

# Sorghum Stunt Mosaic

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## ABSTRACT

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Symptoms of a viruslike disease of sorghum in the Imperial Valley of California included chlorotic and necrotic mottling and streaking of leaves and severe stunting. Electron microscopy of leaf dips and sections showed bacilliform particles measuring  $68 \times 220$  nm, and transmission tests showed that the leafhopper, *Graminella sonora*, was a vector. The virus was transmitted by leafhoppers to sorghum, corn, and wheat but not mechanically transmitted to any plant tested. The name sorghum stunt mosaic is proposed for this apparently new disease.

Three rhabdoviruses that infect members of the Gramineae (3) occur in the continental United States: maize mosaic (not confirmed), oat striate mosaic, and wheat striate mosaic. None of these has been found in California (4), and none of the reported vectors for any grass rhabdovirus is known to occur in

California (R. J. Gill, *personal communication*).

Viruslike symptoms were observed on sorghum (*Sorghum vulgare* Pers. 'Double Dwarf Early Hegari') at the Imperial Valley Field Station of the University of California. Symptoms included chlorotic and necrotic mottling and streaking of leaves (Fig. 1), severe stunting, and drastically reduced seed set. Preliminary electron microscopy showed a rhabdovirus

in infected tissue (5).

We propose the name sorghum stunt mosaic for this apparently new disease. Attempts to identify the rhabdovirus causing sorghum stunt mosaic and to determine its vector are reported here.

## MATERIALS AND METHODS

Leaf dip preparations were made by touching freshly cut surfaces of infected leaves to droplets of 2% phosphotungstic acid, pH 6.8, on Formvar-carbon-coated copper grids and were examined with a Zeiss EM 9S-2 electron microscope. Infected leaf material also was embedded in Spurr's low viscosity epoxy embedding

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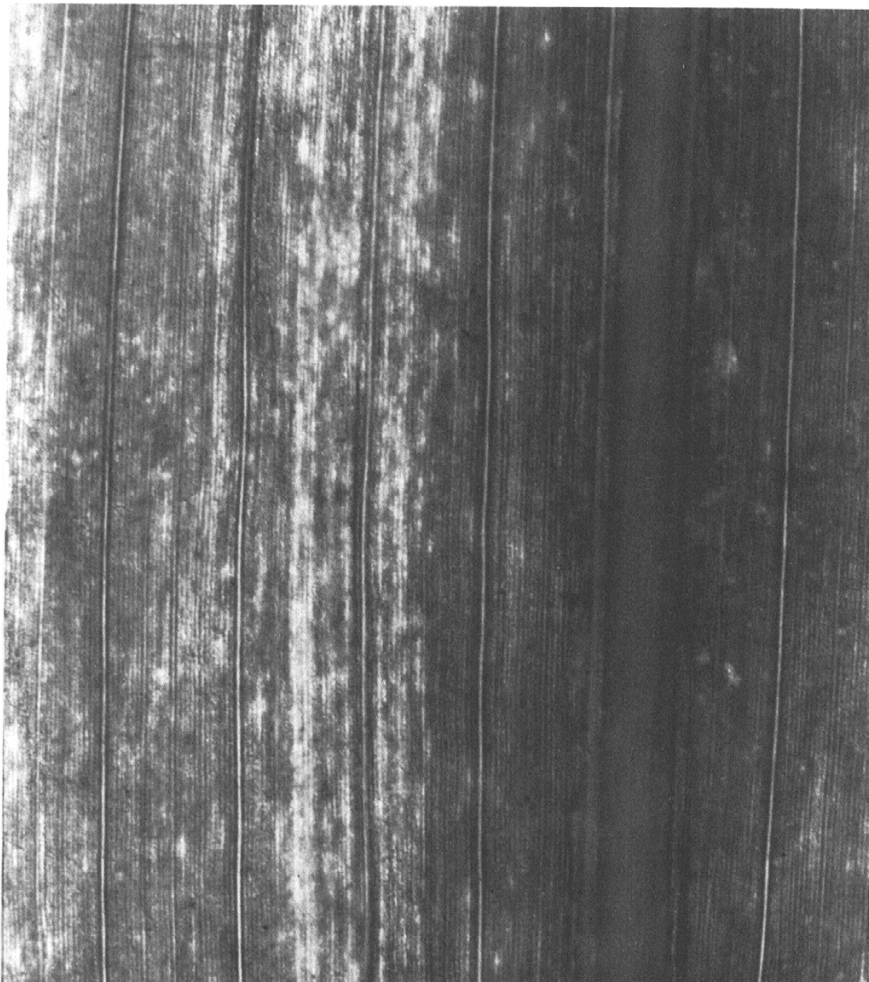


