



## ASSAYING THE PHOTOSYNTHETIC PERFORMANCE OF SALT-AFFECTED SOYBEAN USING CHLOROPHYLL-A FLUORESCENCE TRANSIENTS

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**ABSTRACT:** A pot-cultured trial was designed to evaluate the influences of salinity on soybean [*Glycine max* (L.) Merrill] chlorophyll-a fluorescence as a rapid indicator of photophosphorylation reactions. A factorial experiment with four replicates was planned based on a completely randomized design with five saline degrees (control, 50, 100, 150, and 200 mM NaCl) and five soybean cultivars, Williams and Clark as the wild-type cultivars, M-4 as the mutant cultivar for Williams in addition to M-7 and M-9 as the mutant cultivars for Clark. All measurements were done at two vegetative and reproductive phases under controlled greenhouse circumstance. With increasing salinity levels, chlorophyll index decreased significantly at both phenological phases. Approximately, all the fluorescence parameters responded adversely to saline enhancement at either phase. Entirely, the mutants performed better than their wild-types exposed to saline condition and M-9 exhibited the best performance among them. There was a highly significant relationship between chlorophyll index and total vitality index per either absorption or cross section expressing the key role of chlorophyll content in photosystem functions of soybean under salinity stress.

**Keywords:** Fluorescence, Chlorophyll, Salinity, Stress, Soybean

## INTRODUCTION

Saline water as a worldwide problem has restricted agricultural activities in recent decades. Approximately, it is inevitable to cultivate crops applying either saline irrigation or in saline soils. Therefore, several cultivars have already been bred or derived from different crops in order to tolerate saline conditions. However, there are a few instantaneous or at least rapid procedures to screen cultivars exposed to saline stress. One of the most immediately efficient field methods for evaluating various plant stresses is estimating chlorophyll-a fluorescence. Among diverse chlorophyll-a fluorescence parameters, vitality index (PI) is the most susceptible parameter toward stresses while the ordinary employed  $\Phi_{p_0}$  is not sufficiently responsive to trace chlorophyll-a fluorescence variations [23].

Vitality indices, as new more-sensitive, important stress indicators, are applied to approximate the vitality of photosynthetic apparatus. They are based on three to four components consisting of reaction center densities, maximum quantum yield ( $\Phi_{p_0}$ ), electron transport efficiency from  $Q_A$  to  $Q_B$  ( $\Psi_{E_0}$ ), and electron transport efficiency from  $Q_B$  to the end electron acceptors of PSI ( $\delta_{R_0}$ ). While a stress factor influenced one of them, vitality indices demonstrate the stressful effects visibly [1, 3, and 23]. Similarly, redox potentials associate with oxido-reduction reactions of photochemical components through photosynthesis process, which introduce the functions in chemistry language [20].

Concerning numerous soybean mutants been bred presently and the strategic role of soybean as an oilseed, current evaluation was carried out to contrast chlorophyll-a fluorescence transients of two traditional soybean wild-type cultivars and some of their derived mutant cultivars under salt-stressed conditions.

## MATERIALS AND METHODS

### Experimental condition

The investigation was conducted as a four-replicated factorial pot trial based on a completely randomized design in a climate-controlled greenhouse (30:25 °C diurnal temperature; 40:50 % diurnal RH; 1500  $\mu\text{mol m}^{-2} \text{s}^{-1}$  midday light, 14:10 h diurnal photoperiod) at the College of Agriculture and Natural Resources (University of Tehran, Karaj, Iran) during summer and autumn, 2011. Tungsten (100 W  $\text{m}^{-2}$ ) and white light fluorescent (23 W  $\text{m}^{-2}$ ) lamps illuminated further lighting after removing other artificial light sources throughout night. Every replicate was consisted of three pots.

In the present study, five soybean cultivars comprising of Williams and Clark as the wild-type cultivars in addition to M-4 as the mutant cultivar for Williams, besides M-7 and M-9 as the mutant cultivars for Clark (Seed & Plant Improvement Institute, Karaj, Iran), were investigated. Salinity was treated by adding NaCl (Merck KGaA, Darmstadt, Germany) to tap water (table 1) at five saline degrees: control (tap water), 50, 100, 150, and 200 mM NaCl resembling electrical conductivities (EC): control (table 1), 5, 10, 15, and 20  $\text{dS m}^{-1}$ . Saline treatments began two weeks after planting and persisted until the termination of growth term. Saline degrees further than 50 mM were imposed gently by 50 mM steps every 3 days up to every terminal degree. The soil electric conductivity, evaluated after the trial, was 14, 47, 87, 132, and 181 mM for five saline treatments, in order. The soil and water details, used in this study, are shown in table 1.

Ten inoculated seeds were cultivated in every in 30 × 20 cm pot and plantlets were thinned to four uniform ones afterward. Likewise, 50 and 40 N and P  $\text{kg}^{-1}$ , respectively, were applied as urea (phase V2: growing of second node and/or second trifoliate leaf) and triple superphosphate (before cultivation).

To avoid any moisture deficiency, the pots were intermittently irrigated twice a week using saline and tap water up to field capacity (FC). Moreover, every four weeks, the pots were watered to drain and the drainwater EC of every treatment was measured to be confident about its validity.

**Table 1. Physico-chemical details of soil and water.**

| A. Soil physical details                              |                           |                     |   |                    |  |                  |  |                  |                 |                 |  |
|---|---------------------------|---------------------|---|--------------------|--|------------------|--|------------------|-----------------|-----------------|--|
| $D_b$ ( $\text{g cm}^{-3}$ )                          | Soil Water Content (%)    |                     |   | Soil Particles (%) |  |                  | Soil Texture   |                  |                 |                 |  |
|   | SP                        | FC                  | PWP   | Clay               | Silt   | Sand             |  |                  |                 |                 |  |
| 1.39  | 38                        | 22                  | 9   | 33                 | 39   | 28               | Clay Loam  |                  |                 |                 |  |
| B. Soil chemical details: ions                        |                           |                     |   |                    |  |                  |  |                  |                 |                 |  |
| pH  | EC ( $\text{dS m}^{-1}$ ) | SAR                 | Cations ( $\text{meq } 100 \text{ g}^{-1} \text{ Soil}$ ) |                    |  |                  | Anions ( $\text{meq } 100 \text{ g}^{-1} \text{ Soil}$ ) |                  |                 |                 |  |
|   |                           |                     | $\text{Na}^+$   | $\text{K}^+$       | $\text{Ca}^{2+}$   | $\text{Mg}^{2+}$ | $\text{Cl}^-$  | $\text{HCO}_3^-$ | $\text{CO}_3^-$ | $\text{SO}_4^-$ |  |
| 7.8   | 1.1                       | 0.42                | 1.1   | 4.9                | 6.6  | 7.1              | 2.4  | 4.2              | 3.9             | 7.1             |  |
| C. Soil chemical details: chemicals                   |                           |                     |   |                    |  |                  |  |                  |                 |                 |  |
| CEC ( $\text{meq } 100 \text{ g}^{-1} \text{ soil}$ ) | ESP (%)                   | $\text{CaCO}_3$ (%) | Organic parts (%)   |                    | Macronutrient Parts ( $\text{mg kg}^{-1} \text{ Soil}$ ) |                  |  |                  |                 |                 |  |
|   |                           |                     | OM  | OC                 | Total N  | Available P      | Available K  |                  |                 |                 |  |
| 19.7  | 5.58                      | 10.4                | 0.76  | 0.41               | 0.06   | 8.1              | 96   |                  |                 |                 |  |
| D. Water soluble chemical details                     |                           |                     |   |                    |  |                  |  |                  |                 |                 |  |
| pH  | EC ( $\text{dS m}^{-1}$ ) | SAR                 | Cations ( $\text{meq L}^{-1}$ )                           |                    |  |                  | Anions ( $\text{meq L}^{-1}$ )                           |                  |                 |                 |  |
|   |                           |                     | $\text{Na}^+$   | $\text{K}^+$       | $\text{Ca}^{2+}$   | $\text{Mg}^{2+}$ | $\text{Cl}^-$  | $\text{HCO}_3^-$ | $\text{CO}_3^-$ | $\text{SO}_4^-$ |  |
| 7.4   | 0.7                       | 0.16                | 0.3   | 2.9                | 3.6  | 3.1              | 3.9  | 3.5              | 3.1             | 2.8             |  |

\*  $D_b$ , soil bulk density; SP, saturation point; FC, field capacity; PWP, permanent wilting point; pH, power of  $\text{H}^+$ ; EC, electrical conductivity; SAR, sodium adsorption ratio; CEC, cation exchange capacity; ESP, exchangeable sodium percentage.

### Chlorophyll index measurement

The chlorophyll index (CI) was examined on upper (adaxial) central part -with the exception of the nervure- of the intermediate leaflet belonging to the youngest, intact, fully expanded leaf of three plants' main stem in every replicate at two vegetative (V5: growing of fifth node and/or fifth trifoliate leaf) and reproductive (R5: beginning of seed filling) phases of soybean phenology expressed by Iowa State University of Science and Technology [15].

A portable chlorophyllmeter (Chlorophyll Content Meter CL-01, Hansatech Instrument Ltd., King's Lynn, Norfolk, UK) was applied to detect relative chlorophyll content on the basis of dual absorbance wavelength; 620

nm (red band) which is highly absorbed by chlorophylls and a reference wavelength of 940 nm, from 10:00 to 14:00 h. The records were displayed as chlorophyll index [17].

### Chlorophyll fluorescence measurement

Polyphasic chlorophyll-a fluorescence kinetics (OKJIP) were recorded on the same leaflets of leaves, evaluated the chlorophyll index, with a portable fluorimeter (Pocket PEA, Hansatech Instrument Ltd., King's Lynn, Norfolk, UK) during 10:00 to 14:00 h and thereafter quantified using the JIP-test. All measurements done on fully dark-adapted leaves for 15 min using specific leaf clips. Chlorophyll fluorescence was induced by three-second pulses of red light (peak wavelength of 627 nm) illuminated from the LED filtered via an NIR filter at maximum saturation irradiance of  $3500 \mu\text{mol m}^{-2} \text{s}^{-1}$ . A 16-bit analogue/digital resolution signal with acquainting rate of every 10  $\mu\text{s}$  for the first 300  $\mu\text{s}$ , then every 100  $\mu\text{s}$  up to three ms, subsequently one-ms intervals up to 300 ms, and lastly every 10 ms toward the end, was used for chlorophyll-a fluorescence recording. The fluorescence signal at 50  $\mu\text{s}$  after the origin of illumination was considered as  $F_0$  [21 and 22].

For analyzing fast chlorophyll-a fluorescence transients, providing structural and functional information about photosystem II (PSII) behavior, the JIP-test [21 and 22] was used. The extracted dark-adapted data and equations utilized for calculating chlorophyll-a fluorescence characteristics were explained in table 2.

