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Analysis of Chlorophyll *a* Fluorescence of Two Mangrove Species of Vitória Bay (ES, Brazil) to Natural Variation of Tide

Análise de fluorescência da clorofila *a* de duas espécies de mangue da Baía de Vitória (ES, Brasil) influenciadas pela variação natural da maré

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Abstract

Leaf chlorophyll (Chl) *a* fluorescence parameters (JIP-test, initial fluorescence-*Fo* and potential quantum yield of PS2-*Fv/Fm*) of two mangroves species (*Laguncularia racemosa* Gaerth. and *Rhizophora mangle* L.) were compared in a basin forest, Vitória Bay (Brazil), during the dry and rainy seasons. Measurements were made three hours before high tide (H_1), during high tide (H_2) and three hours after high tide (H_3) using a Handy-PEA fluorometer (*Hansatech*, UK). Leaves were dark-adapted before measurements. Increases in salinities caused a decrease in *Fo* and an increase of *Fv/Fm* in both species and seasons. Increases of salinity around 29.8 % (during H_1), 64.45 % (H_2) and 78.6 % (H_3) (from dry to rainy season) caused an increasing of 12.3, 10.95 and 5.1 % of *Fv/Fm* values in *L. racemosa* and 1.4, 8.2 and 3.75 % in *R. mangle* (H_1, H_2 and H_3, respectively). However, at H_2, the energy fluxes for absorption (ABS/RC) and trapping (TR_0/RC) were significantly reduced in *L. racemosa*. Reductions in ABS/RC result in increase in antenna size of active RCs. In this study, reductions in ABS/RC were consistent to increases in *Fv/Fm* and reductions of energy dissipation at antenna chlorophyll level (DI_0/RC), which would explain the decline on *Fo* values. Fluctuations in salinity level of water during the day induced functional changes of PS2 in both species of mangroves. However, the slight variation of *Fv/Fm* in *L. racemosa* (from 12.3 % in H_1 to 10.95 % in H_2) associated with the decline in ABS/RC, TR_0/RC, and DI_0/RC indicate increased tolerance to salt in *L. racemosa*, in response to changes in the amplitude of tide.

Key words: basin forests, chlorophyll fluorescence, diurnal tide, JIP-test

Resumo

Parâmetros da fluorescência da clorofila *a* (teste JIP, fluorescência inicial- *Fo* e produção quântica potencial do fotossistema 2-*Fv/Fm*) de duas espécies do manguezal (*Laguncularia racemosa* Gaerth. e *Rhizophora mangle* L.) foram comparados em um bosque de bacia, Baía de Vitória (Brasil), durante as estações seca e chuvosa. As medidas foram feitas três horas antes da maré alta (H_1), durante a maré alta (H_2) e três horas após maré alta (H_3) usando um fluorômetro Handy-PEA (*Hansatech*, UK). As folhas foram pré-adaptadas ao escuro antes de cada medida. Aumentos na salinidade causaram um decréscimo em *Fo* e um aumento de *Fv/Fm* em ambas as espécies e estações. Aumentos da salinidade próximos de 29,8 % (durante H_1), 64,45 % (H_2) e 78,6 % (H_3) (da estação seca para a chuvosa) causou um aumento de 12.3, 10.95 e 5.1 % nos valores de *Fv/Fm* em *L. racemosa* e 1.4, 8.2 e 3.75 % em *R. mangle* (H_1, H_2 e H_3, respectivamente). Porém, em H_2, os fluxos de energia por absorção (ABS/RC) e captura (TR_0/RC) foram significativamente reduzidos em *L. racemosa*. Reduções em ABS/RC resultaram em aumentos no tamanho dos centros de reação da antena do FSII. Neste estudo, reduções em ABS/RC foram consistentes com os aumentos em *Fv/Fm* e reduções da dissipação da energia de excitação em nível de clorofila da antena (DI_0/RC), o que poderia explicar o declínio nos valores de *Fo*. Flutuações nos níveis de salinidade da água durante o dia induziram mudanças funcionais do FSII em ambas as espécies. Porém, a pequena variação de *Fv/Fm* em *L. racemosa* (12,3 % - 10,95 % em H_2) associado com o declínio em ABS/RC, TR_0/RC e DI_0/RC indica maior tolerância de *L. racemosa* ao estresse salino frente às mudanças na amplitude das marés.

