TPD SYNDROME INCREASES BARK RESPIRATION IN HEVEA

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A study was conducted in *Hevea* clones (GT-1 and RRII 105) to examine the respiratory rates of the bark tissues from tapping panel dryness (TPD) affected and normal trees and relate to the carbohydrate status of the tissues. Concomitant with an increase in total sugars, sucrose and starch contents in the bark, respiration rate also increased in TPD affected once when compared to healthy bark from normal trees. Bark tissues from the opposite side of the tapping panel of the two clones showed a decrease in the respiration; however, they did not show much variation in their carbohydrate contents in healthy trees. In partially affected GT-1, sucrose and total sugar contents of the bark tissues from the latex yielding (wet) portion of the tapping panel showed a significant decrease compared to the dry portion of the same bark. Carbohydrate content and respiration of wet and dry portions of the panel from TPD trees did not vary in RRII 105 as well. It would appear that increased availability of carbohydrates and increased respiration in the bark must have led to increased biomass production at the cost of rubber biosynthesis in TPD affected *Hevea* trees.

INTRODUCTION

Tapping panel dryness (TPD) is a physiological disorder. When the level of exploitation exceeds the physiological capacity of the tree to generate latex, the rubber tree succumbs to TPD (Chrestin, 1989). In the early stages of TPD syndrome, the tapping panel is dry only at certain places; but in advanced stages dry area increased gradually and led to a total inhibition of rubber biosynthesis. Incidence of TPD increases with high tapping frequency and/or excessive yield stimulation (Commere et al., 1989). A number of studies such as cytological disorder associated with TPD, viruses and viroids and biochemical changes with respect to TPD have been made in recent years (Lim, 1973;

Gomez, 1990; Dian et al., 1995; Krishnakumar et al., 1997). Chrestin (1985) proposed a biochemical explanation involving laticiferous senescence through activation of oxidative stress leading to dysfunction of the laticiferous metabolism. High peroxidase activity and the accumulation of phenols in the TPD affected bark supports this contention (Krishnakumar et al., 1997). In spite of several investigations, the exact causes of TPD syndrome are not clear.

Rubber biosynthesis in TPD affected bark was seriously inhibited although there was adequate supply of carbohydrate for polyisoprene synthesis (Krishnakumar et al., 1997). The conversion of mevalonate into polyisoprene is

an extremely energy consuming process (Jacob and Prevot, 1992) and hence energy metabolism may be a limiting factor for rubber biosynthesis in TPD affected bark tissues. There are no studies reported on bark respiration and energy metabolism in *Hevea*. The present study is aimed at measuring the respiratory rates and carbohydrate composition of bark tissues from TPD affected and healthy trees of two high yielding *Hevea* clones.

MATERIALS AND METHODS

Rubber clones of seventeen year old GT1 and a nineteen year old RRII 105 from the Central Experimental Station of Rubber Research Institute of India (RRII) at Chethackal (90 22' N; 76 o 50' E) were selected for this study. All agronomic practices and plant protection measures were undertaken according to RRII recommendations. These trees were under regular tapping, S/2 d/2 through out the year from the past two years. TPD affected (n=6) and normal (n=7) trees were identified by tapping observations during a two months period prior to bark sample collection. Bark tissues from the tapping panel and the opposite side of the tapping panel was taken from each normal tree. Bark samples were taken from the wet and dry portion of the tapping of the TPD affected trees along with samples from the opposite side of the tapping panel. Samples were taken from the tapping panel at the time of tapping and transported to the laboratory on ice. Soft bark tissues were isolated and used for measurement of respiration while the remaining portions were stored at -60°C for biochemical analysis.

Tissue respiration was measured polarographically using a Clark type oxygen electrode (Hanzatech, UK) as described by Lambers et al., (1983) with modifications. Soft bark tissue (approximately 1 mm thick and 200 mg fresh weight) immersed in two ml of reaction buffer (0.3 M sorbitol, 10 mM NaCl, 10 mM KH₂ PO₄, 2 mM MgSO₄ and 0.1 % BSA with a pH 7.2) in the electrode chamber. Oxygen depletion

rate in the assay buffer was recorded for five minutes at 25°C. Tissues were dried at 70°C and weighed; dry weight of the samples used for calculation and the rate of respiration was presented (nM O₂ g-1 Min⁻¹ dry weight). Analyses were repeated three times in each tissue samples. One hundred-mg (fresh weight) of soft bark tissue was powdered in liquid nitrogen and extracted with 80 per cent ethanol and used for the estimation of total sugars, sucrose and starch.