**Table 2. Extracted and calculated leaf chlorophyll-a parameters owing to fast fluorescence transient (OKJIP) graph [1, 3, 7, 9, 14, and 18 to 25]. With the exception of  $T_M$ , recorded in ms, all the chlorophyll fluorescence characteristics are evaluated in arbitrary unit (a. u.), or in other words, relative unit (r. u.).\***

| Chlorophyll Fluorescence Characteristic                                      |  | Equation                         |                                   |
|--|--|----------------------------------|-----------------------------------|
| <b>A. Extracted Fluorescence Parameters</b>                                  |  |                                  |                                   |
| 1  | Origin or initial or minimum fluorescence value (at 50 $\mu\text{s}$ )   | $F_0 = F_{50\mu\text{s}}$        |                                   |
| 2  | K-step fluorescence value (at 300 $\mu\text{s}$ )  | $F_K = F_{300\mu\text{s}}$       |                                   |
| 3  | J-step fluorescence value (at 2 ms)  | $F_J = F_{2\text{ms}}$           |                                   |
| 4  | I-step fluorescence value (at 30 ms)   | $F_I = F_{30\text{ms}}$          |                                   |
| 5  | Peak or maximum fluorescence value   | $F_P = F_M$                      |                                   |
| 6  | Time to reach $F_M$ , in ms  | $T_M = t_{F_M}$                  |                                   |
| 7  | Complementary area; The above area of the OKJIP graph between $F_0$ and $F_M$  | $\text{Area} = \text{Area}_M$    |                                   |
| <b>B. Technical Fluorescence Parameters</b>                                  |  |                                  |                                   |
| 8  | Variable fluorescence value  | $F_V$                            | $= F_M - F_0$                     |
| 9  | Non-photochemical loss in PSII in dark-adapted state; Ratio of the extrema   | LD                               | $= F_0/F_M$                       |
| 10   | Relative variable fluorescence at the definite time; Number of RCs closed at a definite time   | $V_T$                            | $= (F_T - F_0)/F_V$               |
| 11   | Relative variable fluorescence at the J-step (at 2 ms); Number of RCs, closed at the J-step  | $V_J$                            | $= (F_J - F_0)/F_V$               |
| 12   | Relative variable fluorescence at the I-step (at 30 ms); Number of RCs, closed at the I-step   | $V_I$                            | $= (F_I - F_0)/F_V$               |
| 13   | Initial slope; Maximum rate of primary photochemistry per RC; Net rate of RC closure; Observed rate of $Q_A$ reduction; Approximate value of the initial slope of relative variable fluorescence graph $V_T$ (for $F_0 = F_{50\mu\text{s}}$ )  | $M_0 = (dV/dt)_0$                | $= 4 \times (F_K - F_0)/F_V$      |
| 14   | Relative pool size of PQ; Normalized complementary area (assumed proportion for the number of reduction and oxidation of one $Q_A^-$ molecule during the fast OKJIP transient, and therefore related to the number of electron carriers per electron transport chain); Ability to transport electrons per reaction center; Multiple turnover of $Q_A$ for the RCs' closure; A measure of the energy needed to close all reaction centers | $S_M = EC_0/RC$                  | $= \text{Area}_M/F_V$             |
| 15   | Turnover number of $Q_A$ ; Total number of electrons transferred into electron transport chain; Number of reduction turnovers, oxidation, and re-reduction of $Q_A$ in the time between turning on the light and reaching the $F_M$ ( $T_M$ )  | N                                | $= M_0 \times (1/V_J) \times S_M$ |
| <b>C. Flux Ratios; Quantum Yields and Quantum Efficiencies/Probabilities</b> |  |                                  |                                   |
| 16   | Maximum quantum yield of primary PSII photochemistry (relative to $F_0$ ); PSII activity; Leaf photosynthetic capacity   | $TR_0/DI_0$                      | $= F_V/F_0$                       |
| 17   | Maximum quantum yield of primary PSII photochemistry (relative to $F_M$ ); Maximum efficiency with which an absorbed photon results in $Q_A$ reduction   | $\Phi_{P_0} = \Phi_M = TR_0/ABS$ | $= 1 - LD = F_V/F_M$              |
| 18   | Quantum yield of electron transport flux from $Q_A$ to $Q_B$ of PSII; Efficiency with which an absorbed photon results in electron transport beyond $Q_A^-$ ; Probability that an absorbed photon will move an electron into electron transport further than $Q_A^-$   | $\Phi_{E_0} = ET_0/ABS$          | $= \Psi_{E_0} \times \Phi_{P_0}$  |
| 19   | Quantum yield of reduction in the end electron acceptors of PSI per photon absorbed; Quantum yield of electron transport from $Q_A^-$ to the end electron acceptors of PSI; Quantum yield of electron transport flux until electron acceptors of PSI   | $\Phi_{R_0} = RE_0/ABS$          | $= \Psi_{R_0} \times \Phi_{P_0}$  |
| 20   | Quantum yield of dissipation; Quantum yield of energy dissipation as heat  | $\Phi_{D_0} = DI_0/ABS$          | $= 1 - \Phi_{P_0}$                |

Table-1 continue

|  |   |   |   |   |
|--|---|---|---|---|
| 21   | Efficiency or probability with which a PSII trapped electron is transferred from $Q_A$ to $Q_B$ ; Number of reaction centers open at the J-step (at 2 ms)                               | $\Psi_{E_0} = \Psi_{O_0} = ET_{O_0}/TR_{O_0}$     | = | $1 - V_j$   |
| 22   | Efficiency or probability with which a PSII trapped electron is transferred from $Q_A^-$ to the end electron acceptors of PSI; Number of reaction centers open at the I-step (at 30 ms) | $\Psi_{R_0} = \rho_0 = RE_{O_0}/TR_{O_0}$         | = | $1 - V_i$   |
| 23   | Efficiency or probability with which an electron from $Q_B$ is transferred to the end electron acceptors of PSI   | $\delta_{R_0} = \delta_{O_0} = RE_{O_0}/ET_{O_0}$ | = | $\Psi_{R_0}/\Psi_{E_0}$   |
| 24   | Proportion of electron transport to energy dissipation as heat  | $ET_{O_0}/DI_{O_0}$                               | = | $(ET_{O_0}/RC)/(DI_{O_0}/RC)$   |
| <b>D. Specific Energy Fluxes; Specific Energy Activities</b>                   |   |   |   |   |
| 25   | Absorbed energy flux per RC; Absorbed photon flux per RC; Rate of photon absorption   | ABS/RC  | = | $M_0 \times (1/V_j) \times (1/\Phi_{P_0})$  |
| 26   | Trapped energy flux per RC; Maximum trapped exciton flux per PSII; Maximum rate of $Q_A$ reduction  | $TR_{O_0}/RC$                                     | = | $M_0 \times (1/V_j)$  |
| 27   | Electron transport flux per RC; Electron transport flux from $Q_A$ to $Q_B$ per PSII; Rate of electron transport beyond $Q_A^-$   | $ET_{O_0}/RC$                                     | = | $M_0 \times (1/V_j) \times \Psi_{E_0}$  |
| 28   | Reduction energy flux per RC; Reduction flux of the final PSI electron acceptors; Electron transport flux until PSI acceptors per PSII  | $RE_{O_0}/RC$                                     | = | $M_0 \times (1/V_j) \times \Psi_{R_0}$  |
| 29   | Energy dissipation as heat per RC; Rate of heat dissipation   | $DI_{O_0}/RC$                                     | = | $(ABS/RC) - (TR_{O_0}/RC)$  |
| <b>E. Phenomenological Energy Fluxes; Phenomenological Energy Activities**</b> |   |   |   |   |
| 30   | Absorbed energy flux per CS; Absorbed photon flux per CS  | ABS/CS  | = | $ABS/CS_{Chl} = Chl/CS$<br>or $ABS/CS_{F_0} = F_0$<br>or $ABS/CS_{F_M} = F_M$   |
| 31   | Trapped energy flux per CS; Maximum trapped exciton flux per CS   | $TR_{O_0}/CS$                                     | = | $(ABS/CS) \times \Phi_{P_0}$  |
| 32   | Electron transport flux per CS; Electron transport flux from $Q_A$ to $Q_B$ per CS  | $ET_{O_0}/CS$                                     | = | $(ABS/CS) \times \Phi_{P_0} \times \Psi_{E_0}$  |
| 33   | Reduction energy flux per CS; Electron transport flux until PSI acceptors per CS  | $RE_{O_0}/CS$                                     | = | $(ABS/CS) \times \Phi_{P_0}$  |
| 34   | Energy dissipation as heat per CS   | $DI_{O_0}/CS$                                     | = | $(ABS/CS) - (TR_{O_0}/CS)$  |
| <b>F. Density or Amount or Concentration of Active PSII Reaction Centers**</b> |   |   |   |   |
| 35   | Density or amount or concentration of RCs per ABS; Number of $Q_A$ reducing RCs per PSII antenna chlorophyll  | RC/ABS  | = | $V_j \times (1/M_0) \times \Phi_{P_0}$  |
| 36   | Density or amount or concentration of RCs per CS; Number of RCs per CS  | RC/CS   | = | $(ABS/CS) \times (RC/ABS)$  |
| <b>G. Performance Indices; Vitality Indices**</b>                              |   |   |   |   |
| 37   | PSII vitality index of electron flux per ABS  | $PI_{ABS} = PI_{ABS, PSII}$                       | = | $(RC/ABS) \times [\Phi_{P_0}/(1 - \Phi_{P_0})] \times [\Psi_{E_0}/(1 - \Psi_{E_0})]$  |
| 38   | Total vitality index of electron flux per ABS   | $PI_{ABS, total}$                                 | = | $PI_{ABS, PSII} \times (\delta_{R_0}/(1 - \delta_{R_0}))$   |
| 39   | PSI vitality index of electron flux per ABS   | $PI_{ABS, PSI}$                                   | = | $PI_{ABS, total} - PI_{ABS, PSII}$  |
| 40   | PSII vitality index of electron flux on CS basis  | $PI_{CS} = PI_{CS, PSII}$                         | = | $(RC/CS) \times [\Phi_{P_0}/(1 - \Phi_{P_0})] \times [\Psi_{E_0}/(1 - \Psi_{E_0})]$   |
| 41   | Total vitality index of electron flux on CS basis   | $PI_{CS, total}$                                  | = | $PI_{CS, PSII} \times (\delta_{R_0}/(1 - \delta_{R_0}))$  |
| 42   | PSI vitality index of electron flux on CS basis   | $PI_{CS, PSI}$                                    | = | $PI_{CS, total} - PI_{CS, PSII}$  |
| <b>H. Driving Forces (for Photochemical Activities)**</b>                      |   |   |   |   |
| 43   | PSII redox (oxido-reduction) potential per ABS  | $DF_{ABS} = DF_{ABS, PSII}$                       | = | $\log(PI_{ABS, PSII}) = \log(RC/ABS) + \log(\Phi_{P_0}/(1 - \Phi_{P_0})) + \log(\Psi_{E_0}/(1 - \Psi_{E_0})) = DF_{RC} + DF_{\Phi} + DF_{\Psi}$ |
| 44   | Total redox (oxido-reduction) potential per ABS   | $DF_{ABS, total}$                                 | = | $\log(PI_{ABS, total})$   |
| 45   | PSI redox (oxido-reduction) potential per ABS   | $DF_{ABS, PSI}$                                   | = | $DF_{ABS, total} - DF_{ABS, PSII}$  |
| 46   | PSII redox (oxido-reduction) potential per CS   | $DF_{CS} = DF_{CS, PSII}$                         | = | $\log(PI_{CS, PSII}) = \log(ABS/CS) + \log(PI_{ABS, PSII}) = DF_{RC} + DF_{\Phi} + DF_{\Psi} + DF_{Chl}$  |
| 47   | Total redox (oxido-reduction) potential per CS  | $DF_{CS, total}$                                  | = | $\log(PI_{CS, total})$  |
| 48   | PSI redox (oxido-reduction) potential per CS  | $DF_{CS, PSI}$                                    | = | $DF_{CS, total} - DF_{CS, PSII}$  |

