the drought treatments were initiated. The drought stress treatments were gradually imposed. Three drought-stress levels were considered and compared to a control (CK), in which the soil water potential was kept constant because seedlings were well watered daily. Drought-stressed plants showed low water deficit after 6 d without watering (low stress, LD), moderate stress after 12 d without watering (moderate stress, MD) and severe stress after 24 d without watering (severe stress, SD). Control and drought-stressed seedlings were arranged in a completely randomized design with five replications. Drought stress (four levels, including the control treatment) and varieties (two) were considered as treatments. In total, 40 seedlings were used. The measured plant had 8 leaves.

Absolute water content in soil: For the pots under different drought level, SWC was calculated gravimetricaly. Immediately after measurement of plant sap flow rate, CO₂ assimilation and Chl a fluorescence transient terminated, soil wet mass (SWM) was determined. Then all the pot soil was oven-dried to constant mass at 105 ± 2°C for over 24 h for dry mass determinations of pot soil (SDM). Soil water content (SWC) was calculated from the equation: SWC [%] = (SWM – SDM)/SDM × 100.

Plant sap flow rate: The sap flow rate of Amur Grape has been observed by advanced Photosynthesis Monitor (PTM-48M, Daletown Company Ltd., Nicosia, Cyprus), sap flow collectors were SF-3P Sensor (suitable for 4- to 10-mm stem diameter). The sensors were installed about 0.5–1.0 m above ground, depending on stem diameter, as recommended by the manufacturer. The sensors were covered carefully with two or three layers of aluminium foil in order to protect the sensor from external heat effects.

CO₂ assimilation: Net photosynthetic rate ($P_n$) was measured with a portable photosynthesis system (CIRAS-2; PP-Systems, Hitchin, UK). At the same time, intercellular CO₂ concentration ($C_i$), transpiration rate ($E$), and stomatal conductance ($g_s$) were automatically recorded. Leaves were illuminated by the CIRAS-2 light source with a PPFD of 1,200 µmol m⁻² s⁻¹, at 360 µmol mol⁻¹(CO₂)
concentration, a leaf temperature of 30°C, 70% relative humidity, and flow rate of 196 ml min⁻¹.

**Chl a fluorescence transient measurement and JIP-test:** Chl a fluorescence transient (OJIP) was measured with a Plant Efficiency Analyzer (Pocket-PEA, Hansatech Instruments, King’s Lynn, Norfolk, UK). Fluorescence transients were recorded during a 1-s pulse of red radiation (3,500 µmol m⁻² s⁻¹). The fluorescence signals were recorded within a time scan from 10 µs to 1 s with a data acquisition rate of 100 readings ms⁻¹ for the first 2 ms, and 1 reading ms⁻¹ after 2 ms. All the measurements were done on full dark-adapted (20 min) leaves.

Fluorescence transients were analyzed according to the JIP-test, which is a Chl a fluorescence analysis program (BIOLYZER HP3, Bioenergetics Laboratory, University of Geneva, Switzerland) (Strasser and Strasser 1995, Strasser 1997, Strasser et al. 2004). The following data from the original fluorescence measurements were used: the minimal fluorescence of dark-adapted state or the fluorescence intensity at 50 µs (considered as F₃); the maximal fluorescence of dark-adapted state (F₇₄₃); the fluorescence intensities at 300 µs (K-step). The JIP-test represents translation of the original recorded data to biophysical parameters that quantify the stepwise energy flow through PSII (Van Heerden et al. 2007). The following parameters refer to time zero (onset of fluorescence induction): (1) The specific energy fluxes (per RC) for absorption (ABS/RC), trapping (TRₒ/RC), electron transport (ETₒ/RC) and dissipation (DIₒ/RC); (2) the maximal quantum yield of PSII photochemistry (Fᵥ/F₇₄₃). The formulae used to calculate these biophysical parameters are detailed in Appendix.

The performance index (PIₐₐ₅ₐ), is also calculated (Strasser et al. 2000). The PIₐₐ₅ₐ considers the three main steps that regulate photosynthetic activity by a PSII reaction centre (RC) complex, namely absorption of light energy (ABS), trapping of excitation energy (TR) and conversion of excitation energy to electron transport (ET) (Van Heerden et al. 2007).

**Statistical analysis** was conducted with the software of SAS (JMP 6.0, SAS Institute, Cary, NC, USA). In data sets with parametric distribution, significant differences between means of the treatments means were determined using Student’s t-test at a significant level set to 0.05. Sap flow rates of stem are the averages of three replicates from 3 plants per experiment (drought stress and control); gas-exchange data are the average of individual measurements of 3 plants per experiment; Chl a fluorescence parameters are the average of individual measurements of 6 plants per experiment.

