

DIURNAL VARIATION IN GAS EXCHANGE CHARACTERS OF LEAVES OF *HEVEA BRASILIENSIS* EXPOSED TO SUN LIGHT AT DIFFERENT CANOPY LEVELS AND THEIR CONTRIBUTION TO DRY MATTER PRODUCTION

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ABSTRACT

As in many other crops a significant percentage of leaves in the canopy of a rubber plantation are found under limiting light levels. The photosynthetic capacity of these shaded leaves may play an important role in determining the productivity of the rubber plants. The dry matter production capacity of shade leaves was studied using polybag plants of *Hevea brasiliensis* and grown under different light regimes. The CO₂ assimilation rates and related parameters were monitored through-out the day using a portable photosynthesis system.

The diurnal variation pattern of CO₂ assimilation rates and the internal leaf CO₂ concentration to ambient CO₂ concentration ratio (ci/ca) were different in plants grown under different light regimes. Though peak CO₂ assimilation rates are shown at around 0930h in all treatments, the assimilation rates tend to decline very early in plants grown under full light. During the early and latter parts of the day, plants grown under full light showed significantly higher CO₂ assimilation rates than in plants grown under 40% and 25% of incident light. However, stomatal conductance declined with time of the day and it was similar in all treatments. Low CO₂ assimilation rates later in the day could be either due to accumulation of abscisic acid or due to end product inhibition.

It is apparent that photosynthesis is only marginally high during the major part of the day, *ie* 0930h - 1430h in plants grown in full light. Therefore, sun and shade leaves may be comparable as the daily photosynthetic integral and hence the dry matter production is concerned. Thus, shaded leaves in rubber plantations may significantly contribute to the total dry matter production of the plant.

INTRODUCTION

About 90% of the dry weight of higher plants is derived from CO₂ assimilated by photosynthesis (Zelitch, 1982). The amount of CO₂ assimilated by plants depends on the amount of light intercepted, conversion efficiency of intercepted light and the rate of loss of CO₂ assimilated (Kramer, 1986; Beadle *et al*, 1985 and Nelson, 1988).

In *Hevea brasiliensis* Muell. Arg. a close link exists between the CO₂ assimilation capacity and the economically important component, *ie* latex, of the plant (Nugawela, 1989). The high yielding *Hevea brasiliensis* clones exhibit a retardation of vegetative growth during exploitation for their latex (Templeton, 1969). This effect shows that there is a significant amount of assimilates partitioning towards latex production by the plant.

In many crops the majority of the leaves are found under limiting light levels (Ort and Baker 1988; Acock *et al* 1987). Similarly in *Hevea brasiliensis* plantations it can be observed that the majority of the leaves are found under limiting light levels. Therefore, it is important to study the contribution of leaves found under limiting light, *ie* shade leaves, towards total dry matter production. This information will be useful for breeders for crop improvement.

In sun and shade leaves the CO₂ assimilation rates and their diurnal variation pattern may differ. In order to compare the photosynthetic contribution of sun and shade leaves both of these factors, *ie* CO₂ assimilation rate and its diurnal variation pattern have to be studied. Instantaneous CO₂ assimilation rate of a leaf is not a good indicator of its photosynthetic contribution towards dry matter yield (Zelitch 1982; Nelson, 1988).

It is established that environmental factors like temperature and humidity (Tenhunen *et al* 1985) and plant growth stage (Downton, Grant and Loveys, 1987) influences the diurnal variation pattern in CO₂ assimilation. There is speculation that a feed back inhibition of the photosynthetic apparatus by the assimilates (Azcon-Bieto, 1983) or accumulation of abscisic acid due to water stress (Burschka, *et al*, 1985; Koppers *et al*, 1986a and 1986b) are responsible for diurnal variation in CO₂ assimilation rate. The diurnal variation pattern of CO₂ assimilation rates of sun and shade leaves are not reported earlier.

In this study the CO₂ assimilation rates and their diurnal variation patterns were studied in *Hevea brasiliensis* plants grown under different light regimes. The objective is to compare their photosynthetic contribution and also to find out factors that govern the diurnal variation pattern of CO₂ assimilation rates.

MATERIALS AND METHODS

3.1 Plant materials

Hevea brasiliensis Muell. Arg. plants of genotype RRIC 121 and grown in poly bags (17.7 x 38.1 cm) were used for this study. Plants selected were of same growth stage, *ie* two mature leaf whorls and a dormant bud.

