

POSSIBLE APPLICATION OF THE RATIO OF HMG CoA TO MEVALONATE IN THE BARK OF HEVEA BRASILIENSIS AS AN INDICATOR OF RUBBER YIELD

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ABSTRACT

Reports on the biosynthetic rates of rubber regeneration in the bark during the interval between successive tappings are scanty. It has been demonstrated that HMG-CoA reductase has a direct role in regulating rubber biosynthesis. The ratio of HMG CoA to mevalonate is a possible indication of the activity of HMG-CoA reductase which converts the former to the latter. The ratio has been estimated in the bark samples of four clones with contrasting yield characteristics. The possibility of considering this characteristic as one of the components to be used for identification of high and low yielding clones is suggested.

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Introduction

The two primary limiting factors in latex production in *Hevea brasiliensis* are flow characteristics which govern the quantity of latex obtained on tapping, and the *in situ* regeneration of latex between successive tappings (1). Reports on the biosynthetic rate of rubber regeneration in the bark during the interval between successive tappings are scanty. Investigation of the activities of the enzymes involved in rubber biosynthesis from acetate shows that HMG-CoA reductase (EC1.1.1.34) activity, which is responsible for the formation of mevalonic acid, is much lower and that this enzyme may be a limiting factor in rubber biosynthesis. The significance of low HMG-CoA reductase activity in rubber latex is not clear (2), but a correlation between enzyme activity in latex and dry rubber content was reported (2).

Methodology

In the present study an indirect method for estimating the HMG-CoA reductase activity in the bark is suggested. 3 Hydroxy-3 methyl glutaryl CoA and mevalonate concentrations in the tissue homogenate are estimated by colorimetry and the ratio between the two is taken as an index of activity of the enzyme which catalyzes the conversion of the former to the latter, a lower ratio indicating high enzyme activity and vice versa (3). The ratio, 3 hydroxy-3 methyl glutaryl-CoA/mevalonate was estimated in the bark tissue of four clones belonging to the high yielding and low yielding groups in an attempt to find any possible relationship between the ratio and the yield of rubber.

Bark samples were collected from clones RR11 105, PB-235 (high yielding), Ch4 and Pil B84 (low yielding) from a completely randomized planting in the germplasm garden. There were six replicates per clone. Bark samples were taken from the drainage area in the vicinity of the tapping cut. The samples were collected in polythene bags in ice on the day of tapping, 30 to 60 minutes after complete cessation of latex flow. Soft tissues of the bark were homogenized in 1 percent saline arsenate and diluted with an equal volume of perchloric acid (50ml/litre).

The extract was centrifuged at 3000 rev / min for 15 min. One ml of the filtrate was treated with 0.5ml of 2M freshly prepared hydroxylamine in water. In the case of HMG-CoA, hydroxylamine reagent mixed with an equal volume of 4.5M sodium hydroxide solution was used whereas in the case of mevalonate, hydroxylamine reagent mixed with equal volume of water was used. The solutions were mixed and after 5 min, 1.5ml of ferric chloride was added. After 10 min the colour was read at 540nm against a similarly treated blank. Correlations were worked out between HMG-CoA/Mevalonate ratio, rubber yield and drc. The results indicate that there is significant variation in the ratios (HMG CoA/mevalonate) in high and low yielding clones. Maximum activity was observed in high yielding clones PB 235 and RRII 105 (Figure 1). HMG-CoA/mevalonate ratios were found to be negatively correlated with dry rubber yield ($r = -0.778^*$), (Figure 2), and drc ($r = -0.47^*$).

Discussion and conclusions

The ratio method does not indicate the actual quantity of enzyme present in the tissue. However it gives a reasonable indication of the rate of activity of HMG CoA to mevalonate. Since the enzyme is associated with the endoplasmic reticulum of the latex vessel it is unlikely to be completely represented in the free polysomes found in latex (4). Hence the measurement of enzyme activity in the soft bark tissue which is the truly productive tissue might give a true representation of the *in vivo* situation. The method is simple and rapid and can be utilised as a valuable tool to differentiate between high and low yielding clones.

