

(CC, central cavity; CH, chalazal haustorium; EMB, proembryo; END, endosperm; HY, hypostase; II, inner integument; MH, micropylar haustorium; NU, nucellus; OI, outer integument; SC, seed coat).

rates. However, its remnants are noticeable even at the older stages of the seed. Thus, the endosperm of *Scleria foliosa* differentiates into micropylar haustorium, endosperm proper and chalazal haustorium. As the development proceeds, some of the cells in the central core of the endosperm proper disintegrate as they fail to keep pace with the rapidly expanding peripheral endosperm tissue and results in the formation of a central cavity (Fig. 2) which persists even at maturity.

We are indebted to Professor M. Nagaraj for providing facilities and encouragement, and to Dr. K. Subramanyam and Professor D. A. Govindappa for valuable suggestions. The award of a Junior Research Fellowship by the U.G.C. to one of us (N. D.) is gratefully acknowledged.

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December 6, 1978.

#### SOLUBILITY DIFFERENCES OF POLLINIAL AND POLLEN WALLS OF ASCLEPIADACEAE AND THEIR SIGNIFICANCE

THE chief constituent of pollen exine is sporopollenin—a polymer of carotenoids resistant to biological and chemical degradation<sup>1</sup>. Sporopollenin is also reported to be a major constituent of the sac-like pollinial walls that enclose the pollen grains of *Calotropis*<sup>2,3</sup> and *Pergularia*<sup>4</sup> (Asclepiadaceae). The similarity in the chemical constitution of pollen and pollinial walls is demonstrated by their acetolysis resistance and solubility in the sporopollenin specific solvent of fused KOH as well as by various cytochemical tests.

Although pollen exines are basically composed of sporopollenin, the ectexine (exine-I) and the endexine (exine-II) respond differently to stains<sup>5</sup>. More significantly, solvents like 2-aminoethanol dissolve exine-I while exine-II remains insoluble. This preferential solubility suggests the chemical dissimilarity of the sporopollenin of exine-I and II of pollen walls. Would such zonal differences in solubility exist in the predominantly sporopollenin containing pollinial walls of Asclepiads? This possibility is examined and the results are presented in this report.

Pollinia of the following genera of Asclepiadaceae were tested: *Asclepias*, *Calotropis*, *Ceropegia*, *Daemia*, *Hoya* and *Tylophora*. The controls used were the

pollen grains of 5 non-asclepiadaceous genera (*Cosmos*, *Tridax*, *Zinnia*, *Mussaenda* and *Paspalum*). Both pollinia and pollen grains were treated in cold and hot 2-aminoethanol and in solvent systems of sodium chlorite + acetic acid and ammonium hydroxide + hydrogen peroxide. These reagents have already been tested on pollen walls and are reported to dissolve, degrade or swell exine-I without any apparent action on exine-II<sup>6</sup>.

Cold 2-aminoethanol has only slight reaction on pollen and pollinial walls. Within 2 min of contact with the solvent, the walls take a pale yellow colour and by 10 min the pollen grains appear to be slightly swollen with the intine bulging out through the germ pores. As for the pollinia, there is no evident swelling but the outline of the germinating region becomes wavy and more distinctive. Both pollen and pollinial walls showed no signs of disintegration even after a day in cold aminoethanol.

In contrast, hot 2-aminoethanol reacted with pollen and pollinial walls leading to partial or full dissolution. The exine-I of non-asclepiadaceous pollen grains dissolved but not their exine-II. However, the walls of Asclepiad pollinia completely dissolved without leaving any resistant layer or membrane resembling exine-II.

The process of dissolution of pollinial walls of all Asclepiads in aminoethanol at 70° C is basically similar and may be summarized as follows. Within 1 min of contact with the hot solvent, the pollinium turns pale yellow. The structural continuity of the pollinium is lost within 15 min and as dissolution progresses, wall material appears as globules. By 20 min, no trace of the pollinial wall could be recognised and the enclosed pollen grains separate. Interestingly, the walls of the freed pollen grains retain their identity and remain insoluble even after an hour in the solvent. Thus, the sporopollenin containing pollen and pollinial walls of Asclepiads show differential solubility in hot 2-aminoethanol.

Effects of other solvent reagents are equally revealing. Sodium chlorite + acetic acid system degrades exine-I resulting in loss of structural identity but leaving some residues of the wall material behind. Exine-II is impervious to these chemicals. However, in all Asclepiad genera, these solvents completely degrade the pollinial walls but not the walls of the enclosed pollen grains. Similarly, ammonium hydroxide and hydrogen peroxide, which cause the swelling of exine-I without altering the nature of exine-II, bring parallel responses in pollinial and pollen grain walls respectively of Asclepiads. In short, the action of solvent systems on pollinial walls and exine-I of pollen walls is similar.