RESULTS AND DISCUSSION

Earlier Krishnakumar et al. (1997) reported less respiratory activity in TPD affected rubber bark because of complete inhibition of rubber biosynthesis. Contrary to the earlier findings, bark respiration was higher in TPD affected ones than in the healthy rubber trees of both the clones (Fig. 1a,b). Both the clones showed a significant reduction in the respiratory rates of the bark in the opposite side of the tapping panel as compared to the tapping panel in the healthy and TPD affected trees. Wound induced stimulation in respiration may explain this observation (Roberts et al., 1985).

TPD affected trees had relatively more sucrose than healthy trees in their bark (Fig. 2a, b). Similar trend was observed in the total sugar concentration as well (Fig. 3a, b). This is in conformity with earlier findings of Krishnakumar et al. (1997). In both the clones, there existed a significant positive correlation between respiratory rates and total sugar contents (Fig. 5). Concentration of sucrose was more in the untapped bark than in the tapped bark of healthy rubber trees: This may be because of more drain of sugars from the surrounding places of tapping panel through latex. Starch content remained the same in the tapping panel and in the opposite side of the tapping panel of RRII 105, but it was significantly higher in the latter than in the former in GT1. Similar trend was observed in TPD affected trees of RRII 105 and GT-1. When a healthy tree was compared with a TPD affected one, the starch content increased in the case of the latter (Fig. 4a, b).

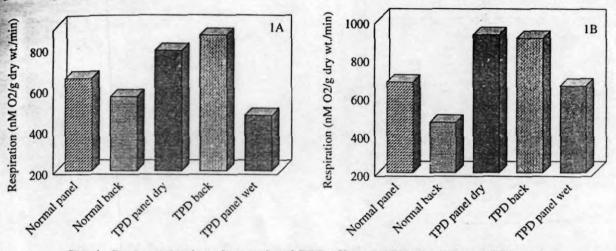


Fig. 1. Bark respiration of normal and TPD affected RRII 105 (1a) and GT1 (1b)

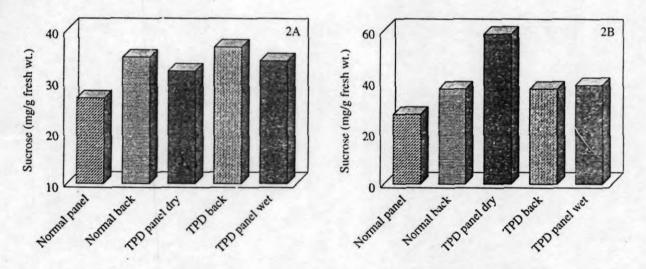
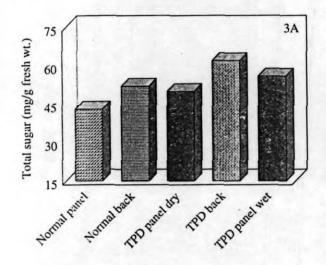


Fig. 2. Sucrose content of normal and TPD affected RRII 105 (2a) and GT1 (2b)

There was relatively more starch per unit sucrose in the untapped areas of both TPD affected and healthy trees of GT1. The ratio was more or less same in the tapped and untapped portions of TPD affected and normal trees in RRII 105. There was relatively more tissue respiration per unit total sugar in the tapped areas than in the untapped areas of normal trees of both the clones. Generally, low respiration per unit sugars was noticed in the dry portion of the TPD trees than in the normal tapping panel. Increased respiratory activity per unit sugar content seem to be related to the rubber biosynthesis capacity of the bark as evident from the results from healthy and dry

trees. Increased carbohydrate content in the untapped bark was related to the decreased bark respiration.

