

# Net CO<sub>2</sub> Assimilation and Chlorophyll Fluorescence in Cocoa Trees\*

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The response of photosynthesis to increase or decrease in photon flux density (PFD) in leaves of cocoa canopy was investigated. The time-course of this response was similar to decrease or increase, but the values were lower in leaf under extended period of low irradiance as compared to sunlit leaf. The fast photosynthetic induction was accompanied by similar changes in transpiration rate. In contrast, stomatal conductance did not show significant variations. Chlorophyll fluorescence values showed inverse relationships as compared to net CO<sub>2</sub> assimilation. The results indicate that there was a fast phase induction of CO<sub>2</sub> assimilation in relation to lightflecks of cocoa leaf without any limitation being imposed by stomatal conductance. The leaves from the most recently hardened flush showed higher photosynthetic efficiency as compared to other flushes.

**Key words:** Photosynthesis, chlorophyll fluorescence, transpiration, irradiance, cocoa.

## INTRODUCTION

The light environment within crop canopies is characterised by large temporal and spatial variability (Ross 1980). Understorey plants receive low level of PFD interspersed with intense lightflecks (Percy 1990). In such situations, the photosynthetic rates often do not attain steady state. Carbon assimilation rate has been measured mostly under steady state conditions. Non-steady photosynthetic induction with increase in PFD in crop canopies have been recently reported (Percy and Seeman 1990, Jackson *et al.* 1991). The rate of CO<sub>2</sub> assimilation is closely related to changes in chlorophyll fluorescence emission (Walker *et al.* 1983, Ireland *et al.* 1984).

In southern India, cocoa is cultivated as a mixed crop under tall arecanut palms where the environmental conditions are favourable. Approximately

30% PFD is transmitted through arecanut canopy which can be utilized by cocoa (Balasimha 1989). The P<sub>N</sub> of cocoa as influenced by light, temperature and VPD under steady-state conditions in field have been reported (Balasimha *et al.* 1991). In this paper, net CO<sub>2</sub> assimilation and chlorophyll fluorescence in cocoa leaves at different irradiance levels and flush position are described.

## MATERIALS AND METHODS

Cocoa (*Theobroma cacao* Linn.) trees cultivated at a spacing of 2.7 × 5.4 m under the shade of 18-year old arecanut palms (*Areca catechu* Linn.) at 2.7 × 2.7 m distance were used in the experiments. Both the crops were fertilized annually with 100 g N, 40 g P<sub>2</sub>O<sub>5</sub> and 140 g K<sub>2</sub>O per tree and were irrigated at 10 day intervals during dry months.

Measurements of PFD, T<sub>leaf</sub>, VPD, P<sub>N</sub>, g<sub>s</sub>, E and C<sub>i</sub> were made from December to May on mature leaves using a LI-6200 portable photosynthesis system (Li-Cor Inc. Lincoln, NE, USA) with 1 leaf chamber. Two sets of experiments were made in leaves at high and low (shade) irradiance in the field. In the first set, leaves exposed to high irradiance were transferred to low-irradiance for a duration and brought back to original high irradiance. In another set, shaded leaves in low irradiance were

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**Abbreviations:** C<sub>i</sub> - intercellular CO<sub>2</sub>, E - transpiration rate, g<sub>s</sub> - stomatal conductance, PFD - photon flux density, P<sub>N</sub> - net photosynthesis, T<sub>leaf</sub> - leaf temperature, VPD - vapour pressure deficit, F<sub>o</sub> - minimal continuous fluorescence, F<sub>m</sub> - maximal fluorescence level, F<sub>v</sub> - variable fluorescence.

transferred to high irradiance by parting the canopy for a period and brought back to original low irradiance. These experiments were done on intact leaves by shifting them from sunfleck to shaded areas and vice versa. The measurements were continued with the Li-6200 during these changes of leaf position. Two leaves/tree in each set and on 15 trees were measured. The LI 6200 was programmed to take 10 measurements at 5s intervals. The PFD ranged from 85 to 1376  $\mu\text{mol m}^{-2}\text{s}^{-1}$  depending on leaf position.

