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Early Selection of Promising High Yielding Hevea *Progenies based on Selected Physiological and Stomatal Characteristics*

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Leaf physiological and stomatal parameters were studied from immature plants grown in the ground nursery. These Hevea *clones differed widely in mature yield. Significant differences were detected in some of the parameters such as light saturated photosynthesis (Psat), stomatal conductance (SC), transpiration rate (TR), mesophyll conductance (MCond) and instantaneous water use efficiency (WUEg) among the clones. The variation in mature yield, girth at opening as well as physiological parameters and stomatal characteristics were not affected by weather. The mature yield showed a positive correlation with Psat, SC, TR and MCond while the girth at opening was positively correlated with instantaneous WUEg. A step-wise multiple regression confirmed results of the simple correlation. About 34% of the differences in mature yield was explained by Psat. The addition of potential conductance index (PCI), a product of stomatal frequency (SF) and square of length of stomatal complex (LSC), as an independent variable increased the predictive power for mature yield by about 55%. The results indicated that Psat singly could identify high yielding clones with 63% accuracy and an error rate of 17% for wrongly identifying low yielders as high yielding clones. When both Psat and PCI were used, all high yielding clones could be identified. It is concluded that both Psat and PCI, which together have successfully identified high yielding clones, have a potential to be used as indirect selection criteria for yield of* Hevea *progenies during the early stages of growth in the nursery.*

Keywords: light saturated photosynthesis; stomatal conductance; transpiration rate; mesophyll conductance; water use efficiency; early selection; mature yield; physiological and stomatal characteristics

Abbreviations: PPFD: Photosynthetic photon flux density, Psat=light saturated photosynthesis, P_N= net photosynthesis, P80= photosynthesis at low irradiance, SC = Stomatal conductance, MCond= Apparent mesophyll conductance, TR=Transpiration rate, WUEg=Instantaneous water use efficiency, EffTR= Efficiency of transpiration rate, QY=Apparent quantum yield, VPD= Vapour pressure deficit, LCP= Light compensation point, gtt = g/tree/tapping.

Hevea is a perennial crop with a long breeding cycle since it takes about 25–30 years from the time of hand pollination to large scale trials before promising clones can be recommended for commercial planting[1]. In the past, there were several attempts at developing an early

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selection technique with the main objective of culling inferior seedlings and selecting the promising ones at an early stage. Tan[2] has given a review on this subject. Currently, yield potentials of progenies are predicted by test-tapping using the modified Hamaker-Morris-Mann technique[3] which involves tapping two to three year old seedlings for three cycles or three months using the 1/2S d/3 tapping system. By this technique, potential yield of progenies can be predicted 3 to 4 years after planting of seedling progenies. This technique developed in the nineteen thirties, is still being widely used by breeders in rubber producing countries although nursery yield has been shown to be a weak predictor for mature yield[1,4]. Given the long breeding cycle of rubber, there is a need to develop a rapid and early selection technique of promising high yielding progenies at the nursery stage when thousands of progenies are produced from hand pollination. This is made possible by using selected physiological traits as criteria in the screening techniques of *Hevea* progenies.

Several researchers have attempted using photosynthetic rate (PR) and stomatal characteristics as an indirect method of yield prediction. Differences in photosynthetic rate have been observed among *Hevea* clones[5–8]. Although correlation studies have shown no significant association between PR and nursery yield[8,9] , a weak positive correlation (r^2=0.23 P<0.1) has been established between PR and mature yield[5]. Photosynthetic rate, a trait with high coefficient of variability (CV), was also shown to be heritable with relatively high broad-sense heritability of 44% indicating that it has sufficient genetic variability to respond to genetic manipulation[5,6].

Clonal differences in stomatal frequency and dimension of stomatal pore in rubber have been reported[10–14] although no significant relationship was found between stomatal frequency and yield potential of the clones studied[13–14].

The weak relationship between PR and mature yield reported by Samsuddin[5] suggests that probably more than one physiological factor may be involved as a determinant of rubber yield. In the present study, leaf photosynthesis and other physiological and stomatal parameters of several *Hevea* clones varying widely in mature yield, as well as environmental factors, were studied with the objective of identifying more parameters likely to be associated with mature yield and girth at opening.