Palavras-chave: bosque de bacia, fluorescência da clorofila, maré diurna, test JIP

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Introduction

Mangrove forest form a unique community in tropical coastal regions and tidal lowlands (TAKEMURA *et al*, 2000). They occupy about 17 million hectares worldwide and are considered among the most productive ecosystems (TUFFERS; NAIDOO; VON WILLERT, 2001). Ecologically, they are considered an essential component in protecting adjacent land from wave and storm erosion and are important components of detrital food chains vital to a variety of marine and estuarine communities (ODUM; HEALD, 1975; TAKEMURA *et al*, 2000).

Studies have established that soil salinity levels, nutrient availability, photosynthetic capacity and the degree of tidal inundation constitute the mainly factors affecting the growth and productivity of mangroves forest (SOBRADO, 1999; NAIDOO; TUFFERS; VON WILLERT, 2002). Although mangroves are adapted to seawater flooding, the frequency and duration of flooding have influenced important physiological processes of natural mangroves (Luzhen *et al.* 2005), such as anatomical development, gas exchange, biomass partitioning, enzymes activity and hormone synthesis (NAIDOO; ROGALLA; VON WILLERT, 1997; YE *et al*, 2004, 2010; CHEN; WANG; LIN, 2005; WANG *et al*, 2007; XIAO *et al*, 2009). In the last years, with the rise of sea level resultant of the rapid changes of climate, the periods of exposure of mangroves to high tide levels has been prolonged (ELLISON; FARNSWORTH, 1997).

The critical tidal level which the mangrove specie can tolerate varies between regions (HE *et al.*, 2007) and the morphological-physiological differences observed between species are consistent with the particular zone they occupy within mangrove ecosystems (BALL, 1986).

High tide is frequently associated to high salinities, affecting the cell turgor. However, many authors have showed that mangroves species are able to tolerate high salt levels (KRAUSS; ALLEN, 2003; SOBRADO, 2005) because they have developed morphological adaptations to salinity, e.g. salt glands, which contribute strongly for acquisition of salt tolerance and to differences in salt tolerance generally observed between species. On the other hand, non-secretors mangroves excluded until 99% of the salt by root ultrafiltration (PALIYAVUTH; CLOUGH; PATANAPONPAIBOON, 2004). A recent study also showed that *Kandelia candel* exhibited a higher capacity to restrict both salt uptake and subsequent accumulation while *Bruguiera gymnorhiza* had a higher capacity to exclude salts from mesophyll cells which resulted in lesser reduction of photosynthesis (LI *et al*, 2008). Salt tolerance level in mangroves likely depends on the NaCl concentration in the external solutions and the duration of salt exposure, associated with specific genetic characteristics (SOBRADO, 2005).

Rhizophora mangle L. and *Laguncularia racemosa* (L.) Gaerth. are dominant mangrove species along of Espírito Santo (ES) State coastlines (Brazil), where they are distributed along of basin forests occupying the medium to high inter-tidal zone of mangrove forest. Thus, are regularly influenced by variations of diurnal tide and interstitial water salinities ranged from 10 to 45 psu. Informations about tolerance to salt stress in these mangroves have been contradictory. Krauss and Allen (2003) affirm that high salinity tends to favor the development, establishment and physiology of mature *R. mangle* compared to *Bruguiera sexangula* (Hawaii, USA). In contrast, it appears that an enhanced salt gland secretion is responsible for higher salt tolerance in *L. racemosa* (SOBRADO, 2005). From the physiological aspects, no comparative study is available concerning the salinity responses of mature *R. mangle* and *L. racemosa* trees in basin forests at ES coastline. Although mangroves have been studied extensively in others regions of Brazil, the mangroves of ES coastline were only partially investigated by Silva, Bernini and Carmo (2005) in studies relating the structure of mangrove forests. The physiological responses of mangrove to natural variation of diurnal tide definitely need studies for a better understanding of the mangroves ecophysiology. Such information would be useful to understand the factors that influence the dominance of *R. mangle* and *L. racemosa* mangroves along ES coastline.