Increased tissue respiration noticed in the wet and dry areas of TPD affected trees that were rather unexpected. Altered activity of enzymes such as polyphenol oxidase, peroxidase, etc., (Krishnakumar et al., 1997) created some artifacts in the polarographic measurement of respiration, although the extent of such interaction may not explain the over all increase in respiration observed in the TPD affected bark. Alternatively, cyanide resistant respiration would have been



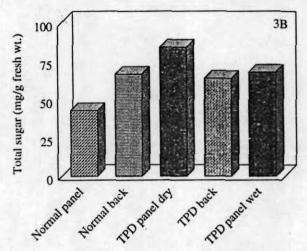
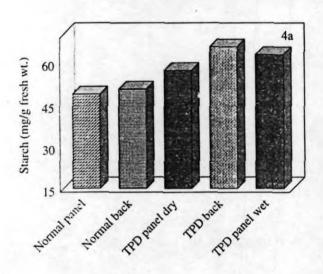


Fig. 3. Total sugar content of normal and TPD affected RRII 105 (3a) and GT1 (3b)



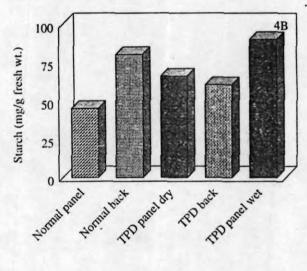


Fig. 4. Starch content of normal and TPD affected RRII 105 (4a) and GT1 (4b)

operated in the TPD affected bark, which led to more respiration with less ATP production. In this context, it may be noted that the TPD affected trees induced excessive biomass in the form of tumors and other secondary growth in the affected region. It appears that the enhanced respiratory activity contributed to biomass production at the cost of rubber biosynthesis in the TPD affected rubber trees. If that is the case, the reason for diversion of energy for biomass production instead of rubber biosynthesis is worth investigating.

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REFERENCES

Chrestin, H. 1985. Etudes des systemes generateurs d'oxygene toxique et des mecanismes detoxifiants. pp.30-32. In: Reuntion encoche seche du 1985, Abidjan, Cote d'Ivoire.

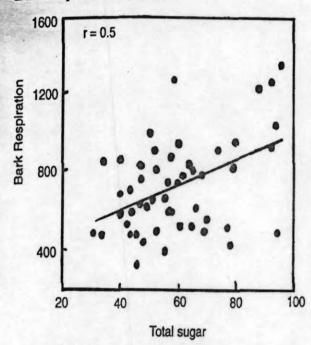


Fig. 5. Relationship between the bark respiration and the total sugars

Chrestin, H. 1989. Biochemical aspects of bark dryness induced by over stimulation of rubber tree with Ethrel. pp.431-441. In: *Physiology of Rubber Tree Latex*. Eds. d' Auzac, J., Jacob, J.L. and Chrestin, H. CRC Press, Boca Raton.

Commere, J., Eschbach, J.M. and Serres, E. 1989.
Tapping panel dryness in Cote d' Ivoire.
pp.83-98. In: Proceedings of IRRDB
Workshop on Tree Dryness, Penang,
Malaysia.

Dian, K., Sangare, A. and Diopoh, J.K. 1995. Evidence for specific variation of protein pattern during tapping panel dryness conditions development in Hevea brasiliensis. Plant Science 105: 207-216.

Gomez, J. B. 1990. Lutoids of *Hevea brasiliensis*: morphological consideration. *Journal of Natural Rubber Research* 5: 231-240.

Jacob, J.L. and Prevot, J. C. 1992. Metabolism of the laticiferous system and its biochemical regulation. pp.116-136. In: Natural Rubber: Biology, Cultivation and Technology. Eds. Sethuraj, M.R. and Mathew, N.M. Elesvier, New York.

Krishnakumar, R. Sreelatha, S., Molly Thomas, Gopalakrishanan, J., James Jacob and Sethuraj, M.R. 1997. TPD alter the biochemical composition in the soft bark tissue. Indian Journal of Natural Rubber Research (Communicated).

Lambers, H., Day, D.A. and Azcon-Bieto, J. 1983. Cyanide - resistant respiration in roots and leaves, Measurement with intact tissues and isolated mitochondria. *Physiologia Plantarum* 58:148-154.

Lim, W.C. 1973. Changes in bacteria free filtrate of *Hevea* latex C-serum from particularly dry tree. *Journal of Rubber Research Institute of Malaysia* 23: 351-355.

Roberts, JA, Tucker, C.A. and Maenads, M.J. 1985. Ethylene and foliar senescence. pp.267-275. In: Ethylene and Plant Development. Eds. Roberts, J.A. and Tucker, C.A. Butter Worths, London.