Chlorophyll fluorescence was measured using the Plant Efficiency Analyser (Hansatech Instruments Ltd., Norfolk, U.K.) in 4 trees. A leafclip was attached to the leaf and shutter closed for dark-adaptation for 30 min. The dark adapted leaf was fitted with the sensor unit over the clip so that it sealed out the light. Holding the clip and sensor unit together, the shutter plate was opened and fluorescence measurements taken at 80% light level of the LED. The stored data were transferred through a software package to a Computer-AT (Hindustan Computers Ltd.) for study. For experiments on leaf position 4 trees for  $P_N$  and 8 trees for fluorescence measurements were used.

## RESULTS AND DISCUSSION

Areca nut canopy transmits about 30% of light which is interspersed with lightflecks and shade areas (Balasimha 1989). Increasing or decreasing PFD in cocoa canopy resulted in a rapid response of  $P_N$  within 20s (Figs. 1, 2). Similar changes in  $E$  was also observed in both high irradiance adopted and shade leaves. Although, the timecourse for  $P_N$  in cocoa leaf under both sets of experiments (i.e., high and low-I) were similar when shifted to decreasing or increasing PFD, the values were higher in sunlit leaf (Fig. 1). The mean values of net  $\text{CO}_2$  assimilation and related parameters for sunlit and shaded leaves are given in Table 1. The chlorophyll fluorescence parameters were, however, higher in shade (low irradiance) leaves as compared to leaves at high irradiance (Table 2, Fig. 3). The reduced photochemical reaction and consequently lower  $P_N$  is thus inversely related to fluorescence characteristics as reported in other species (Krause and Weis 1991).

In contrast to  $P_N$  and  $E$ ,  $g_s$  did not show significant differences in response to varying PFD (Table 1 and

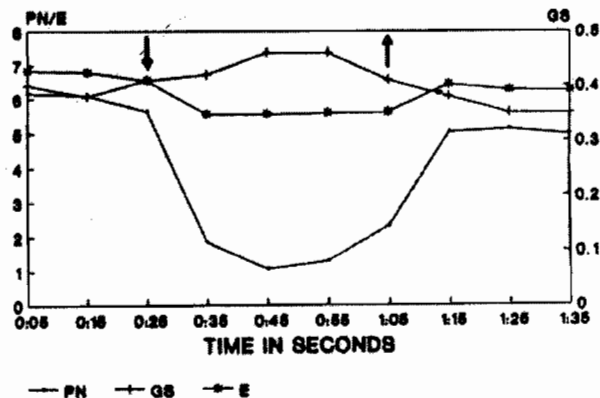


Fig. 1. Time courses of  $P_N$  ( $\mu\text{mol m}^{-2}\text{s}^{-1}$ ),  $g_s$  ( $\text{mol m}^{-2}\text{s}^{-1}$ ) and  $E$  ( $\text{mmol m}^{-2}\text{s}^{-1}$ ) in sun leaf (mean of 15 trees). The PFD levels were 1250-1380  $\mu\text{mol m}^{-2}\text{s}^{-1}$  in full sunlight and 100-150  $\mu\text{mol m}^{-2}\text{s}^{-1}$  in shade. Leaf temperatures were 33 to 36 C and VPD was 1.8 to 2.5 kPa. The PFD was decreased and increased at the arrows.

