

organogenesis, some of these calli, were subcultured to media containing low concentration of KN (0.01–0.5 ppm) and high concentration of NAA (1–3 ppm). Some of these calli produced roots (figure 1D). The roots were about 5 mm long and white. The calli with or without roots were transferred to media supplemented with KN and either BAP or IAA at different concentrations to produce shoots. The calli remained healthy on these media, however, shoot initiation did not occur except in one culture where small shoot development was observed. The duplication of such an experiment was tried but in vain. Addition of growth adjuvants such as CM, yeast extract or casein hydrolysate did not prove successful.

Hypocotyl calli when cultured on a high concentration of cytokinin (KN 1 or 2 ppm) along with NAA (0.1 to 2 ppm) did grow and produced new mass of cells. No organogenesis occurred in these calli. Kinetin alone at different concentrations also produced the same result.

Hypocotyl calli when transferred to liquid MS medium supplemented with 1 ppm each of KN and 2,4-D, produced suspension cultures. These cultures were placed on a shaker. Microscopic examination of the cultures revealed single cell clusters and embryogenic structures of various sizes and shapes (figure 1E–H). When these embryogenic structures were subcultured on solid media or retained on the liquid medium, they merely callused. Further work is needed to achieve dicotyledonous embryoids or young plantlets from these embryogenic structures.

Tissue culture through hypocotyl could be raised successfully and organ regeneration in the form of roots could be manipulated. Further work is needed to induce shoot regeneration on the rooted calli and development of the embryogenic structures from the suspension cultures. If these could be achieved, this method can be used in plant propagation of *Sapium sebiferum*.

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1. Murashige, T., *Ann. Rev. Plant Physiol.*, 1974, 25, 135.
2. Sommer, H. E. and Brown, C. L., In *Plant cell and tissue culture—Principles and application*, (eds) W. R. Sharp, P. O. Larsen, E. F. Paddock and V. Raghavan, Ohio State University Press, Columbus, Ohio, 1979, p. 461.

3. Goncalves, A. N., Machado, M. A., Caldas, L. S., Sharp, W. R. and Amaral Mello, H. D., In *Plant cell and tissue culture—Principles and application*, (eds) W. R. Sharp, P. O. Larsen, E. F. Paddock and V. Raghavan, Ohio State University Press, Columbus, Ohio, 1979, p. 509.
4. Bapat, V. A. and Rao, P. S., *Ann. Bot.*, 1979, 44, 629.
5. Gupta, P. K., Nadgir, A. L., Mascarenhas, A. F. and Jagannathan, V., *Plant Sci. Lett.*, 1980, 17, 259.
6. Venverloo, C. J., *Acta Bot. Neerl.*, 1973, 22, 390.
7. Winton, L., *Forest Sci.*, 1971, 17, 348.
8. Srinivasa Rao, N. K., Narayanaswami, S., Chacko, E. K. and Doreswamy, R., *Curr. Sci.*, 1981, 50, 310.
9. Murashige, T. and Skoog, F., *Physiol. Plant.*, 1962, 15, 437.

WITCHES BROOM DISEASE OF BUTTERFLY FLOWER

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A WITCHES' broom disease is of common occurrence on butterfly flower (*Schizanthus wisetonensis*) in Darjeeling and Sikkim Hills. The disease was first observed in 1976 at the Regional Station of Indian Agricultural Research Institute, Kalimpong. The incidence of the disease ranged from 1–15% in various nurseries. The affected plants were characterised with pronounced stunting, short internodes and reduced laminae. Such plants developed profuse axillary branches thus giving the appearance of witches broom. Occasionally, the affected plants developed violet colour in their leaves and stems. Flowers on these plants were phylloid. The results of transmission and antibiotic therapy are presented in this paper.

The disease was transmitted by grafting and through four types of leaf-hoppers (unidentified) but not by sap inoculations. Butterfly flower plants developed typical symptoms of the disease between 20–50 days after graft inoculation. In the host range studies, larkspur and straw flower were susceptible but *Chenopodium amaranticolor*, *Nicotiana tabacum* v. *xanthi*, potato, brinjal, petunia, tomato, sunnhemp, bottlegourd, sesame and *Sechium edule* failed to take infection. Transmission results were confirmed by back inoculation.