**Results**

**Changes of soil water content in different treatments:** SWC of different treatment decreased significantly from 25.71% in well watered treatment to 17.44%, 12.18%, and 9.44%, in LD, MD, and SD stress treatment, respectively (Table 1).

Table 1. Changes of soil relative water content (RWC) in different drought treatments of Amur Grape. Each value is the mean ± SD (n = 5). Different letters indicate significant difference (P<0.05). CK – control, LD – low drought, MD – moderate drought, SD – severe drought.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>RWC [%]</th>
</tr>
</thead>
<tbody>
<tr>
<td>CK</td>
<td>25.71 ± 0.68a</td>
</tr>
<tr>
<td>LD</td>
<td>17.44 ± 0.95b</td>
</tr>
<tr>
<td>MD</td>
<td>12.18 ± 1.20c</td>
</tr>
<tr>
<td>SD</td>
<td>9.44 ± 1.29d</td>
</tr>
</tbody>
</table>

**Effects of drought stress on sap flow rate:** Drought stress decreased sap flow rate of Amur Grape (Fig. 1A). Sap flow rate decreased from 1.69 ml h⁻¹ in well watered Amur Grape to 1.49 ml h⁻¹ and 0.93 ml h⁻¹ in MD and SD stress in ‘ZuoShanyi’ seedlings respectively, but had a relative increase in LD stress. For ‘ShuangFeng’ seedlings, change of sap flow rate is similar to ‘ZuoShanyi’, but sap flow rate of ‘ShuangFeng’ is lower than ‘ZuoShanyi’ in different treatment.

Fig. 1. Effects of drought stress on sap flow rate in the Amur grape stem (A) and transpiration rate (B) in ‘ZuoShanyi’ and ‘ShuangFeng’, at the end of low, moderate and severe drought stress, values are means ± SD (n = 3). Different letters indicate significant difference (P<0.05). E – transpiration rate, CK – control, LD – low drought, MD – moderate drought, SD – severe drought, S – ShuangFeng, Z – ZuoShanyi.
Fig. 2. Effects of drought stress on intercellular CO₂ concentration ($C_i$) (A), stomatal conductance ($g_s$) (B), net photosynthetic rate ($P_N$) (C), and maximal quantum yield of PSII photochemistry ($F_v/F_m$) (D) of the Amur Grape, values are means ± SD ($n = 3$). Different letters indicate significant difference ($P<0.05$). CK – control, LD – low drought, MD – moderate drought, SD – severe drought, S – ShuangFeng, Z – ZuoShanyi.

**Effects of drought stress on $P_N$, $g_s$, $E$, and $C_i$:** $C_i$ increased by 23.78% and 40.78% in MD- and SD-stressed ‘ZuoShanyi’ seedlings respectively, but decreased 15.7% in LD, compared to the ‘ZuoShanyi’. For ‘ShuangFeng’ seedlings $C_i$ increased by 25.86% and 57.78%, respectively, in MD and SD stress treatment (Fig. 2A) compared to well watered plants, but decreased 17.94% in LD.

$g_s$ and $E$ also decreased significantly as stress developed (Figs. 1B, 2B), but $E$ of ‘ShuangFeng’ is higher than ‘ZuoShanyi’ in different treatment.

$P_N$ decreased from 10.97 µmol m⁻² s⁻¹ in well watered ‘ZuoShanyi’ to 7.97 µmol m⁻² s⁻¹, 0.37 µmol m⁻² s⁻¹ and 0.17 µmol m⁻² s⁻¹ in LD, MD, and SD stress treatments, but $P_N$ of ‘ShuangFeng’ cultivar decreased from 11.23 µmol m⁻² s⁻¹ in well watered seedlings to 6.27 µmol m⁻² s⁻¹, –0.07 µmol m⁻² s⁻¹ and –0.33 µmol m⁻² s⁻¹, in LD, MD, and SD stress treatment respectively (Fig. 2C).