3.2 Experimental area

An area (3m x 15m) close to the Plant Science Department Laboratories at the Rubber Research Institute of Sri Lanka, Agalawatta was chosen.

3.3 Shading

A wooden frame was built over the entire experimental area and was divided into 3 equal sections. Out of these, two sections were covered, the first with one and other with two layers of coir mats to receive 40% and 25% of incident light respectively, to the plants grown underneath. The third section was left uncovered for plants to receive 100% incident light. The light intensity was measured using a quantum sensor (LI-190 SR, Li-Cor Ltd, Lincoln Nebraska, USA). Ten plants were grown at each light level. The plants were spaced at 1.0 x 0.8m in the experimental area.

3.4 Management practices

Watering was done when required. Fertilizer was applied as recommended for polybagged plants by the Rubber Research Institute (Fertilizer to Rubber Advisory Circular No. 85 February, 1980). Dithane M 45 (10g in 4.5 liters of water) and Captan (5g in 1 liter of water) were sprayed alternatively at weekly intervals to control leaf diseases.

3.5 Measurement of gas exchange parameters

3.5.1 Equipment and calibration

Gas exchange parameters, *ie* CO₂ assimilation rate (A), stomatal conductance (g_s), internal leaf CO₂ concentration (c_i) and environmental factors, *ie* light intensity (Q), relative humidity (RH) *etc* were measured using a portable photosynthesis system, (Li-Cor Ltd., Lincoln, Nebraska, USA) comprising of a LI 6200 gas analyzer and a LI 6250 computer and software. The apparatus was calibrated prior to measurements as described by Welles (1986). A medium size leaf chamber adjusted to expose a constant area of the leaf, *ie* 10 cm², to light was used. Atmospheric air (ca=340 ppm) was drawn into the system, by switching the analyzer pump, with the leaf chamber kept open. Then the leaflets were clamped into the leaf chamber to carry-out the measurements.

Measurements were begun when a steady decline in CO₂ was observed. The relative humidity of the system was maintained at 65–70% during the measurements by varying the air flow rate through the magnesium perchlorate desiccant.

3.5.2 Selection of leaves and experimental design

Once the dormant buds developed into a mature leaf whorl under conditions provided leaflets in these whorls were selected to carry-out the measurements. Six plants were selected at random from each treatment for this purpose. CO₂ assimilation, stomatal conductance, internal leaf CO₂ concentration, ambient CO₂ concentration and light intensity were measured simultaneously in the field using the portable photosynthesis system (Li-Cor Ltd., Lincoln, Nebraska, USA). These parameters were measured at 2.5h intervals from 0700h to 1700h. To reduce variation due to sudden changes in the environmental conditions observations were taken sequentially selecting one plant per treatment.

RESULTS

4.1 CO₂ assimilation rates (A)

In all treatments the CO₂ assimilation rates were highest at 0930h. Nevertheless, in plants grown under low light levels the high rates were maintained for a longer period in the day, *eg* in plants grown under 40% and 25% of incident light the high CO₂ assimilation rates exhibited early in the day were maintained up to 1200h and 1430h of the day respectively (Table 1).

Table 1. *The CO₂ assimilation rates (A, $\mu\text{mol m}^{-2}\text{s}^{-1}$) determined at different times of the day in *Ilevea brasiliensis* plants grown under different light environments. Measurements were made under field conditions (each value is the mean of 18 observations).*

Time (h)	Light level in which plants were grown (% of incident light)			
	100	40	25	L.S.D.=0.05
0700	7.36 ^B	5.15 ^B	2.61 ^B	1.86
0930	10.56 ^A	9.92 ^A	6.75 ^A	2.30
1200	7.00 ^B	8.41 ^A	6.08 ^A	3.83
1430	5.97 ^B	5.42 ^B	5.12 ^A	2.04
1700	3.84 ^C	2.13 ^C	1.32 ^B	0.60
Mean	6.95	6.21	4.38	

(Duncan's Multiple Range Test is used to compare CO₂ assimilation rates at different times of the day of a given treatment. Means with the same letter are not significantly different. L.S.D. is used to compare the CO₂ assimilation rates in different treatments in a given time)

Treatment difference in CO₂ assimilation rates are evident only during the early and latter periods of the day, *ie* 0700h and 1700h (Table 1). The treatment difference in CO₂ assimilation rates were not evident in periods with relatively high incident light levels, *ie* 0930h, 1200h and 1430h.