References

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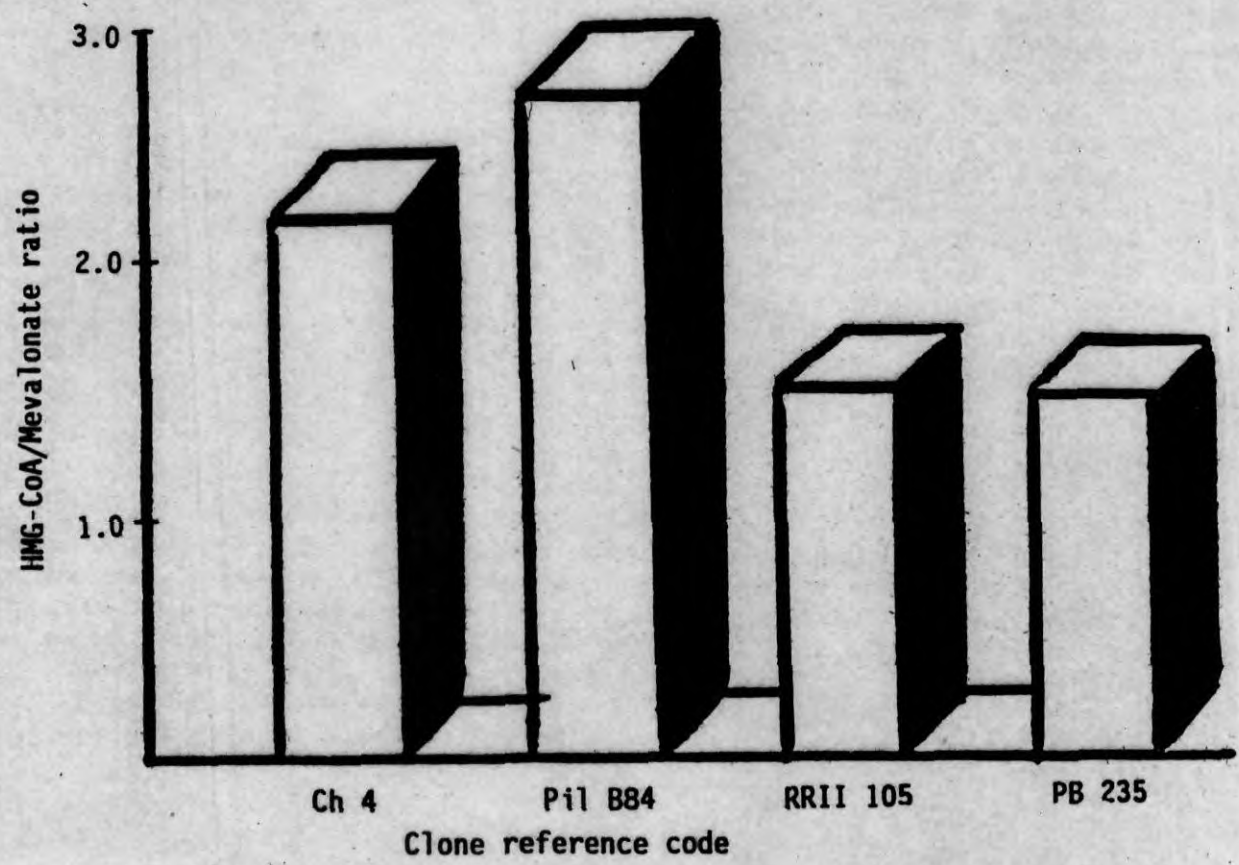


Figure 1: Ratio of HMG-CoA to Mevalonate in the bark of four clones

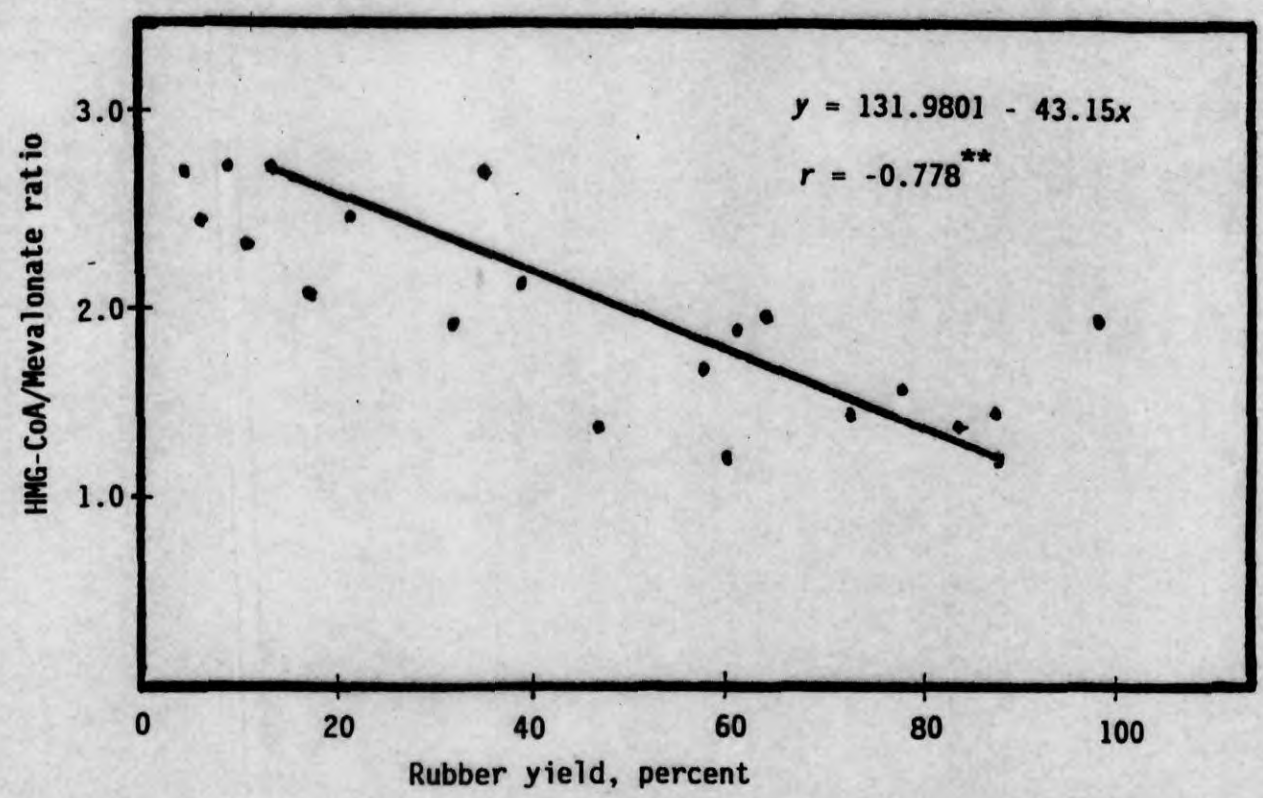


Figure 2: Correlation between HMG-CoA / Mevalonate ratio and rubber yield