**Changes of Chl fluorescence transients:** The polyphasic rise of fluorescence transients (OJIP) was measured with a plant efficiency analyzer. Drought tolerance of two Amur Grapes in response to days of stress was evaluated through analyzing several leaf fluorescence parameters.
PHOTOSYNTHESIS AND ACTIVITY OF PHOTOSYSTEM II IN RESPONSE TO DROUGHT STRESS

In order to compare changes in fluorescence rise kinetic more clearly, original fluorescence curves were normalized between $F_0$ and $F_m$ (Fig. 3A-C). Variable fluorescence ($V_f$) curves of two Amur Grape cultivars are different, the curve of ‘ShuangFeng’ showed a significant difference between control and treatment, however, the curve of ‘ZuoShanyi’ has no significant difference compared to ‘ShuangFeng’. For SD stress treatments, a very clear K-step appeared in ‘ShuangFeng’. In order to compare the changes in the amplitude of the K-step (at about 300 µs) during drought stress, the fluorescence curves were normalized between $F_0$ and $F_j$ (at 2 ms), i.e. $W = (F_t - F_0)/(F_j - F_0)$. The amplitude of the K-step (W$_k$) was expressed as the ratio W (treatment)/W (control) = ΔW. A higher W$_k$ was observed in SD stress than in LD and MD stress, otherwise, a ΔW$_k$ of ‘ShuangFeng’ is higher than that of ‘ZuoShanyi’ in SD stress (Figs. 3B,D).

In order to determine the sensitivity and dynamic ranges of the fluorescence parameters in different drought stresses and different cultivars, we compared the different fluorescence parameters in two cultivars under different drought-stress conditions.

The OJIP fluorescence transients were analyzed according to the JIP-test protocol. Fig. 2D shows the changes of maximal quantum yield of PSII photo-chemistry ($F_v/F_m$). Results demonstrate that $F_v/F_m$ showed a significant difference only under moderate and severe drought condition in ‘ShuangFeng’ but a slight change in ‘ZuoShanyi’.

The effects of drought stress on specific (per excited cross-section of leaf) energy fluxes for light absorption, excitation energy trapping and electron transport are presented in the form of column diagram (Fig. 4A,B). ABS/RC, TR$_{0}$/RC, ET$_{0}$/RC and DI$_{0}$/RC had increased significantly, for acclimation responses to drought stress in ‘ShuangFeng’, but increased significantly only under MD and SD condition in ‘ZuoShanyi’.

Table 2 shows the changes in $F_0$, $F_m$, $F_j$, dv/dt and PI$_{ABS}$ in ‘ShuangFeng’ and ‘ZuoShanyi’ in different treatments. $F_0$ and dv/dt increased significantly comparable to CK in ‘ShuangFeng’, but increased significantly only in MD and SD stress in ‘ZuoShanyi’. $F_m$, $F_j$ and PI$_{ABS}$ had respectively decreased in both varieties. But $F_j$ decreased significantly only in MD and SD stress, PI$_{ABS}$ decreased significantly in LD stress in ‘ShuangFeng’, increased significantly in LD stress in ‘ZuoShanyi’.

Table 2. Changes of $F_0$, $F_m$, $F_j$, dv/dt, and PI$_{ABS}$ in different drought treatments of Amur Grape. Each value is the mean ± SD (n = 6). Different letters indicate significant difference (P<0.05). CK – control, LD – low drought, MD – moderate drought, SD – severe drought, F0 – minimal fluorescence of dark-adapted state; $F_m$ – maximal fluorescence of dark-adapted state, $F_j$ – fluorescence intensity at the J-step (at 2 ms), dv/dt – the initial slope of the relative variable fluorescence, PI$_{ABS}$ – performance index.

<table>
<thead>
<tr>
<th>Variety</th>
<th>Treatment</th>
<th>$F_0$</th>
<th>$F_m$</th>
<th>$F_j$</th>
<th>dv/dt</th>
<th>PI$_{ABS}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>ShuangFeng</td>
<td>CK</td>
<td>369 ± 6$^a$</td>
<td>1,668 ± 11$^a$</td>
<td>1,122 ± 21$^a$</td>
<td>0.774 ± 0.078$^d$</td>
<td>13.3 ± 3.06$^b$</td>
</tr>
<tr>
<td></td>
<td>LD</td>
<td>402 ± 20$^b$</td>
<td>1,656 ± 100$^a$</td>
<td>1,134 ± 19$^a$</td>
<td>0.992 ± 0.091$^c$</td>
<td>7.57 ± 1.05$^{bc}$</td>
</tr>
<tr>
<td></td>
<td>MD</td>
<td>416 ± 32$^b$</td>
<td>1,306 ± 67$^a$</td>
<td>1,150 ± 37$^{abc}$</td>
<td>1.308 ± 0.19$^b$</td>
<td>2.16 ± 1.07$^a$</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>475 ± 33$^a$</td>
<td>1,221 ± 68$^a$</td>
<td>1,177 ± 32$^a$</td>
<td>1.688 ± 0.32$^a$</td>
<td>0.92 ± 0.76$^d$</td>
</tr>
<tr>
<td>ZuoShanyi</td>
<td>CK</td>
<td>334 ± 7$^a$</td>
<td>1,729 ± 7$^a$</td>
<td>1,154 ± 37$^{abc}$</td>
<td>0.761 ± 0.11$^d$</td>
<td>15.8 ± 2.9$^b$</td>
</tr>
<tr>
<td></td>
<td>LD</td>
<td>323 ± 19$^f$</td>
<td>1,746 ± 13$^a$</td>
<td>1,164 ± 69$^{ab}$</td>
<td>0.808 ± 0.11$^d$</td>
<td>21.7 ± 5.1$^a$</td>
</tr>
<tr>
<td></td>
<td>MD</td>
<td>341 ± 8$^{de}$</td>
<td>1,520 ± 80$^b$</td>
<td>1,169 ± 23$^{ab}$</td>
<td>0.941 ± 0.09$^{de}$</td>
<td>8.3 ± 1.3$^c$</td>
</tr>
<tr>
<td></td>
<td>SD</td>
<td>361 ± 11$^{cd}$</td>
<td>1,535 ± 33$^b$</td>
<td>1,183 ± 25$^a$</td>
<td>1.293 ± 0.09$^{b}$</td>
<td>5.2 ± 2.2$^d$</td>
</tr>
</tbody>
</table>