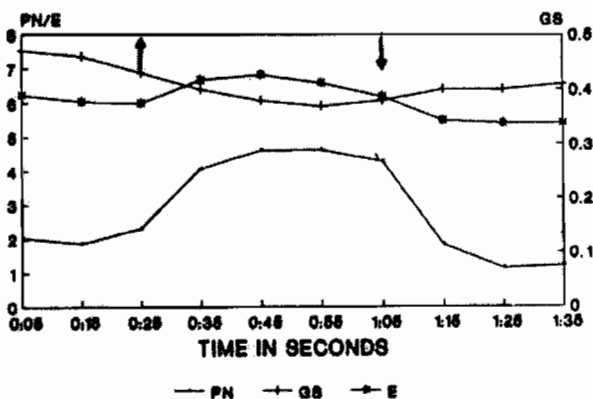


Fig. 2. Time courses of  $P_N$  ( $\mu\text{mol m}^{-2}\text{s}^{-1}$ ),  $g_s$  ( $\text{mol m}^{-2}\text{s}^{-1}$ ) and  $E$  ( $\text{mmol m}^{-2}\text{s}^{-1}$ ) in shade leaf (mean of 15 trees). The PFD levels were 1170-1330  $\mu\text{mol m}^{-2}\text{s}^{-1}$  in full sunlight and 75-100  $\mu\text{mol m}^{-2}\text{s}^{-1}$  in shade. Leaf temperatures were 33-35 C and VPD was 1.9 to 2.5 kPa. The PFD was increased and decreased at the arrows.

Figs. 1, 2). Light saturation for cocoa  $P_N$  is around 400  $\mu\text{mol m}^{-2}\text{s}^{-1}$  of PFD (Balasimha *et al.* 1991), and because of this the  $P_N$  values shown in Table 1 and 3 were similar. Earlier results on other crops had revealed that in a slow phase induction,  $g_s$  and ribulose 1, 5-bisphosphate carboxylase (RubisCo) activity were involved (Woodrow and Mott 1989, Percy 1990, Percy and Seemann 1990, Jackson *et al.* 1991). In cocoa leaf, since no limitation by  $g_s$  was observed, fast phase photosynthetic components may be operating. There is similarly fast photosynthetic induction which may be attributable

**Table 1.** Photosynthetic characteristics of cocoa leaves (mean of 15 trees)

Parameters	Irradiance level*		C.D. (p = 0.01** or 0.05)*
	High	Low	
P <sub>N</sub> ( $\mu\text{mol m}^{-2}\text{s}^{-1}$ )	6.14	1.95	0.47**
g <sub>s</sub> ( $\text{mol m}^{-2}\text{s}^{-1}$ )	0.39	0.46	0.05**
E ( $\text{mmol m}^{-2}\text{s}^{-1}$ )	6.82	6.14	0.56*
C <sub>i</sub> (ppm)	287	319	3.00**
<b>a. Microclimatic variables:</b>			
PFD ( $\mu\text{mol m}^{-2}\text{s}^{-1}$ )	1376	85	
Leaf temperature (°C)	35.0	33.2	
VPD (kPa)	2.40	1.96	

**Table 2.** Chlorophyll fluorescence (arbitrary units) in cocoa leaves (mean of 4 trees)

Parameter	Irradiance level		S.E.M.
	High	Low	
F <sub>o</sub>	725	574	40
F <sub>m</sub>	2886	3502	118
F <sub>v</sub>	2067	2927	142
F <sub>v</sub> /F <sub>m</sub>	0.716	0.835	0.019

**Table 3.** Net photosynthesis and related parameters in relation to leaf age (mean of 4 trees)

Flush	P <sub>N</sub> ( $\mu\text{mol m}^{-2}\text{s}^{-1}$ )	g <sub>s</sub> ( $\text{mol m}^{-2}\text{s}^{-1}$ )	E ( $\text{mmol m}^{-2}\text{s}^{-1}$ )	Internal CO <sub>2</sub> (ppm)
1	6.52	0.39	4.87	305
2	6.15	0.35	4.95	310
3	2.82	0.21	3.37	324
SEM	0.39	0.02	0.18	11