MATERIALS AND METHODS

Preparation of Planting Materials

For the study of leaf photosynthesis and stomatal characteristics, 23 clones of *Hevea brasiliensis* with widely variable mature yields were grown in Field 68 at the Rubber Research Institute of Malaysia, Experimental Station (RRIES), Sungei Buloh, Selangor (at latitude of 03º 07 'N, longitude of 101º 33 'E and day and night temperature of 33/24ºC). The experiment was laid out in a randomised block design with three replicates. Each plot consisted of four plants per clone planted at a planting distance of 3 m × 3 m. The plants were given recommended cultural practices for immature rubber plants with respect to weed control and manuring. Whenever necessary, the shoots were cutback at a height of about 1.5 m to obtain fresh crops of mature leaves for measurement of leaf photosynthesis. Young emerging leaves with a bronze colour were sprayed with fungicide to ensure that the leaves were free of leaf diseases.

Gas Exchange Parameters

In an attempt to narrow down the list of parameters most closely associated with mature yields, a preliminary study was carried out from August 2006 to June 2007. In this preliminary study, net leaf photosynthesis (PN) were measured using LI6400P (LICOR, Oregon, USA) portable open gas exchange system, followed by construction of light response curves. The measurement was done on a sunlit and recently expanded mature leaflet in the terminal whorl on sunny days from 8.30 to 15.00 h with cuvette conditions of the instrument set at ambient (Sample CO_2 set at 350 µmole mol^{-1}, chamber humidity at least 50% and vapour pressure deficit at about 2 units) . Extreme care was taken to use only healthy leaves free from leaf diseases and other physiological maladies such as water logging, malnutrition and water stress. P_N of each clone was measured at various photosynthetic photon flux density (PPFD) (1500, 1200, 400, 150, 100, 50 and 0 µmole m^{-2} s^{-1}) in a decreasing order. Light intensity was provided by a LED source fitted to the leaf chamber. CO_2 gas contained in a mini-cartridge was injected into the open system using a CO_2 injector system (LI-6400-01, LICOR, USA). P_N at each light intensity was recorded after about 3 mins when equilibrium had been attained. P_N /PPFD light response curve was constructed by fitting a 4- parameter function to the data with P_N and PPFD as the dependent and independent variable respectively using a KC-4 programme (Bio-Tek Instruments, USA). An r^2 of more than 99% was obtained for all curves. Outputs such as photosynthesis at saturated irradiance (Psat) and low (P80) irradiance (80 µmole m^{-2} s^{-1}), apparent quantum yield (QY) and light compensation point (LCP) were calculated from the individual curve. This system also simultaneously measures other related parameters such as transpiration rate (TR), stomatal conductance (SC) vapour pressure deficit (VPD) and internal CO_2 concentration (Ci) while other parameters such as instantaneous water use efficiency (WUEg), (Psat/stomatal conductance), transpiration efficiency (ETr) (Psat/TR) and mesophyll conductance (MCond) (Psat/Ci) were calculated.

For each clone, a light response curve of one leaflet per plant per replicate was determined from at least four plants. Records of ambient meteorological data such as air temperature, relative humidity, wind speed, rainfall and solar radiation at each date of measurement were provided by the Subang Meteorological Station under the Meteorological Department of Malaysia, Petaling Jaya, Selangor.

In the second part of the study, Psat were calculated with the instrument set at a survey mode. The instrument was set at the following settings: sample CO_2 concentration set at 400 µmole, flow rate at 500 units, external humidity of 50–70%, block temperature of 30ºC and light intensity at 1500 µmole $m^{-2}s^{-1}$. Psat was recorded after the system had attained stability after about two minutes. For each clone, Psat of three leaflets (with similar physical conditions as those used in the preliminary study) per plant per replicate were measured from at least four plants.