An important technique used to evaluate the photochemical performance of plants under high salinity conditions is the chlorophyll (Chl) *a* fluorescence (KRAUSE; WEIS, 1991). Chl *a* fluorescence provides information on the relationship between structure and function of photosystem 2 (PS2), reaction center (RC), and core complexes in plants salt stressed (YAMANE *et al*, 2000; MISRA; SRIVASTAVA; STRASSER, 2001, PANDA *et al*, 2006). Inhibition of electron transfer by PS2-inhibiting salinity levels causes a rapid chlorophyll fluorescence response, which can be measured using portable fluorometers on dark-adapted leaves. Moreover, when a dark-adapted leaf is illuminated, changes in chlorophyll a fluorescence intensity, known as the Kautsky effect, are observed (GOVINDJEE, 1995). The Kautsky transient shows a fast rise completed in less than 1 s, with a subsequent slower decline towards a steady state. It is well-documented that the rise phase of the transient reflects the primary reactions of photosynthesis (KRAUSE; WEIS, 1991). Transient fluorescence has provided additional and more accurate information (STRASSER; GOVINDJEE 1992; STRASSER; SRIVASTAVA; GOVINDJEE, 1995). For example, it was demonstrated that the fluorescence rise kinetics of the Kautsky transient is polyphasic when plotted on a logarithmic time scale, clearly exhibiting the steps J and I (STRASSER; GOVINDJEE, 1992) between the initial O (F_o) and maximum P level (F_m). Strasser; Srivatava and Tsimilli-Michael (2000) have developed an analysis of the OJIP fast fluorescence rise, measured from 50 μs to 1 s upon irradiation of the photosynthetic sample, and have also described the linkage between this physical signal and the biological functions. Thus, analysis of fluorescence induction (the 'JIP-test') made it possible to analyse the changes in energy transfer within PS2 and the relationships between primary photochemistry and the requirement for electrons in further stages of photosynthetic metabolism with many details (STRASSER; SRIVATAVA; TSIMILLI-MICHAEL, 2000).

The aim of this study was to evaluate the physiological behavior of *R. mangle* and *L. racemosa* growing in the basin forests at Vitória Bay, ES (Brazil) to variations of salinity. For this purpose, we performed the measurements of chlorophyll fluorescence with measurements of salinity three hours before high tide (pre high tide - H_1), during high tide (H_2) and three hour after high tide (post high tide - H_3) in separate two seasons (dry and rainy seasons) simulating the natural variations of tidal level.

Material and Methods

Plant material and experimental conditions - The study site is located in Vitória Bay (20°15 '48"S and 40°18'17"W), Espírito Santo State, southeast Brazil. It comprises basin forest with associated salt flats, where evaporation exceeds precipitation during several months of the year. Tidal amplitudes at the study site ranged between 0.3 and > 1 m. The relative air humidity was approximately 89 %. The annual average air temperature and annual precipitation are showed in figure 1.

The area study is characterized by a dry season

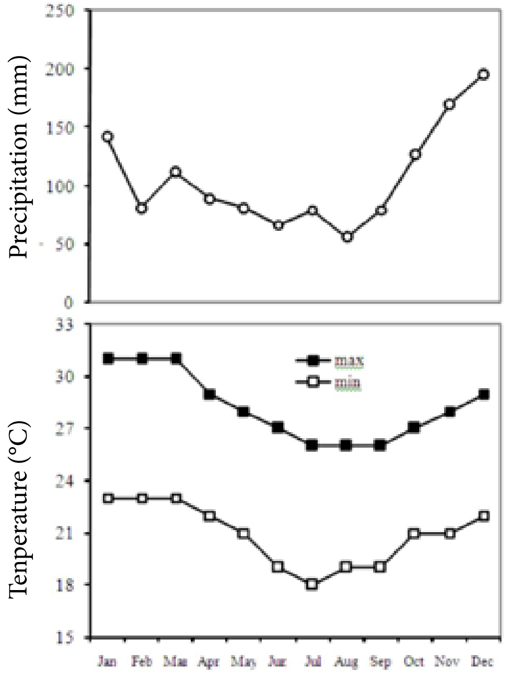


Figure 1. Precipitation and minimal and maximal temperatures of the study site (Vitória Bay) along of year. The dry and rainy season range from April to September and from October to March, respectively.

(April to September) and a rainy season (October to March). Mangrove forest at the study site is composed of three tree species: *Rhizophora mangle* L. (Rhizophoraceae), *Laguncularia racemosa* (L.) Gaerth. (Combretaceae) and *Avicennia germinans* (L.) Stearn. (Acanthaceae). *R. mangle* and *L. racemosa* are the dominant trees at the study site, while *A. germinans* is little represented. All data were collected from ten trees, which varied in height from 2-4 m, three hours before high tidal (pre high tide - H_1), during the high tide (H_2) and three hours after high tide (post high tide - H_3) in separate two seasons (dry and rainy seasons). Chlorophyll fluorescence parameters were determined on fully expanded young leaves of *R. mangle* and *L. racemosa*.

