Bacilliform Particles Associated with Mottled Dwarf of Eggplant (Solanum melongena L.)

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Mottled dwarf, a disease of eggplant (Solanum melongena L.), recently found in Southern Italy, causes a severe stunting of the affected plants accompanied by pronounced mottling and crinkling of the leaves and generalized unfruitfulness. The disease has been transferred by grafting to several cultivars of S. melongena but all attempts to transmit its causal agent mechanically to herbaceous hosts have so far failed (Martelli & Cirulli, unpublished).

In further studies of this disorder, suspected to be caused by a virus, tissues from both naturally and experimentally infected eggplants as well as from healthy seedlings were prepared for electron microscopy. Small fragments of mesophyll tissue were fixed in formaldehyde + glutaraldehyde in cacodylate buffer 0.1 M, pH 7.2 (Karnowsky, 1965), at room temperature for 4 hr and post-fixed in 1.33 % (w/v) osmium tetroxide in s-collidine buffer, pH 7.4. After dehydration through graded ethanol dilutions with two final passages in propylene oxide, the samples were embedded in a mixture of Epon + Araldite (Mollenhauer, 1964).

Thin sections were cut with glass knives on an LKB Ultratome, placed on carbon-coated grids, stained with uranyl acetate and lead citrate and viewed with a Hitachi HU-11 B electron microscope. The microscope was calibrated with a carbon diffraction grating with 54,800 lines/in.

Most of the observed cells contained many heavily stained, elongated or circular elements obviously representing profiles of rod-shaped structures in longitudinal and transverse section respectively (Pl. 1). Evidence supporting the probable virus nature of these structures may be found in the fact that they were consistently present in both naturally and artificially infected tissues but not in the healthy ones and, as discussed below, are morphologically comparable to many recognized plant and animal viruses.

Virus particles were seldom found singly, occurring most frequently in aggregates in which they were either oriented in diverging directions or regularly arranged in a para-crystalline array (Pl. 2 c). Groups of virus particles were usually lodged between the lamellae of the nuclear envelope, which became widely separated (Pl. 1; Pl. 2 a, b), but they also occurred at a distance from the nucleus, encased in membrane-bounded cytoplasmic enclaves originating from the endoplasmic reticulum.

This localization of the virus did not seem accidental. In many sections, particles were seen with their outermost layer continuous with the inner nuclear lamella as though they were 'budding' from it (Pl. 2 a, b), thus indicating that the nuclear envelope and probably also the endoplasmic reticulum had much to do with their development. Virus aggregates were, however, always perinuclear and no evidence of the occurrence of particles inside the nucleoplasm was obtained.

As a rule, two morphologically distinct types of particles were encountered in the same cells: bacilliform with both ends rounded, and bullet-shaped with one rounded and one flat end. The former had rather constant dimensions (2210 ± 108 × 660 ± 45 Å) and were invariably detached or in the process of detaching themselves from the
Transverse section through a naturally infected mesophyll cell of *Solanum melongena* in which the nucleus (N) is surrounded by aggregates of virus particles. Arrow points to particles in membrane-bounded cytoplasmic enclaves.

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(Facing p. 318)
(a) Bacilliform particles with perinuclear localization in a naturally infected cell. The virus particles lie normal to the inner lamella (IL) of the nuclear envelope, with which they are connected in some instances (arrow).

(b) Artificially infected cell with virus particles in a perinuclear space clearly delimited by the outer (OL) and inner (IL) lamellae of the nuclear envelope. Note the bullet-shaped appearance of the particles and their relationship to the inner lamella.

(c) Transverse section through virus particles. The triple-layered nature of the outer coat and the axial channel with inner core are visible.

(d) Bacilliform and bullet-shaped particles in longitudinal section. Note the axial channel and the connexion of the outermost layer with the inner nuclear lamella (IL).

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cellular membranes, appearing free in the perinuclear spaces or in the cytoplasmic
sacs. The latter were variable (usually smaller) in size and in many instances still
connected with their basal flat end to the nuclear lamellae (Pl. 2b) or endoplasmic
reticulum. Only the bacilliform particles, therefore, may be mature virus particles.

Transverse and longitudinal sections (Pl. 2c, d) indicate that complete particles
consisted of a triple-layered outer coat (about 190 Å thick) surrounding an electron-
transparent axial channel (about 310 Å across) which, in turn, contained an electron-
dense core.

The profiles of virus particles in cross-section look strikingly similar to those of
published micrographs of lettuce necrotic yellows (Chambers, Crowley & Francki,
1965), potato yellow dwarf (Macleod, Black & Moyer, 1966) and maize mosaic
(Herold & Munz, 1967) viruses. Also their apparent architecture is consistent with the
four-layered structural model proposed for vesicular stomatitis, an animal virus
(Bradish & Kirkham, 1966), and broccoli necrotic yellows virus (Hills & Campbell,
1968).

Besides the above-listed entities, six additional plant viruses possessing bacilliform
particles have been recorded (Gomphrena virus, Plantago virus, wheat striate mosaic,
winter wheat mosaic, sowthistle yellow vein and rice transient yellows).

Even from a superficial comparison of the characteristics reported in the literature
for these viruses, differences are apparent between them and the one reported here.
These differences lie mostly in particle size and localization within the cells, mechanical
transmissibility and host range; these characters, however, are either not sufficiently
reliable or still too imperfectly known to provide a safe basis for discrimination. Until
more critical information is obtained concerning possible relationships with other
viruses, I suggest that the virus seen within the infected tissues of Solanum melongena
be regarded as a separate entity, for which I propose the name of eggplant mottled
dwarf virus.

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