Stomatal Frequency and Size of Stomatal Complex

Stomatal characteristics such as stomatal frequency and length and width of stomatal complex were determined from leaves used for measurement of leaf photosynthesis in the second part of the study. A stomatal complex is defined as the structure comprising of two guard cells that border a stomatal pore. For each clone, impressions of the abaxial epidermal leaf surface were taken from two fully and recently expanded leaves of the terminal whorl of one plant per replicate. To fix the epidermal section, colourless nail varnish was applied to an area of about 3 cm^2 half-way between the leaf base and leaf apex near the midrib. The epidermal area was then stripped using cellophane tape before placing onto microscopic slides for examination under a light microscope (Olympus BH2) connected to a computer image analysis system (Video test-size 5.0 software). Images on the TV monitor were magnified by about eight-fold. For each leaf replica, length and width of stomatal complex of ten randomly selected stomata were measured at 200× magnification. The number of stomata in five randomly selected microscopic field of view (0.1317 mm^2) per leaf replica was counted at a magnification of 100×. Stomatal frequency was expressed as number per mm^2 of leaf surface.

Stomatal conductance is directly related to size and frequency of stomata on the leaf surface[15]. In this study, SF and LSC were used to estimate total conductance to water vapour using the equation proposed by Holland and Richardson[16]:

$$PCI = (LGC)^2 \times SF \times 10^{-4} \qquad \ldots 1$$

Where *PCI* = potential conductance index, *LGC*=length of guard cell (taken to be equivalent to LSC), *SF*= stomatal frequency.

Statistical Analysis

The analysis of variance of leaf and stomatal parameters were carried out on clone means averaged across dates of measurement using the GLM procedure in SAS (Statistical Analysis System). The probability level of clone mean square was obtained by testing against the error mean square; the clone means over all dates were then compared by Duncan Multiple Range Test at $P< 0.05$.

The relationship of vigour and productivity of mature clones on leaf physiological and stomatal traits of immature plants and ambient meteorological data were examined using simple correlations and step-wise multiple regressions. The data on mature yield, which were means of the first five years of tapping and girth at opening of each clone, were taken from reports published in the RRIM Planters' Bulletin from 1968 to1998 and in the Bulletin of Science and Technology, Malaysian Rubber Board[17].

RESULTS AND DISCUSSION

Simple Correlations

In the preliminary study in which P_N at various light intensities (PPFD) were determined to produce light response curves, we failed to show any significant association between QY, determined as the initial slope of the light response curve, and mature yield suggesting that this parameter is not a determinant of mature yield among *Hevea* clones studied (*Table 1*). These results are contrary to those of Nugawela *et al.*[18] who found significant and positive correlation between QY and yield potential of several *Hevea* clones.

Meteorological parameters recorded during the course of the preliminary study were found to be correlated to each other and to VPD as well as mature yield. However, they had no effect on SC and other physiological parameters. Rainfall was positively correlated with VPD (r^2=0.486*) and wind speed (r^2=0.842***). RH was positively correlated with wind speed (r^2=0.721**), rainfall (r^2=0.80***) but negatively correlated with air temperature (r^2=-0.702***) and mature yield (r^2=-0.464*). Solar radiation was positively correlated with VPD (r^2=0.596**), air temperature (r^2=0.565**) and rainfall (r^2=0.594**).

Unless otherwise stated, the remaining part of the text will refer to results obtained from the second part of the study in which the photosynthesis measuring instrument was set at the survey mode. Using this mode, P_N was measured at one PPFD level only (1 500 µmole m^{-2} s^{-1}) compared to seven levels of PPFD used in the light response curve.

TABLE 1. SIMPLE CORRELATION OF MATURE YIELD AND GIRTH AT OPENING ON LEAF PHYSIOLOGICAL PARAMETERS IN THE PRELIMINARY STUDY

Parameters	QY	SC	VPD
Psat	0.364#	0.699 ***	-0.226 NS
P80	0.049 NS	-0.033 NS	-0.356 #
SC	0.685 ***	–	-0.543 **
Ci	0.667 ***	0.894***	-0.69 ***
TR	0.448 *	0.802***	0.025NS
WUEg	-0.651 **	-0.918***	0.616 **
EffTR	-0.326 NS	-0.593 **	-0.187 NS
MCond	0.088 NS	0.375 #	0.043 NS
Ci/Ca	0.666 ***	0.889***	-0.708***
LCP	-0.131 NS	-0.676 ***	0.364 #
VPD	-0.504 *	-0.543 **	–
MY	0.005 NS	0.28 NS	-0.289 NS
Girth	-0.274 NS	0.217 NS	0.019 NS

NS,#,*,**,***: F-test non-significant or significant at P<0.10, 0.05, 0.01 or 0.001 respectively.
Psat= Photosynthesis at saturated irradiance, P80= Photosynthesis at low irradiance, SC= Stomatal conductance, Ci = Intercellular CO_2 concentration, TR= Transpiration rate, WUEg= Instantaneous water use efficiency, EffTR= Efficiency of transpiration rate, MCond = Mesophyll conductance, Ci/Ca= Ratio of intercellular CO_2 concentration to ambient CO_2 concentration, LCP= Light compensation point, VPD=Vapour pressure deficit, QY=Apparent quantum yield, MY=Mature yield.