\* RC, active PSII reaction center; CS, excited cross section of leaf area; ABS, absorption energy flux; TR, excitation energy flux trapped by a RC and utilized for the reduction of  $Q_A$  to  $Q_A^-$ ; ET, flux of electrons from  $Q_A^-$  into the electron transport chain; RE, reduction of end electron acceptors at the PSI electron acceptor side: NADP (nicotinamide adenine dinucleotide phosphate) and Fd (ferredoxin); DI, dissipation energy flux as heat; EC, ability to electron conductance; PI, performance index; DF, deriving force; O, onset or origin of fluorescence induction; M, maximum of fluorescence induction; T, fluorescence induction at the definite time; Chl, chlorophyll; PSI, photosystem I; PSII, photosystem II;  $Q_A$ , quinone A;  $Q_A^-$ , reduced quinone A;  $Q_B$ , quinone B; PQ, plastoquinone.

\*\* All parameters in parts E, F, G, & H were calculated based on  $F_M$ .

## Data Analysis

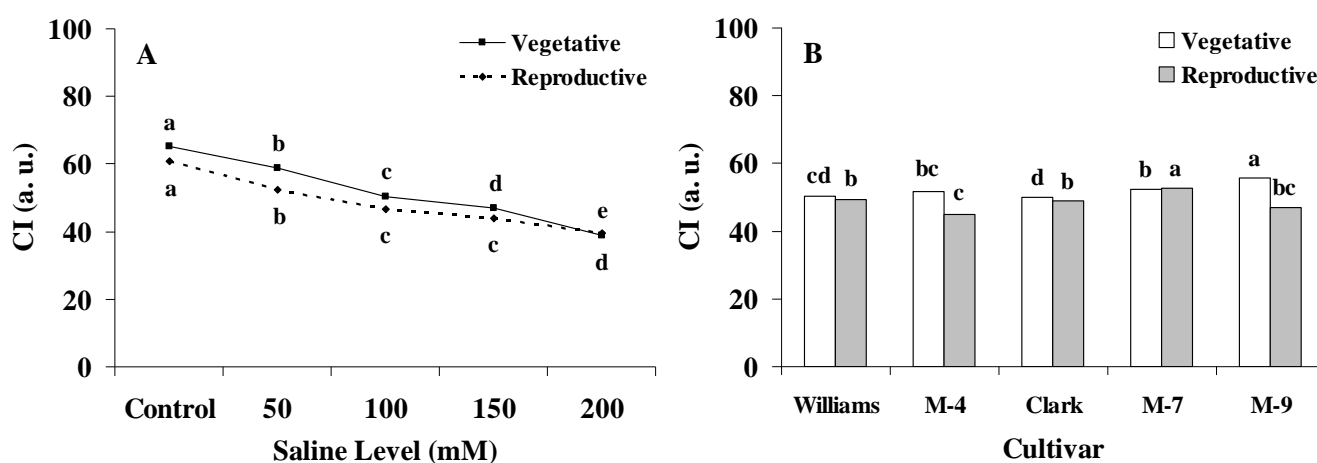
Data were subjected to the general linear model (GLM) for analysis of variance using SAS 9.1 software (SAS Institute Inc., Cary, NC, US, 2003). Significant differences among individual means were determined based on Fisher's protected least significant difference (LSD) test at  $P=0.05$  level and correlation analysis was done considering significance of Pearson's coefficients ( $P=0.05$ ).

## RESULTS AND DISCUSSION

### Chlorophyll index

Chlorophyll index (CI), as an indicator of the leaf chlorophyll content, significantly diminished by enhancing NaCl concentration at both phenological phases; however, there was no meaningful difference between 100 and 150 mM NaCl at reproductive phase (figure 1.A). Although the mutants contained more chlorophyll rather than their wild-types at both phases, the least chlorophyll index at reproductive phase belonged to the mutant M-4 (figure 1. B). This result was a sign of further saline tolerance in mutants comparing to the wild-types. The interactions of salinity  $\times$  cultivar were not significant (not shown).

Besides Miransari and Smith [12], Wang et al. [26] understood salt stress degraded soybean chlorophyll content and chlorophyll index (CI). The studies of Florina et al. [4] and Karimi and Yusef-Zadeh [8] on tomato and grape cultivars, respectively, resulted in the same findings under saline conditions.



**Figure 1. Chlorophyll index variations among (A) salinity levels, and (B) wild-type and mutant soybean cultivars at two vegetative (V5: growing of fifth node and/or fifth trifoliolate leaf) and reproductive (R5: beginning of seed filling) phenological phases. In each line or colored column, identical letters indicate non-significant differences considering LSD (0.05).**

### Chlorophyll fluorescence

Entirely, at both vegetative and reproductive phases, the same trends were observed throughout fluorescence characteristics as increasing salinity worsened fluorescence parameters (tables 3, 4, 5, and 6). None of these parameters showed any significant differences among interactions (not presented).

Against time to maximum fluorescence ( $T_M$ ), the other extracted fluorescence parameters comprising of the minimum, maximum, variable, and also minimum to maximum fluorescence values ( $F_0$ ,  $F_M$ ,  $F_V$ , and  $LD$ , in turn) highly rose by rising salinity levels at either phenological phase, which remarked that further and faster solar energy dissipation occurred in the form of fluorescence when the stress intensified (table 3). The  $LD$  rising pointed out the salinity influenced minimum fluorescence value further than maximum one (table 3). This situation was a direct resultant of rising net rate of closure in reaction centers ( $M_0$ ) caused to greater closed reaction centers in both J and I steps ( $V_J$  and  $V_I$ ) (table 3).

These outcomes were compatible with chlorophyll index as chlorophyll index decreased with increasing saline stress (figure 1.A) meaning that the lower chlorophyll molecules for releasing excited electrons, the more energy emitting as fluorescence. Such a state was mainly due to the extra solar energy absorbed by photosynthetic

pigments, particularly chlorophylls and carotenoids, which had to be dissipated as fast as possible; otherwise, the photosynthetic structures would be damaged [7 and 16]. Between the two ways of energy dissipation (heat and fluorescence) in leaves, the fluorescence was much more rapid, actually. In addition, the declined reaction center densities per both absorption energy flux and cross section (RC/ABS and RC/CS, orderly), containing chlorophyll-a, were in agreement with the decreased chlorophyll index and increased fluorescence values (table 3).

Salinity reduced the relative pool size of plastoquinone ( $S_M$ ), a kind of electron carriers per electron transport chain, at both phenological phases, resulting in rising the turnover number of quinone-a (N) at vegetative phase (table 3) to compensate the low number of plastoquinones and quinones, probably. The N reducing at reproductive phase was possibly related to the reduced ability to electron transport through plant aging (table 3). Contrarily, the vegetative phase demonstrated a meaningful enhancement of N by increasing salinity, seemingly related to more  $Q_A$  turnover in young plants, which could support electron transport in high salinity levels. Likewise, the significantly lower N in less saline levels was presumably cause by the adequate  $Q_A$  for accepting electrons without requiring fast turning over from  $Q_A^-$  to  $Q_A$  for receiving new electrons (table 3), which possibly indicated fewer excited electrons during plant aging.

Apart from non-significant  $V_j$  and  $V_i$  at both phenological phases and non-significant  $S_M$  at vegetative phase, the mutant cultivars demonstrated satisfactory fluorescence characteristics than their wild-types, specifically M-9 (table 3).

These findings were similar to those of Yamane et al. [28] informed in rice which  $F_O$  and  $F_M$  decreased by the same NaCl concentrations. Kafi [6] reported diminished  $T_M$  with a strange fall in  $F_M$  of salt-influenced wheat [6].

As a whole, stress upgraded fluorescence values ( $F_O$ ,  $F_M$ ,  $F_V$ , etc.), reaction center closure in J-step ( $V_j$ ), and LD while it degraded plastoquinone pool size ( $S_M$ ) and reaction center densities per absorption energy flux (RC/ABS) in several investigations [5, 9, 10, 14, and 27].

**Table 3. Extracted and technical fluorescence parameters, besides density/amount of active PS II reaction centers of the wild-type and mutant soybean cultivars exposed to salinity at two vegetative (V5: growing of fifth node and/or fifth trifoliolate leaf) and reproductive (R5: beginning of seed filling) phenological phases.\***