Fig. 1. Symptoms of sorghum stunt mosaic virus on leaf of sweet corn, *Zea mays* 'Golden Bantam.'

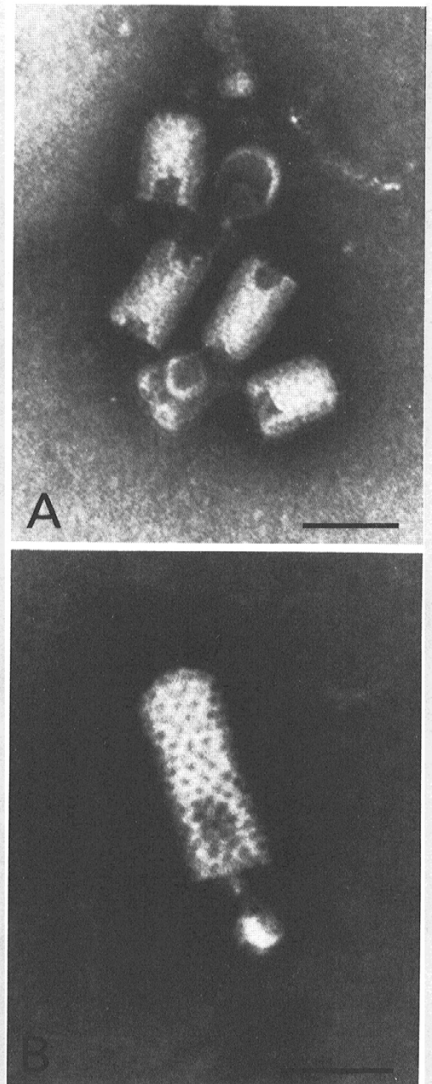


Fig. 2. Virions in negatively stained leaf dip preparations from infected corn: (A) Enveloped particles. (B) Particle lacking lipid envelope. Bar represents 100 nm.

medium after fixation in 3% glutaraldehyde (buffered with 0.01 M Na-K phosphate buffer, pH 7.2) and postfixation with 2% OsO<sub>4</sub> for 1 hr. The embedded material was later sectioned, stained for 10 min in uranyl acetate (30% w/v in absolute methanol), and examined with the electron microscope.

Insects were collected on or near diseased sorghum plants in the field and included the leafhoppers *Graminella sonora* (Ball), *Exitianus picatus* (Gibson), and *Dalbulus maidis* (DeLong); the plant hopper *Delphacodes propinqua* (Fieber); the whitefly *Aleurocybotus occiduus* Russell; and the aphids *Rhopalosiphum maidis* (Fitch) and *Toxoptera graminum* (Rondani).

Insects (50–100 per experiment) were fed on corn, *Zea mays* L. 'Golden Bantam,' 'T. E. Whitmaster,' and 'Early

Triple Sweet'; sorghum *S. vulgare* Pers. 'T. E. Haygrazer,' 'Double TXA + GO35,' and 'Double Dwarf Early Hegari'; and wheat *Triticum aestivum* L. 'Rojo Vejo.' Plants were maintained under greenhouse conditions (day temperatures 21–32 C).