Thus, the procedure used in the survey mode would enable faster measurement of Psat on more leaves, an essential requisite if Psat is to be used as an early selection technique. Precision of the Psat results would improve with the increase in sample size which would eventually reflect the true characteristics of the clones being studied.

Psat and other physiological and stomatal parameters of various clones are presented in *Table 2*. The analysis of variance revealed that significant differences existed among the clones for some of these parameters such as Psat, SC, TR, MCond, WUEg and SF. However, no significant differences in LSC, WSC and PCI could be found between clones.

Table 3 shows simple correlations between girth at opening, mature yield and leaf physiological and stomatal characteristics. The stomatal conductance was positively and significantly correlated with Psat, Ci, TR, Ci/Ca ratio and MCond but was negatively correlated with VPD, WUEg and transpiration efficiency (EffTr). Psat was positively and significantly correlated with TR and SC indicating that it is under stomatal control, but was negatively correlated with WUEg, EffTr and VPD. These results indicated that most of the physiological parameters studied were interrelated to one another as they occured through the stomatal aperture. Psat also appeared to be under mesophyll influence as indicated by its positive and significant

TABLE 2. LEAF PHYSIOLOGICAL AND STOMATAL PARAMETERS IN SEVERAL CLONES OF *HEVEA*

Clone	Psat	SC	TR	MC × 10^2	SF	LSC	WSC	PCI	MY	Girth (cm)
RRIM 902	15.19 a	0.36 ab	5.74 a	5.23 ab	625 abcd	23.8 a	18.2 a	35.4 a	51.4	64.2
RRIM 600	15.05 a	0.336 abc	5.07 abc	5.27 a	645 abcd	25 a	18.8 a	40.3 a	35.1	54.5
PB 350	14.86 ab	0.35 abc	5.42 abc	5.07 abc	626 abcd	24.8 a	18.3 a	38.5 a	59	NA
RRIM 928	14.55 abc	0.346 abc	5.61 ab	5.03 abcd	696 cd	22.1 a	16.3 a	34 a	53.4	60.1
PB 280	14.50 abc	0.339 abc	5.23 abc	5.10 abc	554 ab	23.2 a	17.6 a	29.8 a	35	51.5
PB 260	14.22 abcd	0.317 abc	5.24 abc	4.93 abcde	546 a	24.9 a	18.7 a	33.8 a	50	58
RRIM 2001	14.19 abcd	0.365 a	5.72 a	4.80 abcde	582 abc	26.1 a	19.4 a	39.6 a	57.6+	41.1
PR 255	14.17 abcd	0.312 abc	4.76 abc	4.93 abcde	602 abc	23.3 a	17.9 a	32.7 a	36.5	NA
RRIM 911	13.94 abcd	0.305 abc	4.85 abc	4.90 abcde	605 abc	24.3 a	18 a	35.7 a	47.5	62.9
PB 359	13.39 abcde	0.290 abc	4.82 abc	4.67 abcdef	540 a	24.3 a	17.8 a	31.9 a	38.2	NA
PB 235	13.39 abcde	0.312 abc	4.87 abc	4.57 bcdefg	566 abc	24.8 a	18.4 a	34.8 a	45.1	63.7
RRIM 929	13.36 abcde	0.287 abc	5.04 abc	4.70 abcdef	595 abc	25.1 a	18.6 a	37.5 a	50.6	62.4
PB 366	13.14 bcde	0.287 abc	4.62 abc	4.57 bcdefg	690 bcd	23.4 a	18.1 a	37.8 a	50.2	53.6
RRIM 2016	13.00 bcde	0.300 abc	4.59 abc	4.43 cdefg	680 abcd	24.8 a	19.5 a	41.8 a	56.8+	42.8
RRIM 908	12.87 cde	0.272 abc	4.64 abc	4.50 cdefg	673 abcd	23 a	18.3 a	35.6 a	40.4	64
RRIM 936	12.76 cde	0.307 abc	4.87 abc	4.30 efg	571 abc	24.1 a	17.5 a	33.2 a	38.3+	53.5
RRIM 921	12.72 cde	0.315 abc	5.13 abc	4.30 efg	747 d	22.9 a	18 a	39.1 a	40.6	65.3
RRIM 2015	12.69 cde	0.325 abc	5.21 abc	4.37 defg	634 abcd	23 a	18 a	33.5 a	33.8	43.6
PB 355	12.54 de	0.295 abc	4.50 bc	4.25 efg	581 abc	23.4 a	17.2 a	31.8 a	37.3	60.7
RRIM 2008	12.42 de	0.293 abc	4.79 abc	4.23 efg	573 abc	22.4 a	17.1 a	28.7 a	29.6	45.2
RRIM 938	11.75 ef	0.269 bc	4.57 abc	4.03 fg	623 abcd	24.6 a	18.8 a	37.7 a	38.5+	54.6
RRIC 100	11.57 ef	0.264 c	4.34 c	3.90 g	643 abcd	24.7 a	18.9 a	39.2 a	34.8	NA
RRIM 513	10.56 f	0.170 d	3.28 d	4.03 fg	565 abc	23.8 a	18.7 a	32 a	28.9	NA
Probability level	***	**	**	***	*	NS	NS	NS		
CV	7.65	15.37	11.96	7.64	8.8	4.1	4.1	9.9	21.2	14.5
EMS	0.9342	0.0022	0.3455	0.000012	4871.3	2.454	2.056	23.15	NA	NA

