characteristic feature noted in the root primordium was the absence of root cap (figure 9).

Random vascularization in callus cultures has been reported by several workers[7,8]. Starch accumulation is reported to be essential in mesistematic tissues, leading to organogenesis[9]. However others have found no correlation between bud formation and starch accumulation[3,10]. In our studies the meristems were indistinguishable at early stages of development. Similar reports are also available in the literature[8]. Meristematic activity is found in peripheral regions or in deeper layers depending upon the crop species. In cauliflower, meristematic activity was found in deeper layers while in *Punica granatum* meristems were superficial[8,11]. Although all seed plants have root cap, it was missing in the *in vitro* formed roots. But these may degenerate under culture conditions, as reported in vanilla[12].

4 July 1988

1. Halperin, W., *Annu. Rev. Plant Physiol.*, 1969, **20**, 395.
2. Hicks, G. S., *Can. J. Bot.*, 1987, **65**, 1913.
3. Maherchandani, N., Yadav, N. R., Jain, R. K., Chowdhury, V. K. and Singh, S., *Proc. symp. plant cell and tissue culture of economically important plants*, (ed.) G. M. Reddy, Hyderabad, 1987, p. 337.
4. Gresshoff, P. M. and Doy, C. H., *Planta*, 1972, **107**, 161.
5. Johansen, D. A., *Plant microtechnique*, McGraw Hill, New York, 1940.
6. Misra, N. R., Varghese, T. M., Maherchandani, N. and Jain, R. K., *Plant cell culture in crop improvement*, Plenum Press, New York, 1983, p. 465.
7. Gayatri, M. C., *Phytomorphology*, 1978, **28**, 395.
8. Jaidka, K. and Mehra, P. N., *Can. J. Bot.*, 1986, **64**, 1644.
9. Thorpe, T. A. and Murashige, T., *Can. J. Bot.*, 1970, **48**, 277.
10. Von Arnold, S., *Ann. Bot.*, 1987, **59**, 15.
11. Singh, S. and Mathur, M., *Acta Bot. Indica*, 1987, **15**, 98.
12. Philip, V. J. and Nainar, S. A. Z., *Ann. Bot.*, 1988, **61**, 193.

VIRUS PARTICLES IN CHLOROPLAST PREPARATIONS FROM INFECTED LEAVES OF *NICOTIANA TABACUM*

G. P. RAO and K. SHUKLA
Department of Botany, University of Gorakhpur, Gorakhpur 273 009, India.

THE presence of virus particles inside the chloroplast has been proved by electron microscopy[1,2]. In the present investigation, the presence of virus particles inside the chloroplast has been proved by the following experiment.

Chloroplast was isolated as described earlier[3,4]. The cultures of tobacco mosaic virus (TMV) and potato virus X (PVX) were maintained on systemic host (*Nicotiana tabacum* cv. White Burley) by successive inoculations at intervals of 3 days, in an insect-free glass house. *Chenopodium amaranticolor* Coste et Reyn was used as local lesion host (test plant). The three types of virus inoculums were prepared as follows:

(i) Diseased leaf (with TMV and PVX) showing severe symptoms was macerated and crushed in pestle with distilled water in 1:1 (w/v) ratio (inoculum I).
(ii) Chloroplast was isolated from chlorotic areas of virus-infected leaf. This isolated chloroplast in 0.2 M NaCl solution served as inoculum II.
(iii) Isolated chloroplast (in 0.2 M NaCl solution) from green areas of the same virus-infected leaf served as inoculum III.

For chloroplast isolation, two types of tissues were taken from each of the same virus-infected (TMV and PVX) leaf of *N. tabacum*, i.e. the green and chlorotic tissues. The chlorotic and green patches were cut separately from virus-infected leaves with the cork borer. Fifty g of leaf tissues was taken for chloroplast isolation. After isolation, the chloroplast was kept in 0.2 M NaCl solution at 4°C for 6 h and this was treated as inoculums II and III.

The inoculation was done by rubbing the inoculum gently (I, II and III) on the upper surface of leaves of test plant with 600 mesh carborundum powder. All the inoculums were inoculated separately on the leaves of test plants of the same age and size. Five plants were taken for each experiment. Local lesions were counted after seven days of treatment to compare the potency in infection of each inoculum. All the experiments were repeated thrice and the average value recorded.

Table 1 *Number of local lesions produced by various inoculums*

Source of inoculum	Average no. of local lesions*	
	TMV	PVX
Virus-infected leaf (inoculum I)	142	98
Isolated chloroplast of chlorotic areas of virus-infected leaf (inoculum II)	50	36
Isolated chloroplast from green area of virus-infected leaf (inoculum III)	12	8

*Local lesions on 20 leaves of *C. amaranticolor*.

