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SHORT COMMUNICATION

CHLOROPHYLL FLUORESCENCE TRANSIENTS WITH RESPONSE TO LEAF WATER STATUS IN COCONUT

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Leaf water potential (Ψ_{leaf}) and chlorophyll fluorescence (Chl. F) transients were determined in coconut seedlings in the field under intermittent irrigated (non-stress) and un-irrigated (stress) conditions. Stress was imposed by withholding irrigation to the seedlings. With reduction in Ψ_{leaf}, associated with the increase in soil water deficit as well as micrometeorological variables, the Chl. F traits showed variation. Re-irrigation of the seedlings, although showed recovery of the leaf water status, did not show recovery to the PS II system as revealed by Chl. F values. The reduction in various fluorescence derived ratios viz, Fv/Fm and Fm/Fo as well as t½ (the half rise time from Fo to Fm) implied the blockage of electron flow on the waterside of the PS II and damage to the photochemical apparatus with stress. Observations during different treatment periods clearly revealed that in coconut PS II is highly sensitive to agro-meteorological variables and prolonged drought lead to permanent damage to photosynthetic apparatus. Hence, maintenance of water status of both soil and leaf is highly crucial to protect the PS II system in coconut seedlings during summer months.

Key words: **Chlorophyll fluorescence, leaf water potential, perennial tree, PS II, stress.**

Coconut (*Cocos nucifera* L.), a perennial plantation tree crop is grown mainly under the rain fed conditions of the tropics. Impact of water stress during the summer months (March-May) on the physiology and biochemistry of this crop has been well established (Rajgopal *et al.* 1989, Chempakam *et al.* 1993, Rajagopal and Kasturi Bai 2002). How ever impact of both water and atmospheric stress on the photochemical efficiency of this crop is not understood. Chlorophyll fluorescence of photosynthetic systems which provide information on the cause and mechanism of PS II functioning under stress has been extensively used in various fields of plant physiological studies (Krause and Weis 1991, Govindjee 1995, Srivastava *et al.* 1995). This paper reports the changes in the photochemical efficiency of PS II with the reduction in leaf water potential in coconut seedlings in the field condition. The observations will provide an insight into the physiological status of coconut seedlings under water stress in the field condition.

The observations on leaf water potential (ψ_{leaf}) and chlorophyll fluorescence (Chl. F) transients were carried out in 18 month old coconut seedlings (*Cocos nucifera* L.) growing in the Institute farm (12°30' N, 70°00' E and 11m MSL) in red sandy loam sol in completely randomized block design. Irrigation was provided to the seedlings continuously till saturation of the soil (non stress) and was withheld for twenty days (stress I). The seedlings were re-irrigated continuously for 15 days till saturation of the soil (recovery) and were again stressed by withholding irrigation for 20 days (stress II). The experiment was spread over during the summer months

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of March to April. ψ_{leaf} as well Chl. F transients were measured during all the four periods, i.e. non-stress, stress I, recovery and stress II, in fifteen randomly selected seedlings. Four measurements in the third leaf from the spindle were made from each seedling between 09.30 hr and 10.30 hr.

Chl. F transients were measured in dark-adapted leaf ot coconut seedlings with Morgan, CF-1000, Chlorophyll fluorescence meter, USA and a black light weight clamp for dark adaptation (Rabha and Uprety 1998). Leaflets were dark adapted for 30 minutes before measurements. The Chl. F transients over five seconds were measured after an active pulse of 680 μmol $m^{-2}s^{-1}$. The in built microprocessor of the instrument calculates all the main fast Chl. F parameters such as minimal fluorescence (Fo), maximum fluorescence (Fm) after a flash of actinic radiation and the variable fluorescence (Fv) (i.e. Fm-Fo), photochemical efficiency, (Fv/Fm) and t½ (i.e. the half rise time from Fo to Fm). In addition to the above parameters, there is some quantification of the slow fluorescence transients such as Ft (terminal fluorescence) and Fq (fluorescence quenching, i.e Fm-Ft). Various fluorescence-derived ratios such as fluorescence decrease ratio, (Fm-Ft)/Ft (Jenkins *et al.* 1981), ratio of maximum to ground fluorescence, Fv/Fo (Govindjee *et al.* 1981) and the ratio of variable to ground flourescence, Fv/Fo (Babani and Lichtenthaler 1996) were calculated from these measurements. ψ_{leaf} was measured with Scholanders' Pressure Chamber as modified for coconut (Rajagopal *et al.* 1987). Soil water deficit as well as agro meteorological variables were recorded as described earlier by Rajagopal *et al.* (1989). The mean for the 15 seedlings were subjected to analysis of variance to test for significant differences between the treatments (Panse and Sukhatme 1967). Correlation between ψ_{leaf} and Chl. F traits were worked out to understand the relation between the two as per standard method.