The mean daily CO₂ assimilation rate (A) of plants receiving 100% and 40% of the incident light are comparable and it is relatively low in plants grown under 25% of the incident light (Table 1).

4.2 Internal leaf CO₂ concentration (c_i)

The ratio between atmospheric and internal leaf CO₂ concentration (c_i/c_a) is generally highest early and late in the day, *ie* 0700h and 1700h. It remains low and relatively constant from 0930h to 1430h except in the plants grown in full light. In plants grown in full light c_i/c_a ratio is lowest at 1430h (Table 2).

Irrespective of the time of the day, the c_i/c_a ratio is lowest in the plants grown under full light. The treatment differences in c_i/c_a ratio are not significant at 1700h (Table 2).

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Table 2. *The ci/ca ratio at different times of the day, measured under field conditions, in Hevea brasiliensis plants grown at different light levels. (Each value is the mean of 18 observations)*

Time (h)	Light level in which plants were grown (% of incident light)			L.S.D.=0.05
	100	40	25	
0700	0.874 ^A	0.911 ^A	0.947 ^A	0.028
0930	0.735 ^{CD}	0.770 ^B	0.826 ^B	0.048
1200	0.751 ^C	0.772 ^B	0.809 ^B	0.040
1430	0.704 ^D	0.755 ^B	0.799 ^B	0.053
1700	0.807 ^B	0.868 ^A	0.855 ^B	0.080

(Duncan,s Multiple Range Test is used to compare ci/ca ratio at different times of the day of a given treatment. Means with the same letter are not significantly different. L.S.D. is used to compare the ci/ca ratio in different treatments in a given time)

4.3 Stomatal conductance (gs)

Irrespective of the light environment in which plants were grown the stomatal conductance varied with time of day in a similar pattern, *ie* it decreased with time of day (Table 3). Treatment differences in stomatal conductance are not significant. Though the differences are not significant stomatal conductance is generally high in plants grown in low light levels, *ie* 40% and 25%.

Table 3. *The stomatal conductance ($\mu\text{mol m}^{-2} \text{s}^{-1}$) measured under field conditions at different times of the day in Hevea brasiliensis plants grown at different light levels. (Each value is the mean of 18 observations)*

Time (h)	Light level in which plants were grown (% of incident light)			
	100	40	25	L.S.D.=0.05
0700	0.3970 ^A	0.4339 ^A	0.5103 ^A	0.1308
0930	0.2591 ^B	0.3259 ^B	0.3082 ^B	0.1298
1200	0.1820 ^C	0.2296 ^C	0.2463 ^C	0.1377
1430	0.1665 ^C	0.1669 ^D	0.2123 ^C	0.0907
1700	0.1211 ^C	0.0980 ^B	0.1022 ^D	0.0435

(Duncan's Multiple Range Test is used to compare stomatal conductances at different times of the day of a given treatment. Means with the same letter are not significantly different. L.S.D. is used to compare the stomatal conductances in different treatments in a given time)

4.4 Light intensity (Q)

In all 3 treatments light intensities varied with time of the day in similar pattern. It increased up to 1200h and then decreased gradually (Table 4).

Table 4. *Light intensity ($\mu\text{mol m}^{-2} \text{s}^{-1}$) the plants received in different treatments at different times of day. (Each value is the mean of 18 observations).*

Time (h)	Light Level in which plants were grown (% of incident light)			
	100	40	25	L.S.D.=0.05
0700	343 ^C	199 ^C	76 ^C	92
0930	1139 ^B	556 ^B	272 ^B	204
1200	1431 ^A	771 ^A	466 ^A	236
1430	1307 ^{AB}	599 ^{AB}	364 ^{AB}	222
1700	261 ^C	114 ^C	59 ^C	41

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(Duncan's Multiple Range Test is used to compare light at different times of the day of a given treatment. Means with the same letter are not significantly different. L.S.D. is used to compare the light levels in different treatments in a given time).

4.5 Correlations with variations in CO₂ assimilation rates

The variation in CO₂ assimilation rate with time of day is significantly correlated with that of stomatal conductance (g_s), ci/ca ratio and light intensity (Q) (Table 4).

