

## STAINING PROCEDURE FOR LATICIFEROUS SYSTEM OF *HEVEA BRASILIENSIS* USING OIL RED O

Philipose Omman and C.P. Reghu

Omman, P. and Reghu, C.P. (2003). Staining procedure for laticiferous system of *Hevea brasiliensis* using Oil red O. *Indian Journal of Natural Rubber Research*, 16 (1 & 2) : 41-44.

A staining procedure using 1 per cent Oil red O prepared in 90 per cent isopropyl alcohol was tested for its ability to stain laticifers in *H. brasiliensis*. Laticifers were stained a deep-red maroon with Oil red O, whereas a red colour was developed with Sudan III and Sudan IV. Oil red O took the least time for stain preparation and gave maximum stainability when compared to Sudan III/IV preparations. It was found to be a suitable stain for laticifers of *Hevea brasiliensis*.

**Key words:** *Hevea brasiliensis*, Laticifers, Oil red O, Staining procedure

C.P. Reghu (for correspondence), Rubber Research Institute of India, Kottayam - 686 009, Kerala, India (E-mail: cpreghu@rubberboard.org.in); Philipose Omman, Catholicate College, Pathanamthitta - 689 645, Kerala, India (E-mail: philiposeomman@sancharnet.in).

### INTRODUCTION

Sudan III and IV are non-polar lipid stains (Pearse, 1968; Wigglesworth, 1988) widely used for the histochemical staining of laticiferous tissues in *Hevea brasiliensis* (Gomez *et al.*, 1972; Panikkar, 1974; Abraham *et al.*, 1992; Premakumari *et al.*, 1992; Reghu *et al.*, 1996). Staining methods reported earlier for laticiferous tissue of *H. brasiliensis* are given in Table 1. Hamzah *et al.* (1988) compared conventional staining of bark samples of *H. brasiliensis* using Sudan III, Sudan blue and Oil red O and discussed the colouration

of stained laticifers. But the staining protocol was time consuming. The solvents usually used for Sudan stains are ethyl alcohol, acetic acid and glycerol combinations, which take a minimum period of 48 h for preparation. Moreover, overnight incubation of tissue sections in the staining medium is required for adequate staining of laticiferous system. The formation of precipitate during the preparation of Sudan stain is another drawback, which in turn adversely affects the clarity of stained sections.

Oil red O is a hydrophobic, non-polar,

Table 1. Staining methods for latex vessels of *H. brasiliensis*

Stain	Solvent used	Embedding medium	Duration of staining	Colour developed	Reference
Aqueous safranin and malachite green	Water	Araldite-polybed	2h to overnight	Latex vessel wall carpo blue; latex cyclamen purple	Hamzah <i>et al.</i> (1988)
Sudan III & IV	Methyl alcohol (75%)	Nil Latex vessel rings dissected from KOH (10%) treated bark samples	10-15 min	Red	Zhao Xin Qian (1987)
Oil red 4B	Ethyl alcohol (95%)	Paraffin wax	90 min	Red	Wimalaratna (1973)