| Extracted and Technical Fluorescence Parameters |                     |                     |                       |                      |                       |                      |                      |                      |                    |                   |                       |                      |
|---|---------------------|---------------------|-----------------------|----------------------|-----------------------|----------------------|----------------------|----------------------|--------------------|-------------------|-----------------------|----------------------|
| Stage   | $F_O$               |                     | $F_M$                 |                      | $F_V$                 |                      | $T_M$                |                      | LD                 |                   | $M_O$                 |                      |
|   | Veg                 | Rep                 | Veg                   | Rep                  | Veg                   | Rep                  | Veg                  | Rep                  | Veg                | Rep               | Veg                   | Rep                  |
| <b>Salinity</b>                                 |                     |                     |                       |                      |                       |                      |                      |                      |                    |                   |                       |                      |
| control   | 483.75 <sup>d</sup> | 644.35 <sup>d</sup> | 2998.45 <sup>c</sup>  | 3277.33 <sup>d</sup> | 2514.70 <sup>b</sup>  | 2632.98 <sup>c</sup> | 802.50 <sup>a</sup>  | 647.45 <sup>a</sup>  | 0.16 <sup>c</sup>  | 0.20 <sup>c</sup> | 0.35 <sup>d</sup>     | 0.74 <sup>c</sup>    |
| 50  | 509.60 <sup>c</sup> | 669.57 <sup>c</sup> | 3059.35 <sup>c</sup>  | 3460.88 <sup>c</sup> | 2549.75 <sup>b</sup>  | 2791.30 <sup>b</sup> | 741.60 <sup>b</sup>  | 637.70 <sup>a</sup>  | 0.17 <sup>c</sup>  | 0.19 <sup>d</sup> | 0.57 <sup>c</sup>     | 0.92 <sup>d</sup>    |
| 100   | 568.65 <sup>b</sup> | 757.82 <sup>b</sup> | 3221.65 <sup>b</sup>  | 3616.30 <sup>b</sup> | 2653.00 <sup>a</sup>  | 2858.48 <sup>a</sup> | 685.85 <sup>c</sup>  | 582.75 <sup>b</sup>  | 0.18 <sup>b</sup>  | 0.21 <sup>b</sup> | 0.79 <sup>b</sup>     | 1.09 <sup>c</sup>    |
| 150   | 590.85 <sup>b</sup> | 755.97 <sup>b</sup> | 3267.30 <sup>b</sup>  | 3642.48 <sup>b</sup> | 2676.45 <sup>a</sup>  | 2886.50 <sup>a</sup> | 668.55 <sup>d</sup>  | 555.85 <sup>c</sup>  | 0.18 <sup>b</sup>  | 0.21 <sup>b</sup> | 0.79 <sup>b</sup>     | 1.12 <sup>b</sup>    |
| 200   | 685.85 <sup>a</sup> | 879.05 <sup>a</sup> | 3387.95 <sup>a</sup>  | 3770.80 <sup>a</sup> | 2702.10 <sup>a</sup>  | 2891.75 <sup>a</sup> | 611.70 <sup>c</sup>  | 524.80 <sup>d</sup>  | 0.20 <sup>a</sup>  | 0.23 <sup>a</sup> | 1.68 <sup>a</sup>     | 1.21 <sup>a</sup>    |
| <b>cultivar</b>                                 |                     |                     |                       |                      |                       |                      |                      |                      |                    |                   |                       |                      |
| Williams  | 587.65 <sup>a</sup> | 806.40 <sup>a</sup> | 3274.55 <sup>a</sup>  | 3766.20 <sup>a</sup> | 2686.90 <sup>a</sup>  | 2959.80 <sup>a</sup> | 717.90 <sup>a</sup>  | 624.35 <sup>a</sup>  | 0.18 <sup>ab</sup> | 0.21 <sup>a</sup> | 0.82 <sup>a</sup>     | 0.99 <sup>c</sup>    |
| M-4   | 579.45 <sup>a</sup> | 807.80 <sup>a</sup> | 3173.30 <sup>b</sup>  | 3758.60 <sup>a</sup> | 2593.85 <sup>bc</sup> | 2950.80 <sup>a</sup> | 704.60 <sup>bc</sup> | 590.55 <sup>b</sup>  | 0.198 <sup>a</sup> | 0.21 <sup>a</sup> | 0.86 <sup>a</sup>     | 1.00 <sup>c</sup>    |
| Clark   | 580.95 <sup>a</sup> | 807.27 <sup>a</sup> | 3259.60 <sup>a</sup>  | 3764.73 <sup>a</sup> | 2678.65 <sup>ab</sup> | 2957.45 <sup>a</sup> | 713.95 <sup>ab</sup> | 617.40 <sup>a</sup>  | 0.18 <sup>ab</sup> | 0.21 <sup>a</sup> | 0.83 <sup>a</sup>     | 1.00 <sup>c</sup>    |
| M-7   | 547.05 <sup>b</sup> | 673.90 <sup>b</sup> | 3148.05 <sup>bc</sup> | 3265.70 <sup>b</sup> | 2601.00 <sup>bc</sup> | 2591.80 <sup>b</sup> | 692.60 <sup>cd</sup> | 559.50 <sup>c</sup>  | 0.17 <sup>b</sup>  | 0.20 <sup>b</sup> | 0.84 <sup>a</sup>     | 1.03 <sup>b</sup>    |
| M-9   | 543.60 <sup>b</sup> | 611.40 <sup>c</sup> | 3079.20 <sup>c</sup>  | 3212.55 <sup>c</sup> | 2535.60 <sup>c</sup>  | 2601.15 <sup>b</sup> | 681.15 <sup>d</sup>  | 556.75 <sup>c</sup>  | 0.18 <sup>ab</sup> | 0.19 <sup>c</sup> | 0.83 <sup>a</sup>     | 1.05 <sup>a</sup>    |
|   |                     |                     |                       |                      |                       |                      |                      |                      |                    |                   |                       |                      |
| Stage   | $V_j$               |                     | $V_i$                 |                      | $S_M$                 |                      | N                    |                      | RC/ABS             |                   | RC/CS                 |                      |
|   | Veg                 | Rep                 | Veg                   | Rep                  | Veg                   | Rep                  | Veg                  | Rep                  | Veg                | Rep               | Veg                   | Rep                  |
| <b>Salinity</b>                                 |                     |                     |                       |                      |                       |                      |                      |                      |                    |                   |                       |                      |
| control   | 0.22 <sup>d</sup>   | 0.48 <sup>c</sup>   | 0.39 <sup>d</sup>     | 0.66 <sup>c</sup>    | 105.79 <sup>a</sup>   | 81.83 <sup>a</sup>   | 165.26 <sup>c</sup>  | 125.09 <sup>a</sup>  | 0.55 <sup>a</sup>  | 0.53 <sup>a</sup> | 1646.95 <sup>a</sup>  | 1719.04 <sup>a</sup> |
| 50  | 0.30 <sup>c</sup>   | 0.50 <sup>b</sup>   | 0.48 <sup>c</sup>     | 0.72 <sup>b</sup>    | 95.70 <sup>b</sup>    | 65.87 <sup>b</sup>   | 203.80 <sup>b</sup>  | 122.37 <sup>a</sup>  | 0.39 <sup>b</sup>  | 0.44 <sup>b</sup> | 1200.46 <sup>bc</sup> | 1508.07 <sup>b</sup> |
| 100   | 0.33 <sup>b</sup>   | 0.50 <sup>b</sup>   | 0.53 <sup>b</sup>     | 0.74 <sup>a</sup>    | 83.58 <sup>c</sup>    | 57.81 <sup>c</sup>   | 199.33 <sup>b</sup>  | 124.54 <sup>a</sup>  | 0.35 <sup>c</sup>  | 0.37 <sup>c</sup> | 1114.89 <sup>c</sup>  | 1327.06 <sup>d</sup> |
| 150   | 0.40 <sup>a</sup>   | 0.53 <sup>a</sup>   | 0.53 <sup>b</sup>     | 0.73 <sup>ab</sup>   | 76.77 <sup>d</sup>    | 54.13 <sup>d</sup>   | 164.62 <sup>c</sup>  | 115.33 <sup>b</sup>  | 0.38 <sup>b</sup>  | 0.37 <sup>c</sup> | 1248.23 <sup>b</sup>  | 1360.73 <sup>c</sup> |
| 200   | 0.40 <sup>a</sup>   | 0.50 <sup>b</sup>   | 0.64 <sup>a</sup>     | 0.72 <sup>b</sup>    | 68.02 <sup>c</sup>    | 48.45 <sup>c</sup>   | 309.84 <sup>a</sup>  | 116.44 <sup>b</sup>  | 0.18 <sup>c</sup>  | 0.32 <sup>d</sup> | 596.96 <sup>d</sup>   | 1209.91 <sup>c</sup> |
| <b>cultivar</b>                                 |                     |                     |                       |                      |                       |                      |                      |                      |                    |                   |                       |                      |
| Williams  | 0.31 <sup>a</sup>   | 0.50 <sup>a</sup>   | 0.51 <sup>a</sup>     | 0.72 <sup>a</sup>    | 86.89 <sup>a</sup>    | 60.05 <sup>b</sup>   | 210.38 <sup>ab</sup> | 114.54 <sup>bc</sup> | 0.37 <sup>ab</sup> | 0.41 <sup>a</sup> | 1184.61 <sup>a</sup>  | 1539.68 <sup>a</sup> |
| M-4   | 0.32 <sup>a</sup>   | 0.49 <sup>b</sup>   | 0.52 <sup>a</sup>     | 0.71 <sup>a</sup>    | 87.78 <sup>a</sup>    | 58.91 <sup>b</sup>   | 221.60 <sup>a</sup>  | 118.11 <sup>b</sup>  | 0.35 <sup>b</sup>  | 0.39 <sup>b</sup> | 1097.49 <sup>a</sup>  | 1457.63 <sup>b</sup> |
| Clark   | 0.31 <sup>a</sup>   | 0.50 <sup>a</sup>   | 0.51 <sup>a</sup>     | 0.72 <sup>a</sup>    | 87.43 <sup>a</sup>    | 59.53 <sup>b</sup>   | 210.28 <sup>ab</sup> | 114.12 <sup>c</sup>  | 0.37 <sup>ab</sup> | 0.41 <sup>a</sup> | 1188.12 <sup>a</sup>  | 1528.36 <sup>a</sup> |
| M-7   | 0.31 <sup>a</sup>   | 0.51 <sup>a</sup>   | 0.52 <sup>a</sup>     | 0.71 <sup>a</sup>    | 83.26 <sup>a</sup>    | 65.17 <sup>a</sup>   | 202.42 <sup>b</sup>  | 127.56 <sup>a</sup>  | 0.37 <sup>ab</sup> | 0.41 <sup>a</sup> | 1157.91 <sup>a</sup>  | 1316.61 <sup>c</sup> |
| M-9   | 0.31 <sup>a</sup>   | 0.51 <sup>a</sup>   | 0.52 <sup>a</sup>     | 0.71 <sup>a</sup>    | 84.50 <sup>a</sup>    | 64.42 <sup>a</sup>   | 198.16 <sup>b</sup>  | 129.42 <sup>a</sup>  | 0.38 <sup>a</sup>  | 0.40 <sup>a</sup> | 1179.38 <sup>a</sup>  | 1282.53 <sup>d</sup> |

\* In each column, identical letters point out non-significant differences considering LSD (0.05).

With the exception of the dissipation quantum yield ( $\Phi_{D_0}$ ), the whole quantum yields and efficiencies generally dropped with salinity increasing at either phenological phase. Additionally at both phases, most of the quantum yields and efficiencies responded similarly in 100 and 150 mM NaCl (table 4).

Salinization degraded quantum yield of electron transport flux ( $\Phi_{E_0}$ ) and quantum yield of reduction in end electron acceptors of photosystem I (PSI) ( $\Phi_{R_0}$ ), which displayed a fall in electron transport from  $Q_A$  to  $Q_B$  ( $\Phi_{E_0}$ ) and from reduced  $Q_A$  ( $Q_{A^-}$ ) to the end acceptors of PSI ( $\Phi_{R_0}$ ), in order. Moreover, the efficiencies with which an electron transferred from  $Q_A$  to  $Q_B$  ( $\Psi_{E_0}$ ), from  $Q_B$  to the end electron acceptors of PSI ( $\delta_{R_0}$ ), and from reduced  $Q_A$  ( $Q_{A^-}$ ) to the end electron acceptors of PSI ( $\Psi_{R_0}$ ), diminished with salinity enhancement (table 4). Besides, less  $\Psi_{E_0}$  and  $\Psi_{R_0}$  notified that there were fewer open reaction centers at J and I steps (table 3); therefore, fewer excited electrons could transmit to the electron transport chain. These effects eventually resulted in less maximum quantum yield of primary PSII photochemistry compared to both minimum ( $F_0$ ) and maximum ( $F_M$ ) fluorescence ( $TR_0/DI_0$  and  $\Phi_{P_0}$ , respectively) as indicators of PSII activity or leaf photosynthetic capacity (table 4).