*,**,***: F-test significant at P< 0.05, 0.01or 0.001 respectively.

Duncan's Multiple Range Test for each parameter: α=0.05, DFe=43, EMS=Error Mean Square.

Within a column, means followed by same letters are not significantly different (P<0.05) .

Psat = Photosynthesis at saturated irradiance (μmol(CO_2)$m^{-2}s^{-1}$), SC = Stomatal conductance, TR= Transpiration rate, WUEg= Instantaneous water use efficiency, MC=Mesophyll conductance (Psat/ Ci); SF = Stomatal frequency (mm^{-2}), LSC = Length of stomatal complex(u), WSC= Width of stomatal complex(u), PCI = Index of potential conductance, MY = Mature yield (g/tree/tapping), CV=Coefficient of variability (%), NA = Not available, + = Results obtained from Large Scale clone trial are averages of two years.

TABLE 3. CORRELATION COEFFICIENTS BETWEEN LEAF PHYSIOLOGICAL PARAMETERS AND OTHER PARAMETERS, AND AMONG THEMSELVES FOR 23 *HEVEA* CLONES

Parameters	Psat	SCond	Ci	TR	WUEg	EffTR	MCond	Ci/ Ca	VPD	SF	LSC	WSC	PCI
Psat	-												
SCond	0.865***	-											
Ci	0.342NS	0.684***	-										
TR	0.847***	0.956***	0.572**	-									
WUEg	-0.498*	-0.828***	-0.942***	-0.749**	-								
EffTR	-0.125NS	-0.519*	-0.651**	-0.617**	0.734***	-							
MCond	0.962***	0.726***	0.082NS	0.733***	-0.267NS	0.059NS	-						
Ci/ Ca	0.392NS	0.691***	0.909***	0.624**	-0.887***	-0.632**	0.169NS	-					
VPD	-0.512*	-0.618**	-0.713***	-0.381#	0.689***	0.022NS	-0.356#	-0.644**	-				
SF	0.017NS	0.107NS	0.078NS	0.113NS	-0.101NS	-0.173NS	-0.006NS	0.135NS	0.024NS	-			
LSC	0.195NS	0.102NS	0.118NS	0.082NS	-0.103NS	0.0546NS	0.187NS	0.166NS	-0.165NS	-0.316NS	-		
WSC	-0.074NS	-0.139NS	-0.08NS	-0.145NS	0.089NS	0.116NS	-0.031NS	0.044NS	0.029NS	0.01NS	0.792***	-	0.682**
PCI	0.173NS	0.174NS	0.173NS	0.153NS	-0.177NS	-0.091NS	0.146NS	0.26NS	-1.41NS	0.601**	0.567**	0.682***	-
Girth	0.119NS	-0.26NS	-0.456*	-0.103NS	0.542*	0.235NS	0.15NS	-0.508*	0.347NS	0.164NS	-0.158NS	-0.252NS	-0.03NS
MYield	0.586**	0.519*	0.232NS	0.559**	-0.29NS	-0.208NS	0.549**	0.277NS	-0.153NS	0.25NS	0.408#	0.248NS	0.544**