To control the disease, 500 and 1000 ppm of terramycin (oxytetracycline hydrochloride, 20% active ingredient) was sprayed on inoculated plants in the field and glasshouse. It was observed that 1000 ppm sprays were comparatively more effective in that 40% of the treated plants recovered as against 35% recovery with 500 ppm. These results were confirmed by repeated trials. However, it was not clear whether the recovery was permanent because butterfly flower is an annual and short-duration plant.

A virus disease of *Schizanthus* sp. has been described to be caused by tobacco leaf curl virus¹ which is transmissible to tobacco and tomato. The disease is distinct from witches' broom disease now reported due to non-transmission to these hosts. The symptoms, transmission and recovery by antibiotic therapy are suggestive of the association of mycoplasma-like organisms (MLOs) with butterfly flower witches' broom disease²⁻⁵ studies to confirm the MLOs association by electron microscopy are in progress. The pathogen of this disease has a restricted host range and does not infect hosts of commonly occurring yellow diseases in India⁶⁻¹⁰ and thus forms a new record.

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1. Garg, R. P. *Curr. Sci.*, 1949, **18**, 260.
2. Davis, R.E. and Whitcomb, R.F. *Annu. Rev. Phytopathol.*, 1971, **9**, 119.
3. Doi, Y., Taranaka, M., Yora, K. and Asuyama, H., *Ann. Phytopath. Soc. Jpn.*, 1967, **33**, 259.
4. Ishie, T., Doi, Y., Yora, K. and Asuyama, H., *Ann. Phytopath. Soc. Jpn.*, 1967, **33**, 267.
5. Maramorosch, K., Grandos, R. R. and Hirumi, H., *Adv. Vir. Res.*, 1970, **16**, 135.
6. Ahlawat, Y. S. and Kulshreshtha, U. V., *Indian Phytopathol.*, 1977, **30**, 268.
7. Paul Khurana, S. M., Arai, K., Nagaich, B.B., Vijay Singh and Shatrughna Singh, *Indian Phytopathol.*, 1981, **34**, 532.
8. Varma, A., Asha Sang, Ghosh, S. K., Raychaudhuri, S. P., Chenulu, V.V. and Nam Prakash *Curr. Sci.*, 1974, **43**, 349.
9. Sahambi, H. S. *Plant Disease Problem.*, 1970, p. 340.
10. Varma, A., Chenulu, V. V., Raychaudhuri, S. P., Nam Prakash and Rao, P. S., *Indian Phytopathol.*, 1969, **22**, 289.

PEROXIDASE ACTIVITY AND ITS ISOZYMES IN RELATION TO FLOWER SEX-EXPRESSION IN *RICINUS COMMUNIS* L.

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THE distribution of peroxidase activity and/or its isozymes has been shown to be highly correlative with various plant processes^{1,2}. Recently the role of peroxidase activity and its isozymes have been implicated in flower sex-expression of some plants^{3,4}. Nevertheless, there is no such report in monoecious systems where both staminate and pistillate flowers appear on the same inflorescence but do not have any rudimentary structures of opposite sex. The findings presented here describe the possible role of peroxidase enzyme in sex-expression of *R. communis* where rudimentary sex organs are not found.

Plants of *R. communis* were raised in earthen pots with properly-manured soil. Vegetatively growing apical meristems and male and female flower buds were used for analysis. The peroxidase enzyme was extracted with phosphate buffer (pH 7) at 0°C with 1% sodium ascorbate. The macerates were centrifuged at 16000 g for 20 min at 0°C and supernatants were collected for analysis. The enzyme activity was measured by using H₂O₂-guaiacol reaction mixture as described earlier³.

To separate peroxidase isozymes, disc gel electrophoresis was employed using 10% polyacrylamide gels and applying 50 volt potential and 0.5 mA current per tube till the dye front reached to the bottom of the gel. The reaction mixture used for enzyme assay served as staining agent to localize the bands on the acrylamide gels.

No significant difference was observed in the specific peroxidase activity of vegetatively growing apical meristems (3.1 ± 0.6 units mg⁻¹ protein) and in male flower buds (3.3 ± 0.5 units mg⁻¹ protein) whereas activity on per gram fresh weight basis was high in the apical meristems (1.22 ± 0.4 units) than the male flower buds (0.95 ± 0.2 units). If the specific activity recorded in male flower buds was compared with the female flower buds (2.3 ± 0.7 units mg⁻¹ protein), the former had higher activity than the later whereas no significant difference was observed in the activity taken on per gram fresh weight basis in both male and female (0.8 ± 0.2 units) flower buds. However, no activity was recorded after a few hours of extraction