During the experimental period (March to April) radiation ranged between 273.6 and 351 Wm^{-2}, air temperature between 33°C and 37°C and VPD between 3 and 3.6 K Pa. The soil moisture content, wich was 9% during non-stress period, was reduced to 6.7% during Stress I (with a soil water deficit of 69 mm). After re-watering the soil moisture reached to the original non-stress level, which got reduced to 4.2% (with a soil water deficit of 102 mm) after withholding irrigation during Stress II. This reduction (58%) in soil moisture during Stress II can be attributed to the higher evaporation demand prevailed during the period with high pan evaporation rate (5.5 mm day^{-1}).

Under full irrigation during non stress period ψ_{leaf} ranged between -8.1 and -5.6 bars with a mean of -7.23 bars (CD: 0.974, P=0.05) and Fv/Fm ranged between 0.803 and 0.849 with a mean of 0.82 (CD: 0.02, P=0.05), indicating the seedlings were healthy and not stressed. Imposition of stress resulted in reduction in ψ_{leaf} with associated changes in the Chl F values. Critical analysis of the data (Table 1) during each treatment period clearly indicated the impact of ψ_{leaf} changes on the Chl. F traits. During the non-stress period with high ψ_{leaf} of -7.23 bars all the values of Chl. F traits, viz. Fv/Fm, Fm/Fo, Fv/Fo etc. were within the range. However, with a reduction in ψ_{leaf} by 27% (Stress I), there was a reduction in Fv/Fm (11%), Fm/Fo (25%) Fv/Fo (33%) and t½ (17%) and increase in Fm-ft (38%) and (Fm-ft)/Ft (33%). When the seedlings were re-irrigaed although ψ_{leaf} values showed a recovery to the original level, Chl. F values did not reach to the non stress level indicating damage to the photosynthetic apparatus. Withdrawing irrigation further to a period of 20 days caused higher reduction in ψ_{leaf} (110%) with concomitant changes in the values of Chl. F parameters with higher reduction in Fv/Fm (33%), Fm/Fo (44%), Fv/Fo (57%) and t½ (15%) with an increase in (Fm-Ft)/Ft (37%) thus showing irreparable damage to the PS II system. The results clearly revealed that except for higher reduction in the values during stress II, the trend remained the same during stress I and stress II.

There are reports to show that coconut palms show stress symptoms when exposed to irradiation above 265 Wm^{-2}, temperature >34°C and VPD>2.6 K Pa (Kasturi Bai *et al.* 1988) and soil water deficit > 110 mm (Rajagopal *et al.* 1989). From the results (Table 1) it is clear that seedlings behaved differently during different soil moisture levels. Fv/Fm which has been used to quantify PS II efficiency (Kraus and Weis 1991) showed a strong linear relationship with ψ_{leaf} (Fig. 1). Fm/Fo that serves as a quantitative indicator of the ψ_{leaf} (Govindjee

Table 1. Leaf water potential (ψ_{leaf},-bars) and chlorophyll fluorescence transients (relative fluorescence units) during different treatment periods in coconut seedlings (mean for 15 seedlings).

Parameter	Non stress	Stress I	Recovery	Stress II	CD : P=0.05
ψ_{leaf}(-bars)	7.23	9.17	6.93	14.53	1.90
Fm/Fo	5.62	4.22	4.54	2.52	0.590
Fv/Fo	4.62	3.12	3.45	1.49	0.561
Fv/Fm	0.820	0.733	0.770	0.522	0.074
Ft	0.794	0.779	0.667	0.487	0.117
Fm/Ft	0.314	0.434	0.446	0.400	0.063
Fm/Ft/Ft	0.425	0.574	0.707	0.969	0.208
t½ (ms)	141.87	118.4	133.3	113.3	13.81