Salinity (psu) - Salinity of interstitial water was accomplished immediately after collecting the water in the tubes, using a portable condutometer *SCHOT Mod. LF1*. Interstitial water was collected using 2.5 cm diameter PVC tube inserted at -0.2 m sediment depth, according to the method of Miller and Ullman (2004).

Chl *a* fluorescence - Chl *a* fluorescence transient measurements were conducted on mature dark-adapted and attached leaves at ambient temperature using a portable fluorimeter (Plant Efficiency Analyser built by Hansatech Instruments Ltf. King's Lynn, Norfolk, UK), with high time resolution (10 μs). The leaves were dark adapted for at least 30 min before the measurements were started. The measurement consisted of a single strong 1 s light pulse (with excitation intensity sufficient to ensure closure of all PS2 reaction center provided by an array of six light-emitting diodes peak 650 nm. The Chl *a* fluorescence emission induced by the strong light pulses was measured and digitized between 10 μs and 1 s by the instrument (OUKARROUM; SCHANSKER; STRASSER, 2009).

All leaves exhibit a polyphasic Chl *a* fluorescence rise during the first second of illumination after dark adaptation. The most marked step at 2 ms is called the J-step. The fluorescence rise up to the J-step provides information about single turnover events of the primary reactions of photochemistry, mainly QA reduction. During the time interval from 2 to ~200 ms multiple charge separations occur and the redox components of the electron transport chain become reduced. The different phases of this process show up in the fluorescence rise as the steps J, I and P. The step with the highest fluorescence intensity is called P (peak). In the single turnover range Fo (measured at 50 μs) to F_J (measured at 2 ms), the bands L (at about 100-200 ms) and K (at about 200-400 μs) can often be visualized by subtraction or by calculation of differences between fluorescence transients (STRASSER; SRIVASTAVA; TSIMILLI-MICHAEL, 2004).

From the fast OJIP transients, several bio-energetic parameters were derived according to the equations of the JIP-test using the program BIOLYSER (R. M. Rodriguez, Bioenergetic Laboratory, University of Geneva). The concept of the JIP-test is based on the Energy Flux Theory in Bio-membranes. The JIP-test defines the maximal energy fluxes in the energy cascade for the events Absorption (ABS), Trapping (TR_o), Electron Transport (ET_o) and Dissipation (DI_o) and formulates their link with selected fluorescence experimental signals (Ft) between Fo and Fm (CHRISTEN *et al*, 2007). Using minimal and maximal fluorescence leads to the well accepted expression for the maximum quantum yield of primary photochemistry as $TR_o/ABS = \phi P_o = 1-(F_o/F_m) = F_v/F_m$. The use of fast recording instruments with a time resolution in the microsecond range allows us to measure accurately the slope at the origin of the fluorescence OJIP rise dF/dt0. This allows the calculation of the rate constant for primary photochemistry at the given light conditions as the specific trapping flux [$TR_o/RC = M_o/V_j = (ABS/RC)\phi P_o$]. The combination of the expressions above leads to the average antenna size [$ABS/RC = (M_o/V_j)/(1- Fo/Fm)$]. Furthermore, the energy dissipation at the antenna chlorophylls level (DIo/RC) should be calculated as $DI_o/RC = (ABS/RC) - (TR_o/RC)$ (STRASSER and TSIMILLI-MICHAEL, 2001; STRASSER; SRIVASTAVA; TSIMILLI-MICHAEL, 2004; CHRISTEN *et al*, 2007). In this study, we used the following parameters obtained from data extracted from the OJIP chlorophyll a fluorescence transient: (i) the specific energy fluxes for absorption per reaction centre (ABS/RC), (ii) excitation energy trapping (TR_o/RC), and (iii) energy dissipation at the level of the antenna chlorophylls (DI_o/RC).