Table 1 indicates that the inoculum prepared by chloroplast isolated from the chlorotic tissues (inoculum II) caused similar type of local lesions as caused by inoculum I and also the number of local lesions was found more or less the same. But the inoculum of chloroplast from non-chlorotic areas (green area) from the same infected leaf produced fewer number of local lesions than that of chloroplast of necrotic areas of the same virus-infected leaf with both the viruses.

In the present study the inoculum prepared by isolated chloroplast of virus-infected leaf produces similar local lesions as of virus-infected leaf itself. This proves the presence of virus particles inside the chloroplast of *N. tabacum* leaves.

The authors thank CSIR, New Delhi, for financial assistance.

30 July 1988; Revised 31 October 1988

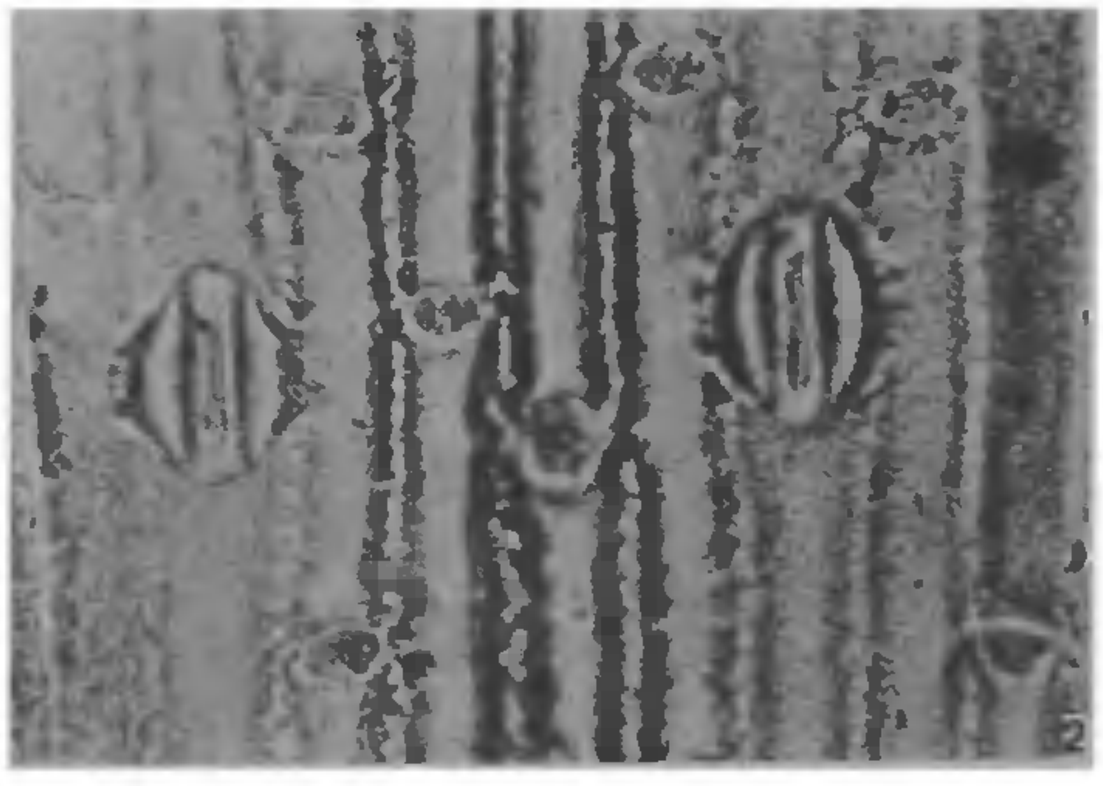
1. Hirai, A. and Wildman, S. G., *Virology*, 1969, **38**, 73.
2. Hagiladi, A., Bourque, D. P. and Wildman, S. G., *Virology*, 1975, **635**, 123.
3. Arnon, D. I., Allen, M. B. and Whatley, F. R., *Biochem. Biophys. Acta*, 1956, **205**, 449.
4. Whatley, F. R. and Arnon, D. I., *Methods Enzymol.*, 1962, **6**, 308.

A RAPID TECHNIQUE FOR OBTAINING LEAF PRINTS FOR STOMATAL COUNT WITH FEVICOL

K. A. NAYEEM and D. G. DALVI
Wheat Research Unit, Marathwada Agricultural University, Parbhani 431 402, India.

STOMATAL resistance to the diffusion of water vapour or carbon dioxide from leaf surface can be regulated by stomatal frequency, stomatal size and opening closing of the stomata. Genetically, it is possible to control stomatal resistance by selecting for suitable stomatal size and frequency[1,2]. The techniques for stomatal examination described earlier[3–6] are time-consuming, and often the method alters the impression of leaf surface and prints are disturbed.

Fevicol (Pidilite Industries Pvt. Ltd, Bombay) is a commonly used synthetic adhesive. It is much easier



Figures 1 and 2. **1.** Application of Fevicol over leaf surface using a needle; **2.** Open and closed stomata of *Triticum dicoccum*.