The differential response of exine-I and II to solvents may be interpreted on the basis of the presence of



several classes of sporopollenins<sup>6</sup>, some of which, as those present in exine-I, are more susceptible to solvents like hot 2-aminoethanol. The insolubility of exine-II is suggested to be associated with its lamellar structure, but lamellar nature is also reported<sup>7</sup> for the soluble pollinal wall raising doubts about assumed correlations between structure and solubility.

Present studies with solvents like 2-aminoethanol indicate that soluble sporopollenin is present in pollinal walls while the pollen grains contained in the pollinium have walls composed mainly of insoluble sporopollenin. In this respect, the pollinal wall of *Asclepiads* is similar to the exine-I and their pollen grain wall resembles to the exine-II of angiosperm pollen walls tested as controls in this demonstration.

We thank Prof. C. A. Ninan for facilities and the UGC for a fellowship to P. S.

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1. Brooks, J. and Shaw, G., In: *Pollen: Development and Physiology* (Ed. by Heslop-Harrison, J.), Butterworths, London, 1971, p. 99.
2. Jalaja, S. and Namboodiri, A. N., *Ind. Sc. Cong. Proc., Part III*, 1975, p. 72.
3. — and —, *Experientia*, 1975, 31, 915.
4. Vijayaraghavan, M. R. and Shukla, A. K., *Ann. Bot.*, 1976, 40, 417.
5. Faegri, K. and Iversen, J., *Text Book of Pollen Analysis*, Munksgaard, Copenhagen, 1964, p. 18.
6. Southworth, D., *Amer. J. Bot.*, 1974, 61, 36.
7. Dan Dicko-Zafimahova, L., *Adansonia*, 1978, 17, 455.

## TWO NEW ADDITIONAL HOSTS OF CITRUS TRISTEZA VIRUS

*Aegle marmelos* Correa. and *Feronia limonia* (L.) Swing. are two species of hard shelled citroid fruit trees indigenous to India. In the course of testing a large number of species, hybrids, varieties of citrus and a few Rutaceous hosts against citrus tristeza virus strains, the above two species were found to be susceptible to severe strain of tristeza virus.

Five Rutaceous hosts, viz., *Aegle marmelos* Correa., *Feronia limonia* (L.) Swing., *Murraya koenigii* L., *Ruta graveolens* L. and *Evodia luytelensis* Dode. were raised under green house condition. Forty-five days old seedlings were inoculated with severe strain (S<sub>1</sub>) of tristeza virus through aphid vector [*Toxoptera citricida* (Kirk.)]. The aphids were fed for 30 minutes to the infected source for acquiring the virus and later 25-30 viruliferous aphids per plant were allowed to feed for 30 minutes more for transmission purposes.

Further, six months old seedlings were inoculated through bark patch grafting with tristeza source and observed for 15 months. The results revealed that *A. marmelos* and *F. limonia* were susceptible and showed external symptoms. The virus was easily transmitted to the hosts by grafting as well as by aphid vector.

On *A. marmelos*, the infected leaves showed cupping, vein clearing and vein corking symptoms after 85-90 days (Fig. 1). Later the leaves became leathery, crinkled and the entire leaf blade became whitish in colour. The infected plants were very much stunted with numerous axillary sprouts. The wood was thin with bumpy bark. Stem pitting which is characteristic for tristeza was not noticed even 15 months after inoculation.



FIG. 1. Vein corking symptom on *Aegle marmelos*.

Stunting was pronounced on *F. limonia*. Cupping and yellowing with marginal chlorosis was noticed in 60 days following inoculation. The leaves became leathery and dropped-off with the advancement of the age of the seedlings. Vein clearing and stem pitting was not observed on infected plants. The bark became bumpy with thin wood. Severe root decay was noticed in the infected seedlings. The other Rutaceous hosts, viz., *M. koenigii*, *R. graveolens* and *E. luytelensis* did not show any symptoms even though the bark patch was green and developed union with the hosts. However, the virus could be recovered from these hosts, when transferred back to acid lime (*Citrus aurantifolia*) which is an indicator host for tristeza virus, through aphid vector indicating thereby that the hosts are carrying the virus symptomlessly.

*A. marmelos* and *F. limonia* were found to be susceptible to tristeza virus and this forms the first report from India and elsewhere. Earlier Vasudeva *et al.*<sup>3</sup> reported transmission of citrus tristeza virus from