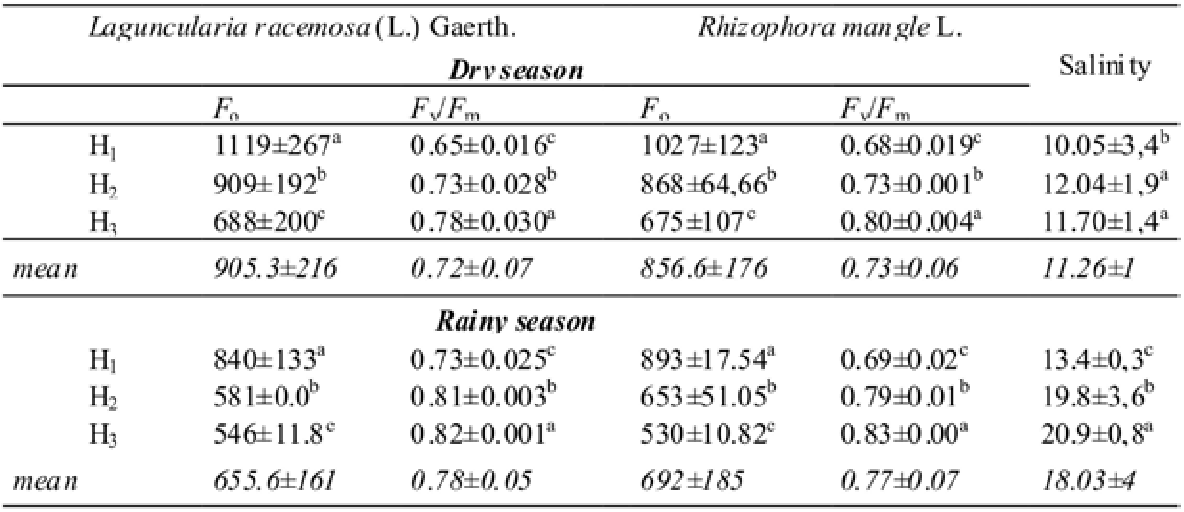
Statistical analysis - Each data collection consisted of ten repetitions, in a randomized block design. Results were examined by one-way analysis of variance (ANOVA) and differences between means were assessed using Tukey test ($p \leq 0.05$).

Results

We first investigated the changes in PS2 photochemistry in the dark-adapted leaves of *R. mangle* and *L. racemosa* under natural variations of diurnal tide during the dry and rainy seasons. Table 1 show that Fo decreased with increasing of salinity. During the rainy season, the decrease in Fo was strongly consistent with the increase in salinity from pre to post high tide, whereas a weaker relationship was observed for dry season which the salinity variation had been slight. In contrast, the potential quantum yield of PS2 (F_v/F_m) increased from pre to post high tide, following the increase of salinity. Under natural conditions, F_v/F_m values were greatest at H_2 (during high tide) and H_3 (post high tide) in both species and seasons (table 1).

ABS/RC and DI_o/RC values declined significantly ($p \leq 0.05$) from H_1 to H_3 in both species and seasons without any difference in TR_o/RC (figure 2). The comparison between two species revealed that ABS/RC and TR_o/RC were significantly reduced in *L. racemosa* mainly at H_2 and H_3 in both seasons (figure 2). Differences in DI_o/RC between species were observed during H_1 in both seasons and H_2 during rainy season (figure 2). However, during H_1 from dry season, the DI_o/RC values were higher in *L. racemosa*.

Table 1. Interstitial water salinity (psu), minimal fluorescence (Fo) and potential quantum yield of PSII (Fv/Fm), in leaves of Laguncularia racemosa (L.) Gaerth and Rhizophora mangle L. mangroves growing in the basin forests at Vitória Bay, Brazil, subjected to different salinity levels under field conditions. Data were collected three times along of day: Three hours before high tide (H1), during high tide (H2) and three hours after high tide (H3) during the dry and rainy seasons. Data (mean ± standard deviation) with different letters indicate a significant difference at p ≤ 0.05 between H1, H2 and H3, Tukey's test (n = 10)



	Laguncularia racemosa (L.) Gaerth.		*Rhizophora mangle* L.		Salinity
		Dry season			
	F_o	F_v/F_m	F_o	F_v/F_m	
H_1	1119 ± 267^a	0.65 ± 0.016^c	1027 ± 123^a	0.68 ± 0.019^c	$10.05 \pm 3{,}4^b$
H_2	909 ± 192^b	0.73 ± 0.028^b	$868 \pm 64{,}66^b$	0.73 ± 0.001^b	$12.04 \pm 1{,}9^a$
H_3	688 ± 200^c	0.78 ± 0.030^a	675 ± 107^c	0.80 ± 0.004^a	$11.70 \pm 1{,}4^a$
mean	*905.3±216*	*0.72±0.07*	*856.6±176*	*0.73±0.06*	*11.26±1*
		Rainy season			
H_1	840 ± 133^a	0.73 ± 0.025^c	893 ± 17.54^a	0.69 ± 0.02^c	$13.4 \pm 0{,}3^c$
H_2	581 ± 0.0^b	0.81 ± 0.003^b	653 ± 51.05^b	0.79 ± 0.01^b	$19.8 \pm 3{,}6^b$
H_3	546 ± 11.8^c	0.82 ± 0.001^a	530 ± 10.82^c	0.83 ± 0.00^a	$20.9 \pm 0{,}8^a$
mean	*655.6±161*	*0.78±0.05*	*692±185*	*0.77±0.07*	*18.03±4*