Table 3. Relationships of the relative activities in absorption flux per reaction center (ABS/RC), trapped energy flux per RC (TR$_{0}$/RC), electron transport flux per RC (ET$_{0}$/RC) and dissipated energy flux per RC (DI$_{0}$/RC) of Amur grape leaf in different drought treatments. Each value is the mean ± SD (n = 6). Different letters indicate significant difference (P<0.05). CK – control, LD – low drought, MD – moderate drought, SD – severe drought.

Fig. 4. Relationships of the relative activities in absorption flux per reaction center (ABS/RC), trapped energy flux per RC (TR$_{0}$/RC), electron transport flux per RC (ET$_{0}$/RC) and dissipated energy flux per RC (DI$_{0}$/RC) of Amur grape leaf in different drought treatments. Each value is the mean ± SD (n = 6). 'ZuoShanyi' (A), 'ShuangFeng' (B). Different letters indicate significant difference (P<0.05). CK – control, LD – low drought, MD – moderate drought, SD – severe drought.
Discussion

Drought affects many physiological and biochemical processes within plants. There are many studies to develop some new screening methods in order to evaluate severity of drought (Boussadia et al. 2008, Van Heerden et al. 2007). In the present study, our results show that the photosynthetic apparatus of ‘ZuoShanyi’ becomes more resistant to drought stress than ‘ShuangFeng’ when grown in arid and semiarid areas. This can be seen by the significantly enhanced resistance of PSIi, as reflected in the changes of the various fluorescence parameters related to PSIi function.

Drought stress effects on sap flow: Sap flow rate of ‘ShuangFeng’ is lower than ‘ZuoShanyi’ in different treatments, but E of ‘ShuangFeng’ is higher than ‘ZuoShanyi’ in different treatments. In addition, air vapour pressure deficit of ‘ShuangFeng’ is higher than ‘ZuoShanyi’ in different treatment (data not shown). These results indicated ‘ShuangFeng’ lost more water than ‘ZuoShanyi’, the more water inside ‘ZuoShanyi’ may keep leaf and whole plant survive. Further, ‘ZuoShanyi’ may be more drought-tolerant.

Drought stress effects on leaf photosynthesis: A significant relationship was determined between NE, Pn, and Ci. Pn and g were decreased significantly as drought stress developed, but Ci shows a decrease in LD stress and increase in MD and SD stresses, suggesting that Ci might not be the limiting factor for photosynthesis in the stressed plant (Dias and Brüggemann 2010). Many studies confirmed that Ci remained constant when Pn decreased apparently (Correia et al. 1990, Sinha et al. 1997). The decrease in Pn might be due to photodamage, while photodamage is often associated with environmental stress, such as drought or temperature (Ohashi et al. 2006, Gamon and Peary 1990). Pn in the leaves is depressed due to stomatal limitation in LD stress, but in MD and SD stresses nonstomatal limitation will be a main factor which depressed the Pn (Galmés et al. 2007), although it has been reported by Flexas et al. (2000) that decreases in Pn were mainly due to stomatal closure and not to decreased photochemical efficiency of PSI in grapevines. The nonstomatal limitation factor may be inhibition in activity of various enzymes of the Calvin cycle and PSI photochemistry (Chaves et al. 2003, Maroco et al. 2002).