Table 4. *The correlation of CO₂ assimilation rate with stomatal conductance (g_s), internal leaf CO₂ concentration (ci/ca) and incident light (Q) level (N=90).*

Parameters	r	Significance probability
g_s	0.311	0.0029 **
ci/ca	-0.494	0.0001 ***
Q	0.574	0.0001 ***

DISCUSSION

In *Hevea brasiliensis* plantations the majority of leaves are found under limiting light levels. Therefore, it is useful to study their contribution to the total dry matter production of the plant.

In *Hevea brasiliensis* plants grown under, 100%, 40% and 25% of incident light, the CO₂ assimilation rates varied significantly with time of day. The pattern of variation in the CO₂ assimilation rate varied with the light environment in which the plants were grown. In plants grown under 100% of incident light CO₂ assimilation rate reached a peak, around 0930h and then declined gradually with time of day. In plants grown under 40% of incident light CO₂ assimilation rates reached a peak, around 0930h and it was maintained until around 1200h. The peak CO₂ assimilation rate was maintained from around 0930h to 1430h in plants grown under 25% of incident light (Table 1). Thus, the diurnal variation pattern in CO₂ assimilation rate is also dependent upon the light environment in which the plant grows. The diurnal

variation in CO₂ assimilation rate is also dependent on the growth stage of the plant (Downton *et al*, 1987) and also on environmental factors like relative humidity (Lang *et al*, 1985) and temperature (Roessler *et al*, 1985). Further, the CO₂ assimilation rates of plants grown under different light levels were comparable when light levels are relatively high, *ie* 0930h–1430h. *Hevea* is a C₃ plant and therefore light saturation may occur even at 40% of the incident light. Further, when leaves develop under low light levels they get adapted to use the available light efficiently. Such adaptations have been reported by Syvertsen (1984), Nelson and Ehlers (1984).

The peak CO₂ assimilation rate is only marginally high in leaves grown under full light when compared with leaves grown under low light. Further, the high CO₂ assimilation rates are maintained for a longer period of the day in plants grown under low light levels. Thus, the daily photosynthetic integral of leaves developed under 40% of incident light is comparable to the leaves developed under full light. Thus, it could be concluded that leaves grown in limiting light *ie* ca. 40% incident, can contribute significantly towards total dry matter production.

Diurnal variation pattern in ci/ca ratio is different in plants grown under full light. In plants grown in low light levels the relatively low ci/ca values are maintained from 0930h to 1430h. Nevertheless, in plants grown under high light the ci/ca ratio increases from 0930h to 1200h and again declines from 1200h to 1430h (Table 2).

The diurnal variation pattern in stomatal conductance is similar in plants grown in different light levels. It decreases steadily with time of day (Table 3). Though the stomatal conductance decreases significantly from 0930h to 1430h the ci/ca ratio remains relatively unchanged during this period, especially in plants grown under low light levels. The relatively unchanged ci/ca values, although the stomatal conductance is decreasing suggests that CO₂ entering in to the leaf is not being utilized for photosynthesis. Hence, it could be concluded that the mesophyll limitations for photosynthesis are increasing with time of day. This is clearly evident as the CO₂ assimilation rates are relatively low later in the day in all 3 treatments, eventhough the incident light levels are alike (Table 1 and Table 4). Similar results have been reported by Koppers *et al* (1986 a). Such limitations in mesophyll cells are attributed to accumulation of abscisic acid (Raschke and Hedrich 1985) or end product inhibition (Azcon - Bieto 1983). It is reported that abscisic acid is biosynthesised within the leaves when they are exposed to long periods of high light (Raschke and Hedrich 1985). Biosynthesis of abscisic acid may have taken place in *Hevea brasiliensis* plants grown in full light decreasing CO₂ assimilation rate early in the day than in plants grown in low light. The variation in CO₂ assimilation rates are either due to that of stomatal conductance or mesophyll limitations (Voncammerer, and Farquhar, 1984).

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In the study it is apparent that variation in CO₂ assimilation rate is significantly correlated with that of stomatal conductance and ci/ca ratio (Table 4).

It is evident that the peak CO₂ assimilation rates of leaves developed under full light are only marginally higher than those developed under 40% incident light. Nevertheless, in leaves developed under 40% and 25% of incident light the peak CO₂ assimilation rates are maintained for a longer period of the day. Therefore, the daily photosynthetic integral is comparable in leaves receiving 40% of the incident light and full light. It is only about 40% less in leaves receiving 25% of the incident light. Hence, shade leaves may also contribute significantly towards the productivity of the plant.

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