Discussion

Our results exhibited decreased Fo with increasing salinity from pre to post high tide and the lowest values of Fo occurred at the salinity levels of 11.7 psu during the dry season and 20.9 psu during the rainy season for *R. mangle* and *L. racemosa*. The sources of Fo include Chl *a* molecules in PS2 and attached light harvesting complex. Decreases in Fo in response to salinity can be related to adjustments at the antenna pigment level or decreases of the excitation trapping efficiency at the active centre of PS2 (YAMANE *et al*, 1997). Salinity tolerance of and *L. racemosa* mangroves, therefore, seemed to be associated with low levels of Fo at H_2 and H_3 (during and post high tide, respectively). No difference in F_o was observed between *R. mangle* and *L. racemosa* mangroves trees growing in basin forest along of ES coastline within of the same salinity level.

Previous studies indicate that Fo should be used as an indicator of stress when associated with potential quantum yield of PS2 (F_v/F_m) measurements (STIRBET; GOVINDJEE, 2011). In the current investigation, potential quantum yield of PS2 incre-

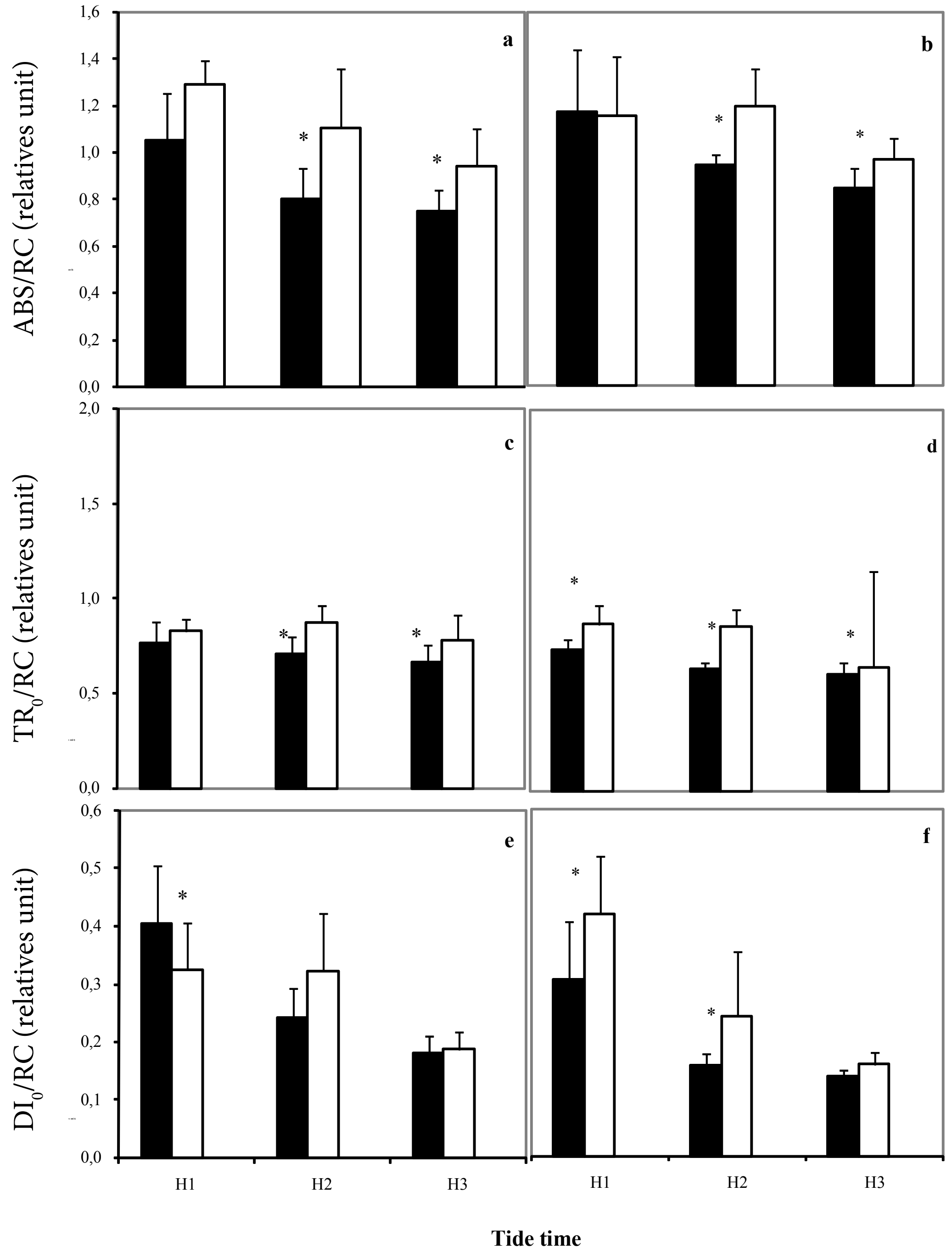


Figure 2. Specific energy fluxes for absorption per reaction centre (ABS/RC – a and b), excitation energy trapping (TRo/RC – c and d) and energy dissipation at the level of the antenna chlorophylls (DIo/RC – e and f) values obtained of (▪) *Laguncularia racemosa* (L.) Gaerth. and (□) *Rhizophora mangle* L. growing in the basin forests at Vitória Bay, Brazil, subjected to different salinity levels under field conditions. Data were collected three times along of day: Three hours before high tide (H_1), during high tide (H_2) and three hours after high tide (H_3) during the dry (a, c and e) and rainy (b, d and f) seasons. Significant differences in JIP-test parameters between species in each time are represented by an asterisk (*) (Tukey test at 5% probability level). (n = 10)

ased with salinity (Table 1). During pre high tide (H_1), potential quantum yield of PS2 values in *L. racemosa* were 0.65 and 0.73 during the dry and rainy season, respectively, representing an increase of 12.3 % in potential quantum yield of PS2 with an increase in salinity around 29.8 %. In contrast, the increase in Fv/Fm values of 1.4 % found in *R. mangle* has been shown poorly significant when compared to that found in *L. racemosa*. This result enforces evidences that the photosynthetic apparatus of *L. racemosa* has higher plasticity in high salt conditions, as evidenced by higher gain in Fv/Fm in H_1, which might reflect an enhanced salt secretion from glands characteristic in this mangrove. In addition, similar results were observed during high tide (H_2). At H_2, with salinity increased to 64.45 % from dry to rainy season, there was an increase in Fv/Fm values of approximately 10.95 and 8.2 % in *L. racemosa* and *R. mangle*, respectively, confirming that increases in salinity resulting from natural variation of diurnal tide changed the dynamic of light absorption reactions with highest intensity in *L. racemosa* mangrove.