According to chlorophyll index depression, as a consequence of saline stress (table 4), it seemed that reduced quantum yields and efficiencies were caused by dropping both electron acceptor amounts i. e.  $Q_A$ ,  $Q_B$ , PQ, etc. and their electron transferal capacity. The decrease in reaction center densities (RC/ABS and RC/CS), quantum yields and quantum efficiencies expressed both photon absorption and electron transport decreased under salinity; however, the electron transport fell relatively further. In fact, electron acceptors were so few that they could even transfer few electrons difficultly. Accordingly, it might be concluded salinity injured the photosynthetic electron transport chain greater in comparison with reaction centers.

By decreasing electron transport per electron chain, the extra-absorbed solar energy, which could not have been transformed to electron flux, had to be dissipated in the form of either fluorescence or heat not to destroy photosynthetic structures. Consequently, in accordance with fluorescence values (table 3), the ratio of electron transport to energy dissipation as heat ( $ET_0/DI_0$ ) dropped while quantum yield of dissipation as heat ( $\Phi_{D_0}$ ) enhanced, simultaneously (table 4). Aside from  $\Psi_{E_0}$ ,  $\Psi_{R_0}$ , and vegetative  $\delta_{R_0}$ , which were not significant, M-9 performed mostly best among the other cultivars, especially at reproductive phase (table 4).

Netondo et al. [13] and Yamane et al. [28] pronounced  $\Phi_{P_0}$  of sorghum and rice, in turn, diminished significantly in salinity greater than 150 mM NaCl, which might have been related to structural variations in PSII; however, the electron transport experienced a mild drop probably due to low impressionability of electron transportation. Yamane et al. [28] related such variations to thylakoid swelling in chloroplasts of mesophyll cells exposed to salt, which injured the thylakoid membranes and grana arrangement. Substantially, whatever the salinity rose, the swollen thylakoids extended in chloroplasts. These damages were induced by lipid peroxidation followed by reactive oxygen species (ROS) generation, mainly hydrogen peroxide and hydroxyl radical, which were among the ordinary ROS seen under salinity stress [28].

Kafi [6] discovered the same results which  $\Phi_{P_0}$  significantly fell with rising NaCl concentration but it did not yet vary among treatments until 100 mM. Less  $\Phi_{P_0}$  was attributed to RuBP (ribulose-1,5-bisphosphate) regeneration disturbance, which was highly dependent on electron transmission from PSII to electron acceptors [6]. Besides, Ripley et al. [14] said the ratio of electron transport to heat dissipation ( $ET_0/DI_0$ ) in sorghum decreased owing to low phosphorous, which was in accordance with the current findings. Moreover, Milivojevic al. [11] found out increasing in arsenic as an injurious element significantly diminished soybean  $TR_0/DI_0$  via lacking phosphorus absorption, the essential element for all energy cycles including photophosphorylation reactions.

On the other side, considering that water is the main media for all biological reactions, drought induced by saline stress could be another cause of decreasing quantum yields and efficiencies, which was announced by Wang et al. in maize [27].

**Table 4. Flux ratios (quantum yields and quantum efficiencies/probabilities) of the wild-type and mutant soybean cultivars exposed to salinity at two vegetative (V5: growing of fifth node and/or fifth trifoliate leaf) and reproductive (R5: beginning of seed filling) phenological phases.\***

| Flux Ratios (Quantum Yields and Quantum Efficiencies/Probabilities) |                                  |                   |                            |                    |                            |                    |                                  |                    |                            |                   |
|---|----------------------------------|-------------------|----------------------------|--------------------|----------------------------|--------------------|----------------------------------|--------------------|----------------------------|-------------------|
| Stage   | TR <sub>O</sub> /DI <sub>O</sub> |                   | Φ <sub>P<sub>o</sub></sub> |                    | Φ <sub>E<sub>o</sub></sub> |                    | Φ <sub>R<sub>o</sub></sub>       |                    | Φ <sub>D<sub>o</sub></sub> |                   |
|   | Veg                              | Rep               | Veg                        | Rep                | Veg                        | Rep                | Veg                              | Rep                | Veg                        | Rep               |
| <b>Salinity</b>   |                                  |                   |                            |                    |                            |                    |                                  |                    |                            |                   |
| control   | 5.22 <sup>a</sup>                | 4.13 <sup>b</sup> | 0.84 <sup>a</sup>          | 0.80 <sup>b</sup>  | 0.65 <sup>a</sup>          | 0.42 <sup>a</sup>  | 0.51 <sup>a</sup>                | 0.27 <sup>a</sup>  | 0.16 <sup>c</sup>          | 0.20 <sup>c</sup> |
| 50  | 5.00 <sup>a</sup>                | 4.21 <sup>a</sup> | 0.83 <sup>a</sup>          | 0.81 <sup>a</sup>  | 0.61 <sup>b</sup>          | 0.41 <sup>a</sup>  | 0.44 <sup>b</sup>                | 0.23 <sup>b</sup>  | 0.17 <sup>c</sup>          | 0.19 <sup>d</sup> |
| 100   | 4.67 <sup>b</sup>                | 3.80 <sup>c</sup> | 0.82 <sup>b</sup>          | 0.79 <sup>c</sup>  | 0.55 <sup>c</sup>          | 0.39 <sup>b</sup>  | 0.39 <sup>c</sup>                | 0.21 <sup>c</sup>  | 0.18 <sup>b</sup>          | 0.21 <sup>b</sup> |
| 150   | 4.53 <sup>b</sup>                | 3.84 <sup>c</sup> | 0.82 <sup>b</sup>          | 0.79 <sup>c</sup>  | 0.52 <sup>d</sup>          | 0.37 <sup>c</sup>  | 0.39 <sup>c</sup>                | 0.21 <sup>c</sup>  | 0.18 <sup>b</sup>          | 0.21 <sup>b</sup> |
| 200   | 4.01 <sup>c</sup>                | 3.30 <sup>d</sup> | 0.80 <sup>c</sup>          | 0.77 <sup>d</sup>  | 0.50 <sup>c</sup>          | 0.38 <sup>c</sup>  | 0.28 <sup>d</sup>                | 0.22 <sup>bc</sup> | 0.20 <sup>a</sup>          | 0.23 <sup>a</sup> |
| <b>cultivar</b>   |                                  |                   |                            |                    |                            |                    |                                  |                    |                            |                   |
| Williams  | 4.64 <sup>ab</sup>               | 3.70 <sup>c</sup> | 0.82 <sup>ab</sup>         | 0.78 <sup>c</sup>  | 0.56 <sup>ab</sup>         | 0.37 <sup>c</sup>  | 0.40 <sup>a</sup>                | 0.22 <sup>b</sup>  | 0.17 <sup>ab</sup>         | 0.21 <sup>a</sup> |
| M-4   | 4.54 <sup>b</sup>                | 3.68 <sup>c</sup> | 0.81 <sup>b</sup>          | 0.78 <sup>c</sup>  | 0.55 <sup>b</sup>          | 0.40 <sup>a</sup>  | 0.39 <sup>a</sup>                | 0.23 <sup>ab</sup> | 0.18 <sup>a</sup>          | 0.21 <sup>a</sup> |
| Clark   | 4.67 <sup>ab</sup>               | 3.69 <sup>c</sup> | 0.82 <sup>ab</sup>         | 0.78 <sup>c</sup>  | 0.56 <sup>ab</sup>         | 0.38 <sup>bc</sup> | 0.40 <sup>a</sup>                | 0.22 <sup>b</sup>  | 0.17 <sup>ab</sup>         | 0.21 <sup>a</sup> |
| M-7   | 4.83 <sup>a</sup>                | 3.89 <sup>b</sup> | 0.83 <sup>a</sup>          | 0.79 <sup>b</sup>  | 0.57 <sup>a</sup>          | 0.38 <sup>bc</sup> | 0.40 <sup>a</sup>                | 0.23 <sup>ab</sup> | 0.16 <sup>b</sup>          | 0.20 <sup>b</sup> |
| M-9   | 4.74 <sup>ab</sup>               | 4.32 <sup>a</sup> | 0.82 <sup>ab</sup>         | 0.81 <sup>a</sup>  | 0.56 <sup>ab</sup>         | 0.39 <sup>ab</sup> | 0.39 <sup>a</sup>                | 0.24 <sup>a</sup>  | 0.17 <sup>ab</sup>         | 0.19 <sup>c</sup> |
| <b>Ψ<sub>E<sub>o</sub></sub></b>                                    |                                  |                   |                            |                    |                            |                    |                                  |                    |                            |                   |
| Stage   | Ψ <sub>E<sub>o</sub></sub>       |                   | Ψ <sub>R<sub>o</sub></sub> |                    | δ <sub>R<sub>o</sub></sub> |                    | ET <sub>O</sub> /DI <sub>O</sub> |                    |                            |                   |
|   | Veg                              | Rep               | Veg                        | Rep                | Veg                        | Rep                | Rep                              | Rep                | Rep                        | Rep               |
| <b>Salinity</b>   |                                  |                   |                            |                    |                            |                    |                                  |                    |                            |                   |
| control   | 0.78 <sup>a</sup>                | 0.52 <sup>a</sup> | 0.61 <sup>a</sup>          | 0.34 <sup>a</sup>  | 0.78 <sup>a</sup>          | 0.66 <sup>a</sup>  | 4.06 <sup>a</sup>                | 2.13 <sup>a</sup>  |                            |                   |
| 50  | 0.73 <sup>b</sup>                | 0.50 <sup>b</sup> | 0.52 <sup>b</sup>          | 0.28 <sup>b</sup>  | 0.72 <sup>c</sup>          | 0.56 <sup>b</sup>  | 3.65 <sup>b</sup>                | 2.12 <sup>a</sup>  |                            |                   |
| 100   | 0.67 <sup>c</sup>                | 0.49 <sup>b</sup> | 0.47 <sup>c</sup>          | 0.26 <sup>c</sup>  | 0.70 <sup>c</sup>          | 0.53 <sup>c</sup>  | 3.12 <sup>c</sup>                | 1.87 <sup>b</sup>  |                            |                   |
| 150   | 0.63 <sup>d</sup>                | 0.47 <sup>c</sup> | 0.47 <sup>c</sup>          | 0.27 <sup>bc</sup> | 0.75 <sup>b</sup>          | 0.57 <sup>b</sup>  | 2.86 <sup>d</sup>                | 1.82 <sup>b</sup>  |                            |                   |
| 200   | 0.63 <sup>d</sup>                | 0.50 <sup>b</sup> | 0.35 <sup>d</sup>          | 0.28 <sup>b</sup>  | 0.56 <sup>d</sup>          | 0.57 <sup>b</sup>  | 2.53 <sup>e</sup>                | 1.64 <sup>c</sup>  |                            |                   |
| <b>cultivar</b>   |                                  |                   |                            |                    |                            |                    |                                  |                    |                            |                   |
| Williams  | 0.69 <sup>a</sup>                | 0.49 <sup>b</sup> | 0.49 <sup>a</sup>          | 0.28 <sup>a</sup>  | 0.71 <sup>a</sup>          | 0.57 <sup>ab</sup> | 3.22 <sup>ab</sup>               | 1.83 <sup>c</sup>  |                            |                   |
| M-4   | 0.68 <sup>a</sup>                | 0.51 <sup>a</sup> | 0.48 <sup>a</sup>          | 0.29 <sup>a</sup>  | 0.70 <sup>a</sup>          | 0.56 <sup>b</sup>  | 3.13 <sup>b</sup>                | 1.89 <sup>bc</sup> |                            |                   |
| Clark   | 0.69 <sup>a</sup>                | 0.49 <sup>b</sup> | 0.49 <sup>a</sup>          | 0.28 <sup>a</sup>  | 0.71 <sup>a</sup>          | 0.57 <sup>ab</sup> | 3.22 <sup>ab</sup>               | 1.83 <sup>c</sup>  |                            |                   |
| M-7   | 0.69 <sup>a</sup>                | 0.49 <sup>b</sup> | 0.49 <sup>a</sup>          | 0.29 <sup>a</sup>  | 0.70 <sup>a</sup>          | 0.59 <sup>a</sup>  | 3.36 <sup>a</sup>                | 1.91 <sup>b</sup>  |                            |                   |
| M-9   | 0.69 <sup>a</sup>                | 0.49 <sup>b</sup> | 0.48 <sup>a</sup>          | 0.29 <sup>a</sup>  | 0.70 <sup>a</sup>          | 0.59 <sup>a</sup>  | 3.29 <sup>ab</sup>               | 2.12 <sup>a</sup>  |                            |                   |