NS,#,*,**,***: F-test non-significant or significant at P < 0.10, 0.05, 0.01, 0.001 or 0.0001 respectively.

Psat = Photosynthesis at saturated irradiance, SC = Stomatal conductance, Ci = Intercellular CO_2 concentration, TR = Transpiration rate, WUEg= Instantaneous water use efficiency, EffTR = Efficiency of transpiration rate, MCond = Mesophyll conductance, Ci/Ca= Ratio of intercellular CO_2 concentration to ambient CO_2 concentration, VPD = Vapour pressure deficit, SF = stomatal frequency, LSC = Length of stomatal complex, WSC = Width of stomatal complex, MYield = Mature yield.

correlation with MCond. However, a detailed study done by Nataraja and Jacob[6] has shown that mesophyll rather than stomatal factors are more important in regulating photosynthesis in *Hevea*. The negative association between VPD and SC, also observed in the preliminary study (*Table 1*), indicated that in woody plants, changes in SC was partly accounted for by changes in ambient VPD the other factor being irradiance[19]. No correlation between stomatal characters (SF, LSC and WSC) and any of the physiological parameters studied was found. Similarly, Samsuddin[10] found no significant relationship between SF or dimension and stomatal conductance.

The mature yield was significantly and positively correlated to Psat, SC, TR, MCond and PCI although taken singly, none of the stomatal characters was found to be associated with the mature yield. Girth at opening was positively correlated with WUEg but negatively correlated with the Ci/Ca ratio.

Step-wise Multiple Regression

Early Selection Criteria for Mature Yield. The association between mature yield and Psat and PCI and between girth and WUEg was confirmed by a step-wise multiple regression analysis which showed that both Psat and PCI were important variables in predicting mature yield (*Table 4*). About 34% of the variation in mature yield was accounted for by Psat; this result agrees with a report given by Samsuddin[5]. The addition of PCI as an independent variable increased the predictive power for mature yield by about 55%.

Accuracy of Regression Equations for Mature Yield. To determine the accuracy of the formula in predicting mature yield, the top eight clones with high mature yield were compared with those with high Psat and PCI. Clones were considered as high yielding if mature yield was similar or higher than yield of PB 260 control (50 gtt); thus the high yielding clones in this study were RRIM 2001, RRIM 2016, RRIM 902, RRIM 928, RRIM 929, PB 366, PB 260 and PB 350.

The results showed that when only Psat was used as a criterion for selection, Psat identified five out of the eight high yielding clones giving 62.5% accuracy; these clones were RRIM 2001, RRIM 902, RRIM 928, PB 350 and PB 260. Psat also wrongly identified three clones (RRIM 600, PB 280 and PR 255) as having high predicted yield while in reality they are low yielders thus giving an error rate of 17%. Samsuddin[5] working on 23 *Hevea* clones had obtained percentage accuracy of about 50% for correctly predicting high yielding clones based on Psat data. The top eight clones with high PCI also had five common clones with the high yielding clones (62.5% accuracy) and an error rate of 17%. Between them, Psat and PCI identified two similar high-yielding clones (RRIM 2016 and RRIM 929) while identifying three other dissimilar high-yielding clones. Consequently, when both Psat and PCI were used as parameters for selection, it was possible to identify all the high yielding clones studied.

The bottom ten clones of Psat had six common clones (RRIM 513, RRIM 2008, RRIM 2015, RRIM 936, RRIC 100 and PB 355) with bottom ten clones of mature yield (60% accuracy). PCI identified eight clones to be low yielders giving an accuracy rate of 80%, while it wrongly identified two high-yielding clones as low yielders hence giving an error rate of 8.7%. This indicates that both Psat and PCI can be used to select for high yielding progenies as well as culling of poor yielders.