However, during post high tide (H_3), salinity increased 78.6 % from dry to rainy season. In these conditions, Fv/Fm slightly increased with salinity in both *L. racemosa* and *R. mangle* (5.1 and 3.75 %, respectively). If one takes into account the absolute values, this increase represents a potential quantum yield of PS2 increased from 0.78 to 0.82 and from 0.80 to 0.83 in *L. racemosa* and *R. mangle*, respectively, from dry to rainy season. The range of Fv/Fm for various monocotyledon and dicotyledonous families with different life styles has been reported to be from 0.75 to 0.85 (Hunt 2003). The high values of Fv/Fm (0.75–0.83 across salinity treatments) indicate that *R. mangle* and *L. racemosa* were able to maintain maximum photochemical efficiency of PS2 at high salinity levels, and demonstrate a very high degree of salinity tolerance relative to other plant species.

Some mangroves species show decreases in Fv/Fm under high salinities that involve protective mechanism (i.e., changes in the pool size of the xanthophyll), which could be reversed upon transfer of plants to a stress-free environment or with changes in the environmental conditions (TAKEMURA, *et al*, 2000; LI *et al*, 2008). Therefore, the responses under salt stress conditions of *R. mangle* and *L. racemosa* mangrove needs to be further verified in controlled conditions to explain increases in Fv/Fm value. Also, factors associated with decrease in Fo (oxi-reducing conditions of the plastoquinone pool, chlororespiration or others) might need to be further elucidated. Thus, the physiological mechanisms associated with the decrease of fluorescence yield and with the increase of photosynthetic performance (evaluated such as Fv/Fm ratio) in mangroves under high salt conditions are not clear. It is possible that increased photorespiration could have contributed to photochemical dissipation of absorbed light energy in this study. In addition, under salt conditions, mangrove photosynthesis is primarily limited by deficiencies in mineral ions such as K+. Thus, salt tolerance in mangroves species is correlated not only with adaptations to excess salt substrate but also to some of the secondary effects of salinity such as water deficit and impaired nutrient acquisition. High concentrations of inorganic ions in mangrove tissues are required to contribute to turgor formation (NAIDOO; TUFFERS; VON WILLERT, 2002). Studies on inorganic ions absorption and accumulation in mangroves plants would contribute to understand salt tolerance in mangroves species growing in Vitória Bay.