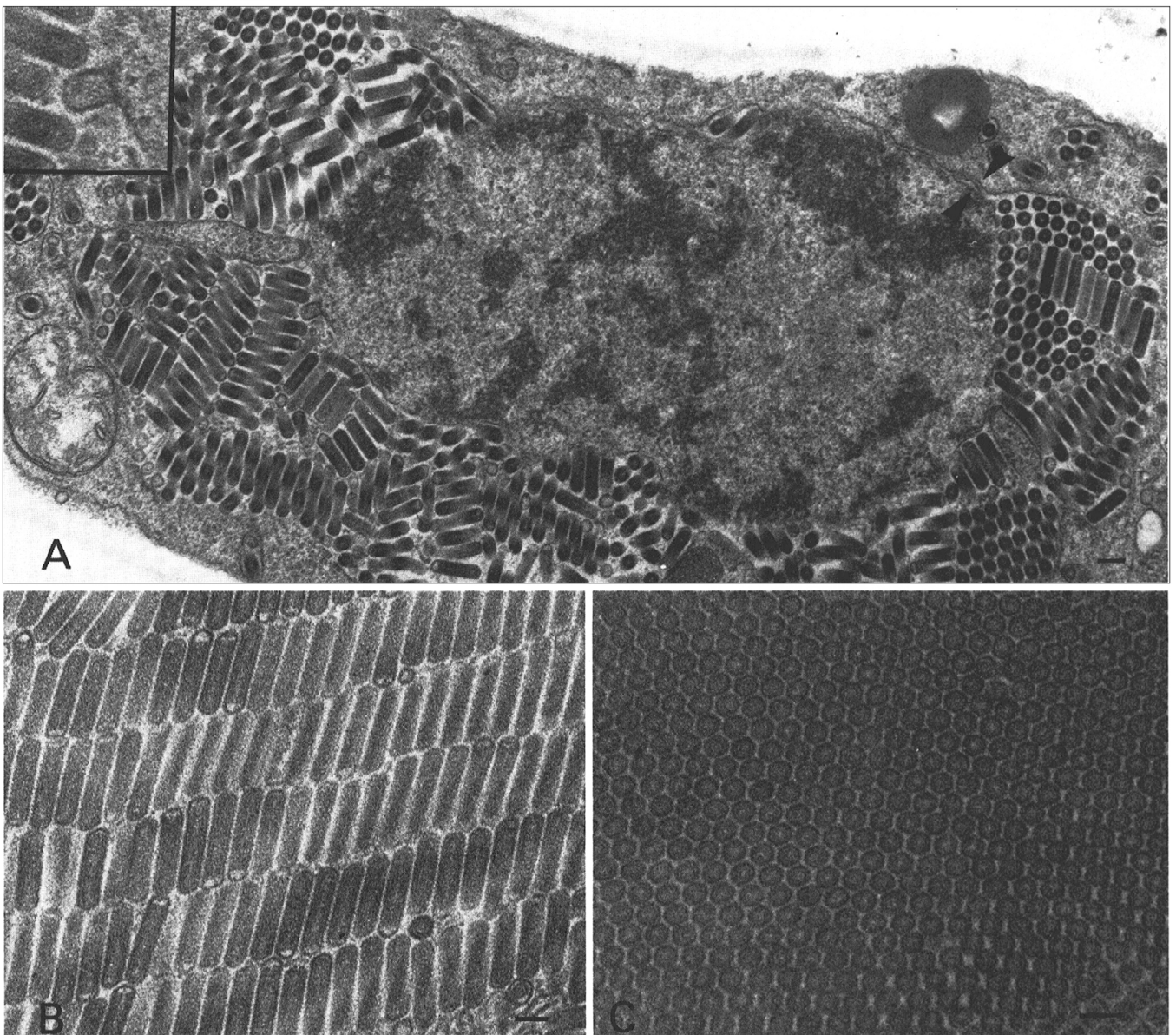
For additional transmission tests with *G. sonora*, 50–100 insects were reared on infected sorghum and allowed to feed on barley (*Hordeum vulgare* L. 'Gus'), oats (*Avena sativa* L. 'Kanota'), and wheat cv. Probred.

Infected corn or sorghum leaves were triturated in 0.01 M Na-K PO<sub>4</sub> buffer, pH 7.2, and the resultant sap was rubbed onto Carborundum-dusted leaves of *Brassica oleracea* L. var. *botrytis*; *Capsicum frutescens* L. 'Yolo Wonder'; *Chenopodium amaranticolor* Coste & Reyn.; *C. quinoa* Willd, *Cucumis sativus* L. 'National Pickling,' *Datura stramonium*

L., *Gomphrena globosa* L.; *Lycopersicon esculentum* Mill.; *Nicotiana glutinosa* L.; *N. sylvestris* Speg & Comes; *N. tabacum* L. 'Turkish,' 'Samsun NN,' and 'Xanthi'; *Phaseolus vulgaris* L. 'Great Northern'; *Vigna unguiculata* (L.) Walp.; and *Vinca rosea* L., in addition to corn, oats, barley, wheat, and sorghum.

