



## **ATP concentration in latex as a biochemical marker for early evaluation of yield in *Hevea brasiliensis***

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### **Introduction**

- Energy metabolism plays an important role in the latex production mechanism in *Hevea*.
- A large supply of biochemical energy in the form of ATP is required to sustain the metabolic activities to regenerate rubber and other sub cellular components such as proteins and nucleic acids after tapping.
- Latex flow also depends on biochemical energy availability.

### Cont'd

- ATP is also the specific substrate for the H<sup>+</sup> pumping ATPase located on the lutoid membrane which regulates the pH of the cytosol and energises the transport of various solutes inside the lutoids (D'Auzac. 1977; Jacob *et al*, 1997; Chrestin *et al*, 1984; Marin *et al*, 1982).
- Thus ATP is a very important regulator in rubber synthesis through its direct effect on metabolic pathways and indirect effect mediated through ATPase activity.

### Previous studies

- The concentration of adenylates (ATP, ADP and AMP), Adenylate energy charge (AEC), ATPase and C-serum pH when measured in high and low yielding clones showed significant positive correlation with latex yield.
- ATP had a large significant correlation with AEC
- ATP concentration in latex was significantly high in high yielding clones both during peak yielding and summer season.

(Sreelatha *et al*, 2004)

## Objective

**To establish the possibility of using ATP as a biochemical marker for early evaluation of yield and to make use of it in crop improvement programme.**

## **CLONES SELECTED FOR THE STUDY (MATURE TREES)**

RRII 105 , RRIM 600, PB 235, PB 260, PB 217 (High yeilders)  
 GT1, PB 5/51, Tjir 1 (Medium yielders)  
 RRII 38, RRII 33 (Low yielders)

**Year of planting : 1992**  
**Location : RRII**  
**Design : RBD**  
**Sample size : Nine trees/clone**

Trees were under S/2 d3 6d/7 tapping system.

The measurements were carried out during the peak yielding season (September, October and November)

### CLONES SELECTED FOR IMMATURE PHASE STUDY

RRII 105 , RRIM 600, PB 235, PB 260, PB 217 (High yeilders)

GT1, PB 5/51, Tjir 1 (Medium yielders)

RRII 38, RRII 33 (Low yielders)

Year of planting : 2006

Location : Central Experiment Station

Design : RBD

Sample size : 15 plants/clone

Latex samples were collected by making small cuts during September, October and November.

### Methodology

Fresh latex samples were extracted with 2.5% Trichloro acetic acid and neutralised with 0.1N KOH and made up to a known volume with 30mM HEPES buffer pH 7.4 (Amalou *et al*, 1992) and quantified using Bioluminescent kit (FL-AA, Sigma) containing luciferin-luciferase.

## Statistical analysis

ANOVA and DMRT were carried out to determine the mean differences in dry rubber yield and latex ATP between clones.

Correlation was worked out with dry rubber yield and latex ATP as well as with latex yield and ATP in corresponding days.

**Table-I Rubber yield (g/t/t) of *Hevea* clones in the first five years of tapping and mean yield over five years**

Clones	Yield (1 <sup>st</sup> yr)	Yield (2 <sup>nd</sup> yr)	Yield (3 <sup>rd</sup> yr)	Yield (4 <sup>th</sup> yr)	Yield (5 <sup>th</sup> yr)	Mean yield Over 5yrs
RRII 105	34.6 <sup>bc</sup>	58.4 <sup>b</sup>	68.3 <sup>b</sup>	84.6 <sup>a</sup>	74.9 <sup>b</sup>	64.1 <sup>b</sup>
RRIM 600	29.4 <sup>cd</sup>	40.7 <sup>d</sup>	48.4 <sup>cd</sup>	55.5 <sup>c</sup>	57.4 <sup>c</sup>	46.3 <sup>c</sup>
PB 217	35.2 <sup>bc</sup>	47.2 <sup>cd</sup>	51.3 <sup>cd</sup>	54.3 <sup>c</sup>	48.5 <sup>d</sup>	47.3 <sup>c</sup>
PB 235	40.4 <sup>b</sup>	69.0 <sup>a</sup>	78.0 <sup>a</sup>	84.5 <sup>a</sup>	87.4 <sup>a</sup>	70.9 <sup>a</sup>
PB 260	48.6 <sup>a</sup>	53.8 <sup>bc</sup>	77.2 <sup>a</sup>	69.6 <sup>b</sup>	82.4 <sup>a</sup>	66.3 <sup>ab</sup>
PB 5/51	21.2 <sup>ef</sup>	26.4 <sup>e</sup>	26.0 <sup>e</sup>	33.4 <sup>d</sup>	39.3 <sup>e</sup>	29.6 <sup>e</sup>
Tjir I	18.9 <sup>f</sup>	30.7 <sup>e</sup>	43.0 <sup>d</sup>	46.3 <sup>c</sup>	43.3 <sup>de</sup>	36.4 <sup>d</sup>
GTI	26.9 <sup>de</sup>	47.6 <sup>cd</sup>	54.1 <sup>c</sup>	49.9 <sup>c</sup>	49.5 <sup>d</sup>	45.2 <sup>c</sup>
RRII 38	8.5 <sup>g</sup>	11.0 <sup>f</sup>	11.3 <sup>f</sup>	13.4 <sup>f</sup>	16.7 <sup>f</sup>	11.7 <sup>f</sup>
RRII 33	8.3 <sup>g</sup>	8.6 <sup>f</sup>	20.4 <sup>e</sup>	23.0 <sup>e</sup>	16.4 <sup>f</sup>	15.8 <sup>f</sup>