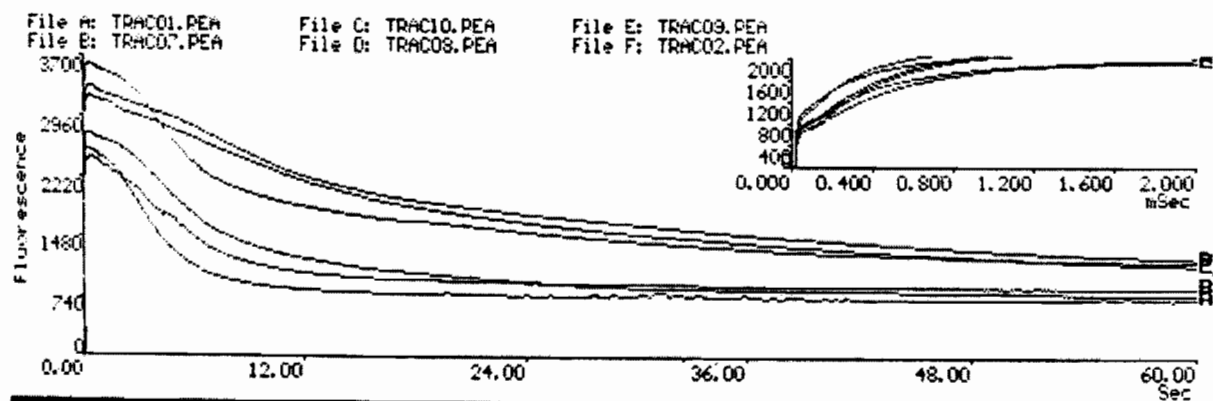
Mean microclimatic variables; PFD: 394±34  $\mu\text{mol m}^{-2}\text{s}^{-1}$

Tair: 29.75 ± 0.06°C

VPD: 1.50 ± 0.08 kPa

to RubisCo regeneration and/or Calvin-cycle metabolites (Kirschbaum and Pearcy 1988, Pearcy 1990). Further, as seen from Fig. 4, if the leaf has been in low irradiance for an extended time, increased PFD enhanced P<sub>N</sub> two-fold, but the values did not reach steady-state rates of sunlit-leaf.

Cocoa exhibits a rhythmic growth of leaves called flushing which occurs periodically. Photosynthetic characteristics were determined in three flush-positions of cocoa. Leaves from the most recently hardened flush (flush 1) exhibited higher net CO<sub>2</sub> assimilation and least rates in oldest (flush 3) leaf (Table 3). This low P<sub>N</sub> was accompanied by significant reduction in g<sub>s</sub> and E. There was an inverse trend with respect of fluorescence characteristics showing higher photosynthetic efficiency in flush 1 (Table 4). The results on P<sub>N</sub> and chlorophyll fluorescence indicate high adaptation of cocoa canopy to shade which is characteristic of an understory plant.

**Fig. 3** Time courses of fluorescence in sun leaves (files A,B,C) and shade leaves (Files D,E,F) of three representative trees. The inset depicts the initial fast data.

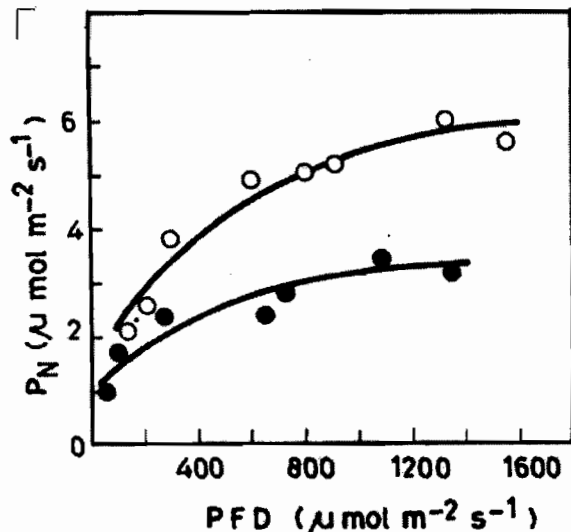


Fig. 4 Relation of  $P_N$  to PFD in sunlit ( $0, r^2 = 0.35$  at  $P = 0.01$ ) and shaded ( $\bullet, r^2 = 0.25$  at  $P = 0.01$ ) leaves of cocoa.

Table 4. Chlorophyll fluorescence (arbitrary units) in relation to leaf age (mean of 8 trees)

Flush	$F_o$	$F_m$	$F_v$	$F_v/F_m$
1	642	2574	1965	0.746
2	675	2693	2017	0.743
3	570	2817	2245	0.795
SEM	19	65	76	0.012

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