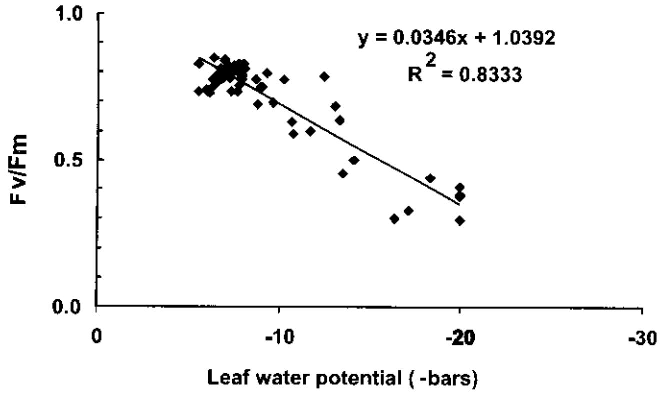

Fig.1. Relationship between leaf water potential and Fv/Fm in coconut seedlings (Each point is mean for four observations)

et al. 1981) showed a fall during stress indicating blockage of the electron flow on the waterside of PS II. A strong linear relationship (R^2=0.69) has been observed between ψ_{leaf} and Fm/Fo. Similarly, Fv/Fo which provides an assessment of the potential photosynthetic activity of healthy as well as stressed plants (Babani and Lichtenthaler 1996) showed linear relationship with ψ_{leaf} (R^2=0.663). The higher fall in Fv/Fo (68%) as well as Fm/Fo (55%) during very severe stress condition as compared to non stress condition can be attributed to the higher increase in Fo (110%) during the period. This coupled with the decrease in Fv/Fm (37%) and Ft (39%) indicated higher inactivation of PS II reaction centers (Genty *et al.* 1989). High correlation has been noticed between Fv/Fm, Fm/Fo and Fv/Fo (Table 2).

The quenching of fluorescence (Fm-Ft) showed an increase during stress I (38%) and as compared to non-stress period an increase of 27% has been observed in this under severe stress period The increase in (Fm-Ft) during stress period has been related to the disruption of oxido-reduction reactions of the primary electron acceptor and on the plastoquinon pool size (Santos *et al.* 1998). The fluorescence decrease ratio (Fm-Ft)/Ft showed significant differences between the treatments. As reported in oak (Epron and Dreyer 1990), the increase in (Fm-Ft)/Ft during stress period can be attributed to the disruption in the flux of electron through PS II.

The value t½ has been used to estimate the antenna size of PS II (Krause and Weis 1991) and is related to the rate of photochemical reaction. The reduction in t½ by 20% during severe stress in coconut seedlings can be attributed to the lowering of the relative number of active PS II centers. A positive correlation observed between t½ and Fv/Fm during severe stress period supports this (Table 2). The higher decrease in Fv/Fm due to stress can be attributed to the higher increase in Fo (110%) and decrease in Fm (20%). As per Krause and Weis (1991) higher Fo value denotes thermal damage to PS II and lower Fv denotes stress damage to the thylakoid. The Fm values are also reported to be proportional to the quantity of chlorophyll molecules in leaf tissue (Miranda *et al.* 1981) and the decrease in Fm values during stress has been attributed to the disruption

Table 2. Correlation between leaf water potential (ψ_{leaf}) and chlorophyll fluorescence transients in coconut seedlings.

	ψ_{leaf}	Fm/Fo	Fv/Fo	Fv/Fm	Ft	Fm-Ft	(Fm-Ft)/Ft
Fm/Fo	0.834**	–	–	–	–	–	–
Fv/Fo	0.814**	0.986**	–	–	–	–	–
Fv/Fm	0.912**	0.926**	0.913**	–	–	–	–
Ft	0.360**	0.462**	0.475**	0.404**	–	–	–
Fm-Ft	0.139	–0.176	–0.247	0.019	–0.170	–	–
Fm/Ft/Ft	–0.237	–0.422**	–0.461**	–0.320*	–0.838**	0.515**	–
t½	0.504**	0.516**	0.532**	0.506**	0.203	–0.222	0.037

*&** indicate significance at 5% and 1% level respectively.

in the phosphorylation of proteins in the thylakoid membranes (Horton and Black 1983).

The observations clearly revealed that in coconut PS II is highly sensitive to soil water deficit and micrometeorological variables. Hence, proper maintenance of soil and leaf water status is highly crucial for protecting the photosynthetic apparatus and survival of coconut seedlings during summer months.

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