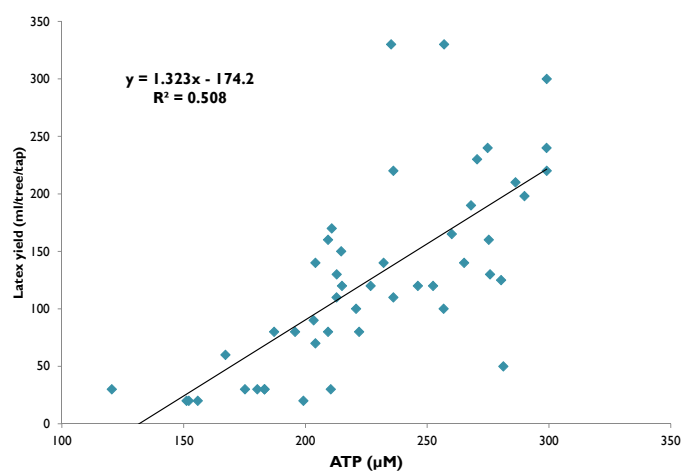
Means followed by a common letter are not significantly different at  $p < 0.05$ .

Table-I Rubber yield and ATP in different *Hevea* clones during peak season (Sep-Nov)

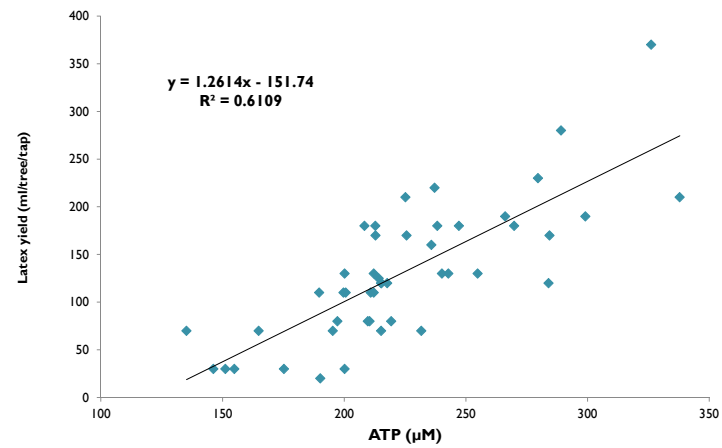
Clones	Yield (g/t/t)	ATP ( $\mu$ M)
RRII 105	91.8 <sup>a</sup>	278.1 <sup>a</sup>
RRIM 600	61.5 <sup>b</sup>	253.6 <sup>bc</sup>
PB 217	55.0 <sup>bc</sup>	218.4 <sup>de</sup>
PB 235	63.2 <sup>b</sup>	232.9 <sup>cd</sup>
PB 260	59.9 <sup>b</sup>	245.1 <sup>bc</sup>
PB 5/51	36.3 <sup>cd</sup>	197.7 <sup>c</sup>
Tjir I	35.7 <sup>cd</sup>	202.6 <sup>e</sup>
GTI	46.9 <sup>bc</sup>	263.2 <sup>ab</sup>
RRII 38	25.7 <sup>d</sup>	137.1 <sup>f</sup>
RRII 33	15.6 <sup>d</sup>	144.6 <sup>f</sup>

Means followed by a common letter are not significantly different at  $p < 0.05$ . Each value is the mean of 9 plants per clone x three months.