The study showed that *Hevea* yield was influenced by both Psat and total stomatal conductance estimated by PCI (the product

TABLE 4 . STEP-WISE MULTIPLE REGRESSIONS OF GIRTH AND MATURE YIELD ON LEAF PHYSIOLOGICAL AND STOMATAL PARAMETERS

Equation	Dependent variable	Regression equation	R^2
1	Girth at opening (G)	G = -16.241 + 1.567 WUEg*	0.294
2	Mature yield (MY)	MY= -18.106 + 4.607 Psat **	0.344
3	Mature yield (MY)	MY= -52.044 + 3.988Psat ** + 1.190 PCI**	0.546

*,**: F-test significant at P< 0.05 or 0.01 respectively.
Psat= Photosynthesis at saturated irradiance, WUEg= Instantaneous water use efficiency,
PCI= Potential conductance index

of SF and square of LSC). TR was implicated in this relationship since at the leaf level, the rate of TR was controlled largely by SC which depended on stomatal frequency and dimension[15]. This was in agreement with the simple correlation results obtained in the present study which showed significant correlation between SC and TR and between MY and TR (*Table 3*). It is generally known that water relations of plants have a large influence on plant productivity. This is even more pertinent in *Hevea* since much water is removed from the tree when latex is harvested during tapping. The water lost through tapping must be replenished from the soil through the transpiration stream. The high transpiration rate induces cooling effect of the canopy; this is important for tree productivity since many leaf biochemical and physiological processes such as leaf photosynthesis and floral bud retention in cotton have lower optimum temperature than ambient air temperature[20,21]. This is in agreement with results of the present study which showed the importance of total conductance and hence transpiration in influencing yield of *Hevea* clones. It is probable that in high yielding *Hevea* clones, photosynthesis is sustained by canopy cooling arising from high rate of transpiration.

Early Selection Criterion for Vigour. Besides yield, vigour as measured by girth of the trunk, is another important selection criteria used by plant breeders in *Hevea* breeding programme. High vigour is an important trait associated with early tapping and high yield[22]. The prediction of trunk size would be useful in rubber forest plantation where bole size is highly valued.

Significant correlation between girth at opening and instantaneous water use efficiency (WUEg) (*Table 3*) was further confirmed by results of a step-wise multiple regression as shown in *Table 4*. This observation probably underscores the important role played by environmental factors in determining vigour (as estimated by girth) of *Hevea* during the immature phase, since WUEg in the present study was shown to be closely related to vapour pressure deficit (VPD) (*Tables 1* and *3*). However, in our study, no significant correlation was observed between girth at opening and meteorological parameters such as air temperature, relative humidity, wind speed, rainfall and relative humidity (data not presented). This was probably because the meteorological parameters had been collected over a short period only during measurements of leaf physiological parameters. Elsewhere, it was reported that environmental factors such as site, cumulative sunshine hours and rainfall of the location have a large influence on immature vigour of *Hevea*[22–24], with the last two variables collected over a period of five years shown to be capable of determining potential vigour of a clone. Jayasekera *et al.*[23] also reported that the final size of a rubber tree can also be estimated from post-tapping tree size at eight years after planting.

CONCLUSION

This study shows that a regression model based on Psat and PCI has a potential to be used as an indirect early selection criteria for high-yielding *Hevea* clones. Apart from the initial cost of acquiring a LI6400P photosynthesis measuring instrument (LICOR, USA) and the setting up of an anatomical laboratory, the research does not require other costly capital expenditure. The technique is fast and precise enabling it to be used during the early stages of a breeding programme when a large population of progenies (about 1 400) are produced. Besides saving time, the use of Psat and PCI as early selection criteria would also provide savings in cost, space and labour. However, more research is required before the technique can be used as an accurate and reliable alternative to the nursery screening technique by test-tapping. The first step is to determine the accuracy of the regression model in predicting mature yields of progenies released from the breeders' hand pollination programme. This requires validation of the regression models for their accuracy in identifying high-yielding progenies at each stage of the breeders selection programme namely the nursery selection, small scale clone trial and large scale clone trial. Since our results indicate weather data do not have any effect on leaf parameters, the measurement of Psat and stomatal characteristics can be carried out over several months depending on the size of progenies produced from crossings without any adverse affect on the final results.

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