Variability of Venezuelan Isolate of Maize Dwarf Mosaic Virus in Sorghum

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ABSTRACT

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A virus isolated from sorghum in Venezuela in August 1980 was identified as maize dwarf mosaic virus. The virus caused a severe reaction in seven sorghum cultivars resistant to maize dwarf mosaic virus, particularly Tx2536 and RTx430, but did not infect the immune QL3 and QL11. The virus infected Johnsongrass and was serologically related to maize dwarf mosaic virus but not to sugarcane mosaic virus A or H. It is proposed that the Venezuelan virus is a variant of maize dwarf mosaic virus strain A.

Grain sorghum (*Sorghum bicolor* (L.) Moench) is an important feed grain in Venezuela; about 400,000 ha was planted in that country in 1980 (12). Virus diseases caused the greatest loss of all factors reducing yield of grain sorghum in Venezuela in 1980–1981 (5). Highest virus disease incidence occurred in sorghum planted in August. Ordosgoitty and Malaguti (8) reported the first virus disease of sorghum in Venezuela in 1969, and later Ordosgoitty (6) and Ordosgoitty and Gonzales (7) found both maize dwarf mosaic virus (MDMV) and sugarcane mosaic virus (SCMV) strains A and H in Venezuelan sorghum fields. In 1979, Riccelli (12) reported that the Venezuelan isolate of the virus infecting sorghum studied by Ordosgoitty and Viera (9) caused mottling, chlorosis, red leaf, necrosis, and death of the whorl in sorghum.

Ordosgoitty and Viera (9) had also found that the virus could be transmitted mechanically and by the aphid *Rhopalosiphum maidis* (Fitch) to corn (*Zea mays* L.), grain sorghum, Johnsongrass (*S. halepense* (L.) Pers.), *S. arundinaceum* Stapf., and raoulgrass or itchgrass (*Rottboellia exaltata* L.). Ordosgoitty et al (10) reported the virus particle size as 12 × 755 ± 10 nm. Riccelli (12) and Ordosgoitty et al (8,10) agreed that the virus was a new strain of SCMV. They based their conclusions on the high frequency of necrosis that occurred in lines that reacted only mildly to MDMV strain A (MDMV-A) (14), particularly Tx2536 and RTx430. Riccelli (12) observed that sorghum accession QL3, which is immune to MDMV-A, was susceptible to the Venezuelan virus isolate.

Since the first observations in 1969, the virus attacking sorghum has increased in Venezuela and in 1977 reached epidemic proportions. The virus has been reported in every region where sorghum is grown, reaching devastating levels where highly susceptible hybrids were planted (5). Mena (4) estimated the 1977 incidence at 30% for commercial fields of the Mexican hybrid Master 911 in Guarico. Riccelli (13) observed all degrees of virus symptoms in Venezuelan sorghum. This investigation was undertaken to determine the identity of the virus, whether MDMV immunity of sorghum accessions QL3 and QL11 had broken down, and the reaction of MDMV-A resistant lines.

MATERIALS AND METHODS

In August 1980, at the invitation of M. Riccelli and H. A. Mena, R. W. Toler and D. T. Rosenow visited Venezuela and collected sorghum leaf samples and field data at Macapo, Maracay, and Villa de Cura. Lines in the international virus nursery, the standard-line virus nursery, and the international sorghum disease and insect nursery were rated for virus symptoms as follows: 0 = no symptoms, MSRL = mosaic and slight red leaf, MRL = mosaic and red leaf, MN = mosaic and necrosis. Samples of suspect virus-infected and one sample of healthy-appearing tissue were collected from nine sorghum lines and species and returned to the virus laboratory at Texas A&M University. These samples, collected at Villa de Cura, Macapo, and Gonzalito, included QL3 (India and Texas seed sources), QL11, QL3 × BTx618, and RTx430 sorghums; *S. halepense*; and *R. exaltata*.

Quick-dip preparations of diseased and healthy sorghum tissues were examined with a Hitachi HS7S electron microscope. Sap from a cut edge of a leaf was allowed to flow into a drop of distilled water on a Formvar-coated

Table 1. Reaction of grain sorghum lines and accessions to natural infection by the Venezuelan virus and to mechanical inoculation with maize dwarf mosaic virus strain A (MDMV-A)[a]