Positive significant correlation between latex yield and [ATP] (September)



Positive significant correlation between latex yield and [ATP] (October)



Positive significant correlation between latex yield and [ATP] (November)

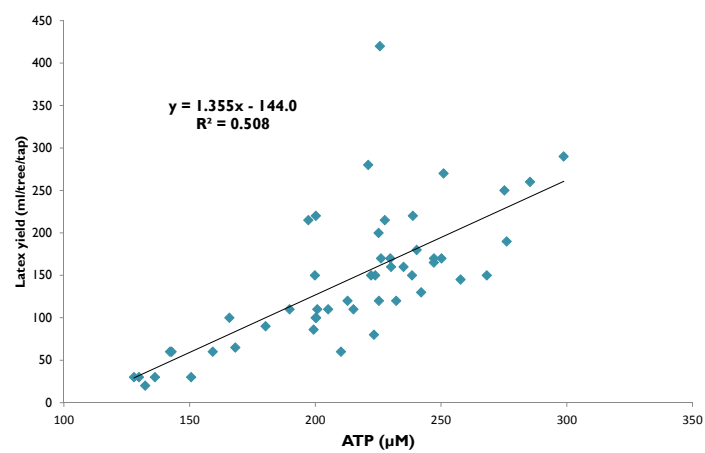


Table-3 Latex ATP ( $\mu\text{M}$ ) in immature plants of ten *Hevea* clones of known yield potential

Clones	Yield class	ATP (2 <sup>nd</sup> yr)	ATP (3 <sup>rd</sup> yr)	ATP (4 <sup>th</sup> yr)	ATP (5 <sup>th</sup> yr)
RRII 105	High	339.2 <sup>a</sup>	334.8 <sup>a</sup>	294.7 <sup>a</sup>	299.5 <sup>a</sup>
RRIM 600	High	306.2 <sup>ab</sup>	304.2 <sup>ab</sup>	236.0 <sup>bcd</sup>	249.8 <sup>b</sup>
PB 217	High	253.7 <sup>bc</sup>	245.7 <sup>bc</sup>	204.2 <sup>cd</sup>	207.9 <sup>c</sup>
PB 235	High	301.6 <sup>ab</sup>	286.6 <sup>ab</sup>	257.4 <sup>ab</sup>	239.6 <sup>b</sup>
PB 260	High	284.5 <sup>ab</sup>	282.8 <sup>ab</sup>	222.3 <sup>bcd</sup>	228.4 <sup>bc</sup>
PB 5/51	Medium	197.9 <sup>cd</sup>	208.2 <sup>cd</sup>	194.9 <sup>d</sup>	199.5 <sup>c</sup>
Tjir I	Medium	277.9 <sup>ab</sup>	244.4 <sup>bc</sup>	204.1 <sup>cd</sup>	207.9 <sup>c</sup>
GTI	Medium	319.9 <sup>a</sup>	304.6 <sup>ab</sup>	247.9 <sup>bc</sup>	236.7 <sup>b</sup>
RRII 38	Low	192.3 <sup>d</sup>	174.8 <sup>d</sup>	157.1 <sup>c</sup>	136.1 <sup>d</sup>
RRII 33	Low	180.3 <sup>d</sup>	155.2 <sup>d</sup>	148.2 <sup>c</sup>	142.2 <sup>d</sup>


Means followed by a common letter are not significantly different at  $p < 0.05$   
 Each value is the mean of 15 plants per clone x three months (Sep, Oct and Nov).

Table-4 Correlation between latex ATP in immature plants and mature yield over five years

	ATP Two yr old plant	ATP Three yr old plant	ATP Four yr old plant	ATP Five yr old plant
Mature tree yield	0.70**	0.53**	0.78**	0.79**

\*\* correlation is significant at  $p < 0.01$  (2-tailed)





### Conclusions

- ❖ The direct relationship observed between latex yield and [ATP] suggests the direct role of ATP in controlling rubber biosynthesis.
- ❖ Low yielding clones always showed lower concentrations of ATP, both at immature and mature phases.
- ❖ **Latex [ATP] in young seedlings is a good indicator of yield potential in the mature phase (with a high coefficient of determination).**



### Practical utility

- Measurement of ATP is a simple, reliable and low cost technique and can be done in a few drops of latex.
- ATP finger-printing is being used to screen potentially high yielding clones by RRII.