\* In each column, identical letters point out non-significant differences considering LSD (0.05).

While reduction energy flux per cross section (RE<sub>O</sub>/CS) at either phase besides electron transport flux per cross section (ET<sub>O</sub>/CS) at vegetative phase dropped, the other specific and phenomenological fluxes grew coordinately at both phases in response to increasing salinity. In addition, other than non-significant vegetative specific fluxes, all fluxes in mutants especially M-9 responded more appropriately rather than the wild-types (table 5).

It was presumed the decrease in reaction center densities caused by salinity eventuated in perfectly filling all reaction center capacity comparing to lower salinity levels, which ultimately enlarged ABS/RC, TR<sub>O</sub>/RC, and ET<sub>O</sub>/RC, in order. Even though the reaction centers and the electron acceptors dropped, it appeared the photosynthetic system could transfer a few excited electrons released by a few reaction centers.

On the other hand, ABS/CS and TR<sub>O</sub>/CS enhancement with NaCl indicated the cross section reached to the maximum potential for transferring electrons; hence, the electron acceptors were further and further involved in electron transferring relative to lower saline treatments. Actually, fewer electron acceptors in cross section (table 4) resulted in exceeding the proportion of absorbed and trapped energy fluxes to cross section (table 5).

ET<sub>O</sub>/CS trended oppositely at vegetative and reproductive phases. It seemed the younger plants had lower electron transporters in electron transport chain, which caused to completely fill electron transport capacity with increasing TR<sub>O</sub>/CS along salinity enhancement. Nevertheless, through plant aging, electron transporters appeared to enhance and could transport the electrons coordinately while TR<sub>O</sub>/CS increased by salinity (table 5).

The enhancement of ET<sub>O</sub>/RC eventuated in increasing RE<sub>O</sub>/RC, which demonstrate more end PSI electron acceptors in reaction centers reduced by excited electrons. On the contrary, RE<sub>O</sub>/CS displayed reduced trend with salinity enhancement, expressing electron transport flux toward PSI declined because of saturating electron acceptor capacity in electron transport chain (table 5).

It was natural that in such a situation, the energy dissipation as heat per either reaction center (DI<sub>O</sub>/RC) or cross section (DI<sub>O</sub>/CS) rose to disperse the excessive absorbed solar energy not to damage the photosynthetic structures (table 5) besides the fluorescence enhancement (table 3).



Ripley et al. [14] achieved similar consequences on sorghum but did not find any significant differences in  $ET_o/RC$ . Falqueto et al. [3] remarked that  $DI_o/RC$  could be affected by the proportion of active to inactive reaction centers; therefore,  $DI_o/RC$  could predict proportional amount of inactive reaction centers. Similar prediction could be done by  $DI_o/CS$  about relative rate of inactive cross section.

**Table 5. Specific fluxes/activities and phenomenological fluxes/activities of the wild-type and mutant soybean cultivars exposed to salinity at two vegetative (V5: growing of fifth node and/or fifth trifoliate leaf) and reproductive (R5: beginning of seed filling) phenological phases.\***

| Specific Energy Fluxes/Activities                |                       |                      |                       |                      |                       |                       |                       |                      |                     |                     |
|--|-----------------------|----------------------|-----------------------|----------------------|-----------------------|-----------------------|-----------------------|----------------------|---------------------|---------------------|
| Stage  | ABS/RC                |                      | TR <sub>o</sub> /RC   |                      | ET <sub>o</sub> /RC   |                       | RE <sub>o</sub> /RC   |                      | DI <sub>o</sub> /RC |                     |
|  | Veg                   | Rep                  | Veg                   | Rep                  | Veg                   | Rep                   | Veg                   | Rep                  | Veg                 | Rep                 |
| <b>Salinity</b>                                  |                       |                      |                       |                      |                       |                       |                       |                      |                     |                     |
| control  | 1.86 <sup>d</sup>     | 1.91 <sup>d</sup>    | 1.56 <sup>d</sup>     | 1.53 <sup>d</sup>    | 1.21 <sup>d</sup>     | 0.79 <sup>c</sup>     | 0.94 <sup>c</sup>     | 0.53 <sup>c</sup>    | 0.30 <sup>d</sup>   | 0.37 <sup>c</sup>   |
| 50   | 2.55 <sup>c</sup>     | 2.30 <sup>c</sup>    | 2.19 <sup>c</sup>     | 1.86 <sup>c</sup>    | 1.55 <sup>b</sup>     | 0.94 <sup>d</sup>     | 1.11 <sup>b</sup>     | 0.52 <sup>c</sup>    | 0.43 <sup>c</sup>   | 0.44 <sup>d</sup>   |
| 100  | 2.90 <sup>b</sup>     | 2.73 <sup>b</sup>    | 2.38 <sup>b</sup>     | 2.16 <sup>b</sup>    | 1.59 <sup>b</sup>     | 1.07 <sup>b</sup>     | 1.12 <sup>b</sup>     | 0.57 <sup>b</sup>    | 0.51 <sup>b</sup>   | 0.57 <sup>b</sup>   |
| 150  | 2.62 <sup>c</sup>     | 2.68 <sup>b</sup>    | 2.15 <sup>c</sup>     | 2.13 <sup>b</sup>    | 1.35 <sup>c</sup>     | 1.01 <sup>c</sup>     | 1.01 <sup>c</sup>     | 0.57 <sup>b</sup>    | 0.47 <sup>bc</sup>  | 0.55 <sup>c</sup>   |
| 200  | 5.74 <sup>a</sup>     | 3.13 <sup>a</sup>    | 4.58 <sup>a</sup>     | 2.40 <sup>a</sup>    | 2.90 <sup>a</sup>     | 1.19 <sup>a</sup>     | 1.63 <sup>a</sup>     | 0.68 <sup>a</sup>    | 1.16 <sup>a</sup>   | 0.73 <sup>a</sup>   |
| <b>cultivar</b>                                  |                       |                      |                       |                      |                       |                       |                       |                      |                     |                     |
| Williams   | 3.11 <sup>a</sup>     | 2.50 <sup>d</sup>    | 2.54 <sup>a</sup>     | 1.96 <sup>c</sup>    | 1.71 <sup>a</sup>     | 0.97 <sup>c</sup>     | 1.16 <sup>a</sup>     | 0.55 <sup>b</sup>    | 0.58 <sup>a</sup>   | 0.54 <sup>b</sup>   |
| M-4  | 3.26 <sup>a</sup>     | 2.62 <sup>a</sup>    | 2.65 <sup>a</sup>     | 2.05 <sup>ab</sup>   | 1.78 <sup>a</sup>     | 1.05 <sup>a</sup>     | 1.20 <sup>a</sup>     | 0.59 <sup>a</sup>    | 0.61 <sup>a</sup>   | 0.56 <sup>a</sup>   |
| Clark  | 3.09 <sup>a</sup>     | 2.51 <sup>cd</sup>   | 2.53 <sup>a</sup>     | 1.97 <sup>c</sup>    | 1.70 <sup>a</sup>     | 0.97 <sup>c</sup>     | 1.16 <sup>a</sup>     | 0.55 <sup>b</sup>    | 0.56 <sup>a</sup>   | 0.54 <sup>b</sup>   |
| M-7  | 3.14 <sup>a</sup>     | 2.55 <sup>bc</sup>   | 2.58 <sup>a</sup>     | 2.02 <sup>b</sup>    | 1.74 <sup>a</sup>     | 0.99 <sup>bc</sup>    | 1.17 <sup>a</sup>     | 0.57 <sup>ab</sup>   | 0.56 <sup>a</sup>   | 0.53 <sup>b</sup>   |
| M-9  | 3.07 <sup>a</sup>     | 2.56 <sup>b</sup>    | 2.51 <sup>a</sup>     | 2.07 <sup>a</sup>    | 1.68 <sup>a</sup>     | 1.02 <sup>ab</sup>    | 1.13 <sup>a</sup>     | 0.60 <sup>a</sup>    | 0.56 <sup>a</sup>   | 0.49 <sup>c</sup>   |
| <b>Phenomenological Energy Fluxes/Activities</b> |                       |                      |                       |                      |                       |                       |                       |                      |                     |                     |
| Stage  | ABS/CS                |                      | TR <sub>o</sub> /CS   |                      | ET <sub>o</sub> /CS   |                       | RE <sub>o</sub> /CS   |                      | DI <sub>o</sub> /CS |                     |
|  | Veg                   | Rep                  | Veg                   | Rep                  | Veg                   | Rep                   | Veg                   | Rep                  | Veg                 | Rep                 |
| <b>Salinity</b>                                  |                       |                      |                       |                      |                       |                       |                       |                      |                     |                     |
| control  | 2998.45 <sup>c</sup>  | 3277.33 <sup>d</sup> | 2514.70 <sup>b</sup>  | 2632.98 <sup>c</sup> | 1957.90 <sup>a</sup>  | 1365.23 <sup>b</sup>  | 1534.40 <sup>a</sup>  | 903.83 <sup>a</sup>  | 483.75 <sup>d</sup> | 644.35 <sup>d</sup> |
| 50   | 3059.35 <sup>c</sup>  | 3460.88 <sup>c</sup> | 2549.75 <sup>b</sup>  | 2791.30 <sup>b</sup> | 1863.40 <sup>b</sup>  | 1407.70 <sup>ab</sup> | 1338.50 <sup>b</sup>  | 782.70 <sup>bc</sup> | 509.60 <sup>c</sup> | 669.57 <sup>c</sup> |
| 100  | 3221.65 <sup>b</sup>  | 3616.30 <sup>b</sup> | 2653.00 <sup>a</sup>  | 2858.48 <sup>a</sup> | 1772.20 <sup>c</sup>  | 1417.98 <sup>a</sup>  | 1246.95 <sup>c</sup>  | 753.13 <sup>c</sup>  | 568.65 <sup>b</sup> | 757.82 <sup>b</sup> |
| 150  | 3267.30 <sup>b</sup>  | 3642.48 <sup>b</sup> | 2676.45 <sup>a</sup>  | 2886.50 <sup>a</sup> | 1688.20 <sup>d</sup>  | 1364.83 <sup>b</sup>  | 1263.80 <sup>bc</sup> | 777.30 <sup>bc</sup> | 590.85 <sup>b</sup> | 755.97 <sup>b</sup> |
| 200  | 3387.95 <sup>a</sup>  | 3770.80 <sup>a</sup> | 2702.10 <sup>a</sup>  | 2891.75 <sup>a</sup> | 1710.25 <sup>cd</sup> | 1435.75 <sup>a</sup>  | 959.25 <sup>d</sup>   | 816.63 <sup>b</sup>  | 685.85 <sup>a</sup> | 879.05 <sup>a</sup> |
| <b>cultivar</b>                                  |                       |                      |                       |                      |                       |                       |                       |                      |                     |                     |
| Williams   | 3274.55 <sup>a</sup>  | 3766.20 <sup>a</sup> | 2686.90 <sup>a</sup>  | 2959.80 <sup>a</sup> | 1846.25 <sup>a</sup>  | 1462.35 <sup>a</sup>  | 1310.70 <sup>a</sup>  | 834.45 <sup>a</sup>  | 587.65 <sup>a</sup> | 806.40 <sup>a</sup> |
| M-4  | 3173.30 <sup>b</sup>  | 3758.60 <sup>a</sup> | 2593.85 <sup>bc</sup> | 2950.80 <sup>a</sup> | 1770.55 <sup>ab</sup> | 1513.25 <sup>a</sup>  | 1239.60 <sup>ab</sup> | 858.35 <sup>a</sup>  | 579.45 <sup>a</sup> | 807.80 <sup>a</sup> |
| Clark  | 3259.60 <sup>a</sup>  | 3764.73 <sup>a</sup> | 2678.65 <sup>a</sup>  | 2957.45 <sup>a</sup> | 1838.43 <sup>a</sup>  | 1461.68 <sup>b</sup>  | 1305.83 <sup>ab</sup> | 836.93 <sup>a</sup>  | 580.95 <sup>a</sup> | 807.27 <sup>a</sup> |
| M-7  | 3148.05 <sup>bc</sup> | 3265.70 <sup>b</sup> | 2601.00 <sup>bc</sup> | 2591.80 <sup>b</sup> | 1793.80 <sup>ab</sup> | 1275.15 <sup>c</sup>  | 1264.35 <sup>ab</sup> | 750.45 <sup>b</sup>  | 547.05 <sup>b</sup> | 673.90 <sup>b</sup> |
| M-9  | 3079.20 <sup>c</sup>  | 3212.55 <sup>c</sup> | 2535.60 <sup>c</sup>  | 2601.15 <sup>b</sup> | 1742.93 <sup>b</sup>  | 1279.05 <sup>c</sup>  | 1222.43 <sup>b</sup>  | 753.40 <sup>b</sup>  | 543.60 <sup>b</sup> | 611.40 <sup>c</sup> |