Entry	Venezuelan virus	MDMV-A
SC063-11E (IS1269 dr.)	M	MRL
SC175-14E	M	MRL
Tx3197	MN	MRL
Atlas	MN	MRL
NM31	MN	MRL
RTx7000	MN	MRL
BTx618	MN	MRL
BTx3048	MN	MRL
BTx378	MN	MRL
RTx430	MN	MSRL
Tx2737	MN	MSRL
Tx2536	MN	MSRL
ATx399 × RTx430	MN	M
SA394	MN	M
Tx412	MN	M
BTx623	MN	M
RTAM428	M	M
RIO	M	M
Tx09	M	M
SA8735	M	M
BTx398	M	M
BTx624	M	M
BTx625	M	M
Tx414	M	M
Tx7078	M	M
BTx399	M	M
BTx3566	M	M
Sc097-14	M	M
QL3 (India)	0	0
QL3 (Texas)	0	0
QL11	0	0
ATx618 × QL3	0	0

[a] MN = mosaic and necrosis, MRL = mosaic and red leaf, M = mosaic, MSRL = mosaic and slight red leaf, and 0 = no symptoms.

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electron microscope grid. The drops were air-dried, stained for 1 min with a 2% solution of phosphotungstic acid (pH 6.5), and then scanned for virus particles.

Diseased sorghum samples were also assayed by serologically specific electron microscopy (SSEM) (1–3) using antisera to the following viruses: wheat streak mosaic, maize mosaic, maize chlorotic dwarf, maize chlorotic mottle, sugarcane mosaic strain A, sugarcane mosaic strain H, maize dwarf mosaic strain A, and maize dwarf mosaic strain B. Ouchterlony gel diffusion tests were performed in addition to SSEM using the method of Purcifull and Batchelor (11).

For ultrastructure examination, healthy and diseased leaf tissue pieces (1–2 mm) were successively fixed in each of the following at 25 C: 2% glutaraldehyde in 0.01 M phosphate buffer (pH 7.1) for 3 hr, 2% osmium tetroxide in 0.1 M phosphate buffer (pH 7.1) for 1 hr, and 1% uranyl acetate for 16 hr. Following fixation, specimens were dehydrated in a graded ethanol series, placed in acetone for 2- to 15-min intervals, and embedded in a graded Epon 812 plastic. Sections were cut on an LKB ultramicrotome, stained in uranyl acetate for 1 min and 0.5% lead citrate for 1 min, and examined with the electron microscope.

RESULTS AND DISCUSSION

Quick-dip assays of healthy and diseased tissue of all nine accessions and species were negative for virus particles. In the ultrastructure study, only RTx430, *S. halepense*, and *R. exaltata* contained flexuous rods and pinwheel inclusions. None of the samples of QL3, QL11, or QL3 × BTx618 contained virus particles or pinwheels. In gel-diffusion tests, there were reactions between the Venezuelan virus and MDMV-A antiserum, but none with the other seven antisera. SSEM tests were negative for all the viruses tested except MDMV-A. Sap extracts from RTx430, *S. halepense*, and *R. exaltata* were positive when reacted with MDMV-A antiserum, showing 24, 27, and 16, respectively, particles per electron microscope field at ×20,000. Sap from *R. exaltata* also reacted positively to SCMV-A antiserum with 20 particles per field, indicating infection with two viruses. QL3, QL11, and QL3 × BTx618 were negative with all antisera tested.

A comparison of symptoms between naturally infected sorghum accessions in Venezuela and those mechanically infected with MDMV-A in Texas is shown in Table 1. The major differences occurred with BTx378, RTx430, Tx2737, Tx2536, BTx623, Tx412, and SA394, all of which were more severely affected when inoculated with the Venezuelan virus (necrosis of growing point). The line that best differentiated the Venezuelan virus and MDMV-A was RTx430.

The QL-numbered sorghum lines that were immune to MDMV-A in Texas and Australia were also immune to the Venezuelan virus, although they were earlier reported to be susceptible to the Venezuelan virus (12). Possibly the earlier report was of some other disease. According to Riccelli (12), Ordosgoitty and Viera reported SCMV strains H and A and MDMV in Venezuela and concluded that the virus in sorghum was a strain of SCMV. We believe that the Venezuelan virus is a strain of MDMV rather than SCMV for the following reasons: a) Both 2n and 4n *S. halepense* are hosts, whereas strains of SCMV do not have Johnsongrass as a host; b) neither the Venezuelan virus nor MDMV-A infects Krish derivatives QL3 or QL11; c) sorghum breeding lines that were generally susceptible to the Venezuelan virus (12) were generally susceptible to MDMV, although RTx430, Tx412, BTx378, BTx623, Tx2737, Tx2536, and SA394 (Table 1) were more susceptible to the Venezuelan isolate than to MDMV-A; and d) the Venezuelan isolate was serologically related to MDMV-A and not to SCMV-A or -H. We propose that the Venezuelan isolate is a variant of MDMV rather than SCMV.

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