Other extracting solutions tested, with only sweet corn as a host, were 0.005 M borate buffer, pH 9.0; 0.18 M phosphatocitric acid buffer, pH 7.0; 0.5 M borate buffer plus 0.15% thioglycolic acid, pH 8.0; 0.5 M Na citrate plus 0.5% mercaptoethanol; 0.1 M Na citrate; 0.1 M tris buffer, pH 7.2; 0.2 M acetate buffer, pH 4.5; and distilled water.

Inoculated plants were examined for development of symptoms and for virions in leaf dip preparations or sectioned leaf tissue.



**Fig. 3.** Thin sections of corn leaf tissue infected with sorghum stunt mosaic virus: (A) Perinuclear envelope filled with virions in leaf parenchyma cell. Arrows point to separation of inner and outer membranes of perinuclear envelope; insert shows virion being enveloped by budding through inner membrane of perinuclear envelope. (B) Virus aggregate in longitudinal section. (C) Virus aggregate in cross section. Bar represents 100 nm.

## RESULTS

Particles observed in leaf dip preparations were bullet-shaped, and 200 measured particles averaged  $63 \times 95$  nm (Fig. 2A). Only rarely were particles found that had no apparent lipid membrane (Fig. 2B).

In thin sections of infected tissue, bacilliform particles were found in large numbers in leaf parenchyma cells. Virions were confined primarily to the space between the inner and outer membranes of the perinuclear envelope. No particles were observed within the nucleus or existing freely in the cytoplasm. Those observed in the cytoplasm were surrounded by a membrane that appeared to be contiguous with the outer membrane of the perinuclear envelope. In many sections, there was evidence of new particles becoming enveloped by budding through the inner membrane of the perinuclear envelope (Fig. 3A). Large, densely packed viral aggregates were found in many cells. Particles in aggregates were uniform in size and morphology. Mean dimensions of 1,000 measured particles were  $68 \times 220$  nm (Fig. 3B and C).

Of the insects tested, only *G. sonora* was a vector of sorghum stunt mosaic virus (SSMV), transmitting it to corn, wheat, and sorghum. Symptoms on all three hosts were similar to those on field-grown sorghum. Acquisition time was less than 1 day, and incubation time averaged 10–12 days. Transmission efficiency was quite variable, however,

and depended on the temperature in which plants and insects were reared. In general, no transmission occurred at day temperatures below 24 C, but insect populations increased. At higher temperatures (up to 32 C), leafhoppers transmitted SSMV efficiently (up to 100%), but insect populations declined rapidly to zero. Insects fed on healthy plants maintained normal life cycles at both low and high temperatures. Because of problems in handling the vector, these results are based on limited data.

Attempts to mechanically transmit SSMV were unsuccessful.

## DISCUSSION

Symptoms, particle morphology, and cytological evidence suggest that SSMV is related to maize mosaic virus (MMV), which occurs in Hawaii and many areas of the Caribbean (2) and may also be present in the continental United States (1,7).

Based on our current knowledge, it is possible that SSMV and MMV are not identical. The vector of MMV, *Peregrinus maidis*, has not been found in California, *G. sonora* is not known to transmit MMV, and MMV is not known to infect wheat. SSMV may also be distinct from other known rhabdoviruses of Gramineae. The vector, *G. sonora*, a vector of maize chlorotic dwarf virus (6), is not known to transmit any other rhabdovirus, even though other members of this genus do. Furthermore, known vectors of rhabdoviruses infecting grasses have not been

found in California. In addition, SSMV infects corn, sorghum, and wheat; no other rhabdovirus infects all three hosts (3). It is possible, however, that a complex of two or more viruses cause SSMV.

SSMV could cause serious economic losses in California. To date, however, SSMV has been found only in Imperial County and only on two occasions.

## ACKNOWLEDGMENTS

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