\* In each column, identical letters point out non-significant differences considering LSD (0.05).

Totally, all vitality indices and redox potentials at both phenological phases revealed significant decreased responses to salinity increase (table 6) with no significances in interactions (data not shown). Within two phenological phases, the significant drop of whole vitality indices and redox potentials along salinity enhancement clearly explained the saline stress extremely affected the soybean photochemical components of photosynthetic apparatus. As the other fluorescence parameters (tables 3, 4, and 5), M-9 performed best with saline increasing; yet, M-9 performance was not significantly different from other cultivars in some traits (table 6).

Total vitality indices ( $PI_{ABS, total}$  and  $PI_{CS, total}$ ) detected the photosynthetic apparatus reflections up to the PSI end electron acceptors [23]. Van Heerden et al. [25] discovered a decrease in nitrogen-deficient soybean  $PI_{ABS}$  while soybean  $PI_{ABS}$  enhanced rapidly following nitrogen application. Ripley et al. [14] experienced  $PI_{ABS}$  reduction in eight- and twelve-week sorghum exposed to low phosphorous stress, which was owing to the significant reduction in  $\Phi_{Po}$ ,  $\Phi_{Eo}$ , and  $\Psi_{Eo}$ . Additionally, PSII vitality index was depended on the number of reaction centers. Consequently, photosystems consisted of more reaction center densities (RC/ABS) trapped energy more effectively ( $TR_o/ABS$ ) and presented efficient electron transport ( $ET_o/ABS$ ) besides their dissipated energy as heat or fluorescence ( $DI_o/RC$  and  $DI_o/ABS$ ) reduced [14].

Desotgiu et al. [2] announced that the decrease in leaf water potential followed by the stomata closure eventuated in  $PI_{ABS, total}$  declining, which had been previously affirmed as one of the salinity impacts. However, Ripley et al. [14] noted that restriction in amount of phosphorus-containing intermediates taking part in electron transport caused to decrease in vitality indices, which was ordinary. Furthermore, Van Heerden et al. [25] reported vitality indices and redox potentials of soybean genotypes were partially influenced by dark chilling stress in such a manner that could be utilized as rapid, economic facilitated indicators for genotype screening.

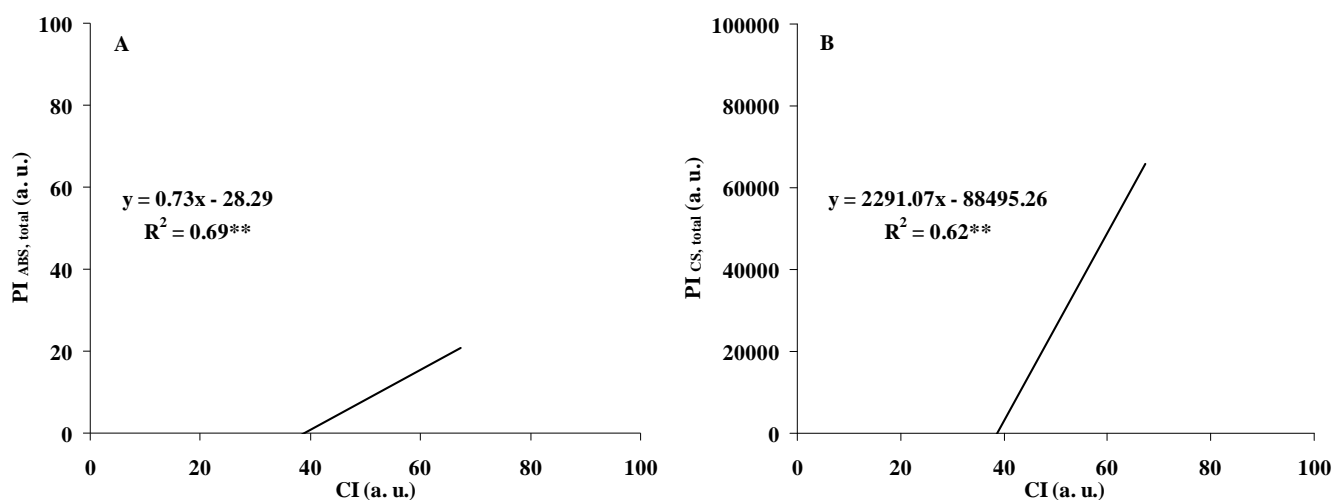
**Table 6. Performance/vitality indices and driving forces of the wild-type and mutant soybean cultivars exposed to salinity at two vegetative (V5: growing of fifth node and/or fifth trifoliolate leaf) and reproductive (R5: beginning of seed filling) phenological phases.\***