The importance of fluorescence parameters for water-stress assessment: In order to gain more insight into the enhanced resistance of PSIi to drought stress, the structure and function of PSIi were evaluated according to the JIP test by measuring the fast Chl fluorescence transients, which can be used as a tool to detect and estimate the PSIi behavior. The parameters of phenomenological energy fluxes can be visualized by means of an energy pipeline model of the photosynthetic apparatus in the form of the leaf model (Strasser 1987, Strasser et al. 1996, Krüger et al. 1997). However, more importantly, we observed that the decrease in the number of active RCs was much greater in severe drought stress than that in control. The well watered plants have no inactivation of the RC.

Many studies demonstrated that drought stress resulted in injuries to oxygen-evolving complex (Guissé et al. 1995). Oukarroum et al. (2007) considered that changes of the L and K-bands of the fluorescence transients OJIP could predict the vitality of leaves and the tolerance of the varieties to drought stress. The effect of the different treatments on ‘ZuoShanyi’ and ‘ShuangFeng’ was evident by comparing the shape of the corresponding fluorescence transient curves. A similarity between the two Amur Grape cultivars was the appearance of a clearly defined K-peak at around 300 µs (ΔW-peak), which suggests inhibition or damage of the oxygen-evolving complex (OEC) (Stasser 1997, Wen et al. 2005). Previous studies have found that there is appearance of a K-peak when leaves are exposed to elevated temperature (Jiang et al. 2006, Li et al. 2009). Appearance of K-step under drought stress may be due to lower stomatal conductance and transpiration rate, which increase leaf temperature (Van Heerden et al. 2007). One difference, however ΔW value of ‘ShuangFeng’ is higher than ‘ZuoShanyi’, suggesting oxygen-evolving complex of ‘ZuoShanyi’ have greater resistance than ‘ShuangFeng’. Thereby, the appearance of K-step can be used as a convenient stress indicator and specifically attributed to a damage on the oxygen-evolving complex on a practical basis (Strasser 1997, Wen et al. 2005).

As noted before, drought stress has relatively slight effect on the parameter F/Fm, which decreases only under SD stress condition (Lu and Zhang 1999, Zlatev and Yordanov 2004, Oukarroum et al. 2007). Many studies confirmed parameters derived from the JIP-test to be very sensitive to indicate the physiological status of the plant under drought stress (Wen et al. 2005). PIABS may be a good parameter for monitoring and evaluating drought stress (Van Heerden et al. 2007). Our results showed that PIABS had a highly significant change comparable to CK in ‘ShuangFeng’, but decreased significantly only in MD and SD stress in ‘ZuoShanyi’. Similar changes were observed in barley, where PIABS of Ait Baha had an increase in drought stress, suggesting the cultivar is more tolerant to drought stress (Oukarroum et al. 2009). Our results further indicated that the photosynthetic apparatus of ‘ZuoShanyi’ becomes more resistant to drought stress than ‘ShuangFeng’.

In addition, ABS/RC, TR/RC, ET/RC, DL/RC, F, Fm, and dv/dt started to change significantly only in MD or SD stress in both cultivars. It must be stressed that TR/RC expresses the maximal rate of the closure of RCs.
ABS/RC = $M_0/(1/V_J)(1/\phi_{po})$

TR$_\psi$/RC = $M_0/(1/V_J)$

ET$_\psi$/RC = $M_0/(1/V_J) \psi_{ps}$

DI$_\psi$/RC = (ABS/RC) - (TR$_\psi$/RC)

The initial slope of the relative variable fluorescence (dv/dt)$_0$ can be expressed as the initial increment per ms using the time interval of 250 μs where: (dv/dt)$_0 \approx 4(F_{300\mu s} - F_0)/(F_m - F_0)$.

In the JIP-test the Equation $[4(F_{300\mu s} - F_0)/(F_m - F_0)]$ is represented by the parameter $M_0$, and is defined as the net rate of PSII closure.

Performance indexes

$PI_{ABS} = (RC/ABS)[\phi_{ps}/(1 - \phi_{ps})][\psi_{ps}/(1 - \psi_{ps})]$


Appendix

Summary of JIP-test formulae using data extracted from the OJIP transients (Van Heerden et al. 2007):

Specific fluxes or specific activities

ABS/RC = $M_0/(1/V_J)(1/\phi_{po})$

TR$_\psi$/RC = $M_0/(1/V_J)$

ET$_\psi$/RC = $M_0/(1/V_J) \psi_{ps}$

DI$_\psi$/RC = (ABS/RC) - (TR$_\psi$/RC)


Strasser, B.J., Strasser, R.J.: Measuring fast fluorescence transients to address environmental questions: the JIP test. – Z.X. WANG et al.