Many studies have only used the ratio of variable to maximal chlorophyll a fluorescence (Fv/Fm) as parameter to screen for salt tolerance in mangroves. However, a growing body of evidence indicates that the Fv/Fm ratio is often insensitive to salt and not always a suitable parameter for indicating salt tolerance of plants (VAN HEERDEN *et al*, 2003; VAN HEERDEN; STRASSER; KRÜGER, 2004). Force, Critchley and Rensen (2003) demonstrated the advantage of using a number of JIP-test derived fluorescence parameters to evaluate PS2 function, rather than using only a single parameter such as the Fv/Fm ratio. Like previously found by Strauss *et al.* (2006), the JIP-test parameters are most sensitive in assessment of plant stress. In this study, we evaluated the specific energy fluxes for absorption per reaction centre (ABS/RC), excitation energy trapping (TRo/RC) and energy dissipation at the level of the antenna chlorophylls (DIo/RC) (figure 2), which would be sensitive enough to evaluate the responses in the mangrove species to ultimately rank them according to salt tolerance.

Diurnal variations of salinity conditions adversely affected the energy fluxes for absorption per reaction centre (ABS/RC) of both *L. racemosa* and *R. mangle* mangroves growing in the basin mangrove forest of Vitória Bay (figure 2A-B). At H_2, where the salinity increased by 39 %, ABS/RC was significantly reduced in *L. racemosa* compared to *R. mangle*. These results are consistent to that increase of Fv/Fm values described above. According Mehta *et al.* (2010) ABS/RC, which reflects the effective antenna size of an active reaction centers, is calculated as a total number of photons absorbed by Chl molecules of all RCs divided by total number of active RCs. Thus, ABS/RC values are influenced by ratio of active/inactive RCs. At H_2 and H_3, the lower values of ABS/RC in *L. racemosa* resulted in higher decrease in antenna size of active RCs in this specie. Taken together, it would appear that *L. racemosa* is physiologically more sensitive than *R. mangle* in it salt responses in field conditions. Reports in the literature (SOBRADO, 1999; PANDA *et al*, 2006) confirm that high salinity conditions adversely affect photosynthetic performance of mangrove under field conditions.

The low ABS/RC values also were followed by reduced values ($p \leq 0.05$) of excitation energy trapping $[TR_o/RC = = M_o/V_j = (ABS/RC)\phi P_o]$ in *L. racemosa* (figure 2C-D). Reductions in energy dissipation at antenna chlorophyll level (DI_o/RC) (figure 2E-F) such as observed in *L. racemosa* and *R. mangle* at H_1, H_2 and H_3, in both seasons, would explain the decline observed in Fo values (table 1). The declined values of DIo/RC from H_1 to H_3 are in line with the increase in potential quantum yield of PS2 in both *L. racemosa* e *R. mangle* mangrove species.

Conclusions

Variations in salinity resultant of natural variations of diurnal tides influenced the photosynthetic responses of both *L. racemosa* and *R. mangle* mangroves plants during dry and rainy seasons. Likewise, *R. mangle* and *L. racemosa* trees growing in basin mangrove forest along of ES coastline showed differences in minimal fluorescence emission in relation to natural changes in salinity. In contrast, the slight variation of Fv/Fm increment in *L. racemosa* (from 12.3 % in H_1 to 10.95 % in H_2) would indicate higher salt tolerance in this specie in response to changes in the diurnal tide. In addition, analyses of energy flux through PS2 permitted observe the lower specific energy fluxes for absorption per reaction center (ABS/RC), which reflected in lower excitation energy trapping (TRo/RC) and lower energy dissipation at antenna chlorophylls levels (DIo/RC) in *L. racemosa*. These results reflect genetic differences related to acquisition of morphological and physiological adaptations that permit survival in saline environment such as mangrove ecosystem of Vitória Bay.

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