| Performance/Vitality Indices                         |                        |                   |                         |                    |                       |                    |                       |                      |                        |                       |                       |                       |
|--|------------------------|-------------------|-------------------------|--------------------|-----------------------|--------------------|-----------------------|----------------------|------------------------|-----------------------|-----------------------|-----------------------|
| Stage  | PI <sub>ABS,PSII</sub> |                   | PI <sub>ABS,total</sub> |                    | PI <sub>ABS,PSI</sub> |                    | PI <sub>CS,PSII</sub> |                      | PI <sub>CS,total</sub> |                       | PI <sub>CS,PSI</sub>  |                       |
|  | Veg                    | Rep               | Veg                     | Rep                | Veg                   | Rep                | Veg                   | Rep                  | Veg                    | Rep                   | Veg                   | Rep                   |
| <b>Salinity</b>                                      |                        |                   |                         |                    |                       |                    |                       |                      |                        |                       |                       |                       |
| control  | 10.36 <sup>a</sup>     | 2.32 <sup>a</sup> | 39.92 <sup>a</sup>      | 4.72 <sup>a</sup>  | 29.56 <sup>a</sup>    | 2.40 <sup>a</sup>  | 32020.00 <sup>a</sup> | 7600.80 <sup>a</sup> | 125980.00 <sup>a</sup> | 15394.80 <sup>a</sup> | 93960.00 <sup>a</sup> | 7794.00 <sup>a</sup>  |
| 50   | 5.34 <sup>b</sup>      | 1.86 <sup>b</sup> | 13.72 <sup>b</sup>      | 2.34 <sup>b</sup>  | 8.38 <sup>b</sup>     | 0.48 <sup>b</sup>  | 16361.00 <sup>b</sup> | 6432.30 <sup>b</sup> | 42156.00 <sup>b</sup>  | 8086.20 <sup>b</sup>  | 25794.00 <sup>b</sup> | 1653.90 <sup>b</sup>  |
| 100  | 3.25 <sup>c</sup>      | 1.37 <sup>c</sup> | 7.76 <sup>bc</sup>      | 1.55 <sup>c</sup>  | 4.51 <sup>bc</sup>    | 0.19 <sup>b</sup>  | 10482.00 <sup>c</sup> | 4941.40 <sup>c</sup> | 25063.00 <sup>bc</sup> | 5620.30 <sup>c</sup>  | 14581.00 <sup>b</sup> | 678.90 <sup>b</sup>   |
| 150  | 2.96 <sup>c</sup>      | 1.28 <sup>d</sup> | 8.84 <sup>bc</sup>      | 1.76 <sup>c</sup>  | 5.88 <sup>bc</sup>    | 0.47 <sup>b</sup>  | 9671.00 <sup>c</sup>  | 4668.60 <sup>c</sup> | 28915.00 <sup>bc</sup> | 6339.80 <sup>c</sup>  | 19244.00 <sup>b</sup> | 1671.20 <sup>b</sup>  |
| 200  | 1.21 <sup>d</sup>      | 1.04 <sup>e</sup> | 1.64 <sup>c</sup>       | 1.39 <sup>c</sup>  | 0.43 <sup>c</sup>     | 0.35 <sup>b</sup>  | 4098.00 <sup>d</sup>  | 3928.60 <sup>d</sup> | 5537.00 <sup>c</sup>   | 5235.10 <sup>c</sup>  | 1439.00 <sup>b</sup>  | 1306.50 <sup>b</sup>  |
| <b>cultivar</b>                                      |                        |                   |                         |                    |                       |                    |                       |                      |                        |                       |                       |                       |
| Williams   | 4.45 <sup>ab</sup>     | 1.52 <sup>b</sup> | 13.50 <sup>a</sup>      | 2.22 <sup>bc</sup> | 9.06 <sup>a</sup>     | 0.70 <sup>bc</sup> | 14130.00 <sup>a</sup> | 5656.50 <sup>a</sup> | 42622.00 <sup>a</sup>  | 8181.90 <sup>a</sup>  | 28492.00 <sup>a</sup> | 2525.40 <sup>ab</sup> |
| M-4  | 3.96 <sup>b</sup>      | 1.56 <sup>b</sup> | 11.21 <sup>a</sup>      | 2.10 <sup>c</sup>  | 7.25 <sup>a</sup>     | 0.55 <sup>c</sup>  | 12190.00 <sup>a</sup> | 5782.90 <sup>a</sup> | 34276.00 <sup>a</sup>  | 7811.40 <sup>a</sup>  | 22086.00 <sup>a</sup> | 2028.50 <sup>b</sup>  |
| Clark  | 4.48 <sup>ab</sup>     | 1.51 <sup>b</sup> | 13.48 <sup>a</sup>      | 2.17 <sup>bc</sup> | 9.01 <sup>a</sup>     | 0.66 <sup>bc</sup> | 14149.00 <sup>a</sup> | 5616.60 <sup>a</sup> | 42307.00 <sup>a</sup>  | 8027.60 <sup>a</sup>  | 28158.00 <sup>a</sup> | 2411.00 <sup>ab</sup> |
| M-7  | 5.03 <sup>ab</sup>     | 1.60 <sup>b</sup> | 16.73 <sup>a</sup>      | 2.53 <sup>ab</sup> | 11.69 <sup>a</sup>    | 0.96 <sup>ab</sup> | 15983.00 <sup>a</sup> | 5045.50 <sup>b</sup> | 54382.00 <sup>a</sup>  | 8034.10 <sup>a</sup>  | 38399.00 <sup>a</sup> | 2988.60 <sup>ab</sup> |
| M-9  | 5.21 <sup>a</sup>      | 1.73 <sup>a</sup> | 16.96 <sup>a</sup>      | 2.74 <sup>a</sup>  | 11.75 <sup>a</sup>    | 1.01 <sup>a</sup>  | 16179.00 <sup>a</sup> | 5470.00 <sup>a</sup> | 54064.00 <sup>a</sup>  | 8621.20 <sup>a</sup>  | 37885.00 <sup>a</sup> | 3151.20 <sup>a</sup>  |
| <b>Driving Forces (for Photochemical Activities)</b> |                        |                   |                         |                    |                       |                    |                       |                      |                        |                       |                       |                       |
| Stage  | DF <sub>ABS,PSII</sub> |                   | DF <sub>ABS,total</sub> |                    | DF <sub>ABS,PSI</sub> |                    | DF <sub>CS,PSII</sub> |                      | DF <sub>CS,total</sub> |                       | DF <sub>CS,PSI</sub>  |                       |
|  | Veg                    | Rep               | Veg                     | Rep                | Veg                   | Rep                | Veg                   | Rep                  | Veg                    | Rep                   | Veg                   | Rep                   |
| <b>Salinity</b>                                      |                        |                   |                         |                    |                       |                    |                       |                      |                        |                       |                       |                       |
| control  | 0.99 <sup>a</sup>      | 0.36 <sup>a</sup> | 1.55 <sup>a</sup>       | 0.66 <sup>a</sup>  | 0.55 <sup>a</sup>     | 0.29 <sup>a</sup>  | 4.47 <sup>a</sup>     | 3.89 <sup>a</sup>    | 5.02 <sup>a</sup>      | 4.17 <sup>a</sup>     | 0.55 <sup>a</sup>     | 0.29 <sup>a</sup>     |
| 50   | 0.72 <sup>b</sup>      | 0.27 <sup>b</sup> | 1.13 <sup>b</sup>       | 0.37 <sup>b</sup>  | 0.41 <sup>c</sup>     | 0.10 <sup>b</sup>  | 4.21 <sup>b</sup>     | 3.81 <sup>b</sup>    | 4.62 <sup>b</sup>      | 3.90 <sup>b</sup>     | 0.41 <sup>c</sup>     | 0.10 <sup>b</sup>     |
| 100  | 0.51 <sup>c</sup>      | 0.13 <sup>c</sup> | 0.89 <sup>c</sup>       | 0.19 <sup>c</sup>  | 0.37 <sup>c</sup>     | 0.05 <sup>c</sup>  | 4.02 <sup>c</sup>     | 3.69 <sup>c</sup>    | 4.39 <sup>c</sup>      | 3.74 <sup>cd</sup>    | 0.37 <sup>c</sup>     | 0.05 <sup>c</sup>     |
| 150  | 0.47 <sup>c</sup>      | 0.11 <sup>d</sup> | 0.94 <sup>c</sup>       | 0.23 <sup>c</sup>  | 0.47 <sup>b</sup>     | 0.12 <sup>b</sup>  | 3.98 <sup>c</sup>     | 3.67 <sup>d</sup>    | 4.46 <sup>c</sup>      | 3.79 <sup>c</sup>     | 0.47 <sup>b</sup>     | 0.12 <sup>b</sup>     |
| 200  | 0.08 <sup>d</sup>      | 0.02 <sup>e</sup> | 0.19 <sup>d</sup>       | 0.14 <sup>d</sup>  | 0.11 <sup>d</sup>     | 0.12 <sup>b</sup>  | 3.61 <sup>d</sup>     | 3.59 <sup>c</sup>    | 3.72 <sup>d</sup>      | 3.71 <sup>d</sup>     | 0.11 <sup>d</sup>     | 0.12 <sup>b</sup>     |
| <b>cultivar</b>                                      |                        |                   |                         |                    |                       |                    |                       |                      |                        |                       |                       |                       |
| Williams   | 0.55 <sup>ab</sup>     | 0.16 <sup>b</sup> | 0.95 <sup>a</sup>       | 0.29 <sup>b</sup>  | 0.39 <sup>a</sup>     | 0.13 <sup>ab</sup> | 4.07 <sup>a</sup>     | 3.74 <sup>ab</sup>   | 4.46 <sup>a</sup>      | 3.87 <sup>a</sup>     | 0.39 <sup>a</sup>     | 0.13 <sup>ab</sup>    |
| M-4  | 0.51 <sup>b</sup>      | 0.17 <sup>b</sup> | 0.89 <sup>a</sup>       | 0.29 <sup>b</sup>  | 0.37 <sup>a</sup>     | 0.12 <sup>b</sup>  | 4.02 <sup>b</sup>     | 3.75 <sup>a</sup>    | 4.39 <sup>a</sup>      | 3.87 <sup>a</sup>     | 0.37 <sup>a</sup>     | 0.18 <sup>b</sup>     |
| Clark  | 0.56 <sup>a</sup>      | 0.16 <sup>b</sup> | 0.96 <sup>a</sup>       | 0.29 <sup>b</sup>  | 0.40 <sup>a</sup>     | 0.13 <sup>ab</sup> | 4.07 <sup>a</sup>     | 3.74 <sup>ab</sup>   | 4.47 <sup>a</sup>      | 3.86 <sup>a</sup>     | 0.40 <sup>a</sup>     | 0.13 <sup>ab</sup>    |
| M-7  | 0.57 <sup>a</sup>      | 0.17 <sup>b</sup> | 0.95 <sup>a</sup>       | 0.33 <sup>ab</sup> | 0.39 <sup>a</sup>     | 0.16 <sup>a</sup>  | 4.07 <sup>a</sup>     | 3.69 <sup>c</sup>    | 4.45 <sup>a</sup>      | 3.85 <sup>a</sup>     | 0.38 <sup>a</sup>     | 0.16 <sup>a</sup>     |
| M-9  | 0.58 <sup>a</sup>      | 0.22 <sup>a</sup> | 0.95 <sup>a</sup>       | 0.38 <sup>a</sup>  | 0.37 <sup>a</sup>     | 0.16 <sup>a</sup>  | 4.06 <sup>ab</sup>    | 3.72 <sup>b</sup>    | 4.43 <sup>a</sup>      | 3.88 <sup>a</sup>     | 0.37 <sup>a</sup>     | 0.16 <sup>a</sup>     |

\*In each column, identical letters point out non-significant differences considering LSD (0.05).

## Relationships between photosynthetic pigment attributes

Total vitality indices (PI<sub>ABS, total</sub> and PI<sub>CS, total</sub>), indicating the photosynthetic pigment functions of the leaf, strongly correlated with chlorophyll index (figure 2). This consequence hinted the vitality index was highly dependent on chlorophyll content per either absorption or cross section.



**Figure 2. Correlation of chlorophyll index with (A) PSII absorption and cross section vitality indices (PI<sub>ABS, PSII</sub> and PI<sub>CS, PSII</sub>), (B) PSI absorption and cross section vitality indices (PI<sub>ABS, PSI</sub> and PI<sub>CS, PSI</sub>), and (C) total absorption and cross section vitality indices (PI<sub>ABS, total</sub> and PI<sub>CS, total</sub>) in the wild-type and mutant soybean cultivars exposed to salinity at two vegetative (V5: growing of fifth node and/or fifth trifoliolate leaf) and reproductive (R5: beginning of seed filling) phenological phases (P=0.05).**

## CONCLUDING REMARKS

Decreased chlorophyll index under saline condition influenced the fluorescence parameters of soybean adversely so that the least redox potential of photophosphorylation reactions ( $DF_{S_{ABS}}$  and  $DF_{S_{CS}}$ ) was observed in the highest salinity level. These consequences demonstrated the directly declining impact of salinity on reduction-oxidation rate of photosystem components, which ultimately might lead to diminish the photosynthesis. On the other hand, mutants were more tolerant to salinity rather than their wild-types. In addition, among mutants derived of the same wild-type (Williams), the later mutant (M-9) usually performed better than the former mutant (M-7), showing that the breeding process has been rather successful in this case.

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