New Sources of Resistance in Sorghum (*Sorghum bicolor*) Germplasm Are Effective Against a Diverse Array of *Potyvirus* spp.

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Abstract

Seifers, D. L., Perumal, R., and Little, C. R. 2012. New sources of resistance in sorghum (*Sorghum bicolor*) germplasm are effective against a diverse array of *Potyvirus* spp. Plant Dis. 96:1775-1779.

Sorghum is a host to numerous *Potyvirus* spp. and its germplasm encompasses a wide range of infection responses to these viruses. We determined how 183 mini-core-collection sorghum germplasm accessions responded to mechanical inoculation with *Maize dwarf mosaic virus* (MDMV) in growth regimes in which they were maintained at 30°C followed by 16°C for 5 days. Accessions that appeared immune to MDMV in this initial screening were evaluated for their response in a similar temperature maintenance regime to mechanical inoculation with MDMV, *Sugarcane mosaic virus* strain MDB (SCMV-MDB), *Sorghum mosaic virus* (SrMV), *Zea mosaic virus* (ZeMV), and Kansas, Nigerian, and Australian isolates of *Johnsongrass mosaic virus* (JGMV-KS, -N, and -Aus, respectively). In both experiments, MDMV systemically infected all accessions except international sorghum accession number (IS) 7679 and IS 20740. These accessions also proved resistant to MDMV, SCMV-MDB, SrMV, and JGMV-N but both were susceptible to the JGMV-KS and JGMV-Aus isolates. IS 7679 but not IS 20740 was resistant to infection with ZeMV. These observations suggest that IS 7679 and IS 20740 might serve as new sources of resistance to several *Potyvirus* spp. that systemically infect sorghum.

Sorghum (*Sorghum bicolor* (L.) Moench) is a host to several virus species of the family *Potyviridae,* including *Maize dwarf mosaic virus* (MDMV) (formerly MDMV-A), *Sugarcane mosaic virus* strain MDB (SCMV-MDV; formerly MDMV-B), *Johnsongrass mosaic virus* (JGMV), *Sorghum mosaic virus* (SrMV), and *Zea mosaic virus* (ZeMV). Of these, MDMV and SCMV-MDB are most often associated with the disease called maize dwarf mosaic of corn and sorghum (21,22). JGMV has also been documented to naturally infect sorghum in the United States (14,17). A few instances of natural infection of sorghum with SrMV in Texas have been reported (9), and ZeMV occurs in Israel, where it infects maize and sorghum (18).

Maize dwarf mosaic of sorghum has been recognized as an important disease of sorghum in the United States (22). Two principal vectors of MDMV and SCMV-MDB are the corn leaf aphid, *Rhopalosiphum maidis* (Fitch), and the greenbug, *Schizaphis graminum* Rond. (4,7). At warm temperatures, sorghum plants respond initially to infection with mosaic symptoms but produce a red-leaf symptom when they are exposed to cooler temperatures of about 16°C (12,21). This symptom is a consequence of the necrosis induced by infection. The term "red-leaf" is also applied to sorghum genotypes that produce tan pigments during necrosis (10). In earlier literature, the mosaic reaction was described as a "resistant" response (8), although it demonstrated susceptibility to systemic virus infection. Other authors have described this "mosaic-only" response to infection at different temperatures as resistance, "tolerance", or as a reaction following infection that should be preferred when screening germplasm (3,5,10,22). Sorghum that respond to MDMV exclusively with mosaic symptoms have been shown to suffer less yield loss when infected, ranging from 31.4% for 'Tx 412', to no loss of yield for 'Tx 414', while 'CK-60', which develops necrosis, experienced a 47.6% yield loss (5). By contrast, yield reductions of sorghum infected with SCMV-MDB were least severe for those with only mosaic and most severe in lines with red-leaf symptoms (1).

Genetic resistance in sorghum to MDMV has been historically derived from 'Krish'. In this photoperiod-sensitive forage sorghum cultivar from India, resistance is controlled by a single dominant gene designated 'Krish' (13). In Australia, the Krish gene was also found to confer resistance to systemic infection with JGMV (19), although it is ineffective against the JGMV-Kr strain (15). The Krish gene also protects sorghum from systemic infection with SCMV-MDB (10). Another breeding resource is 'Wiru' (international sorghum accession number [IS] 8789), a photoperiod-sensitive tall sorghum line which has been found to be immune to mechanical inoculation with MDMV and SCMV-MDB (10). Because germplasm with claims to resistance should confer protection in the setting in which it is intended to be grown, sources with resistance should be characterized regarding the mode of virus transmission. For example, the Nigerian grain sorghum line 'Q7539' was observed in Australia to resist natural infection with JGMV (15) but proved susceptible to systemic infection by mechanical inoculation (11). The resistance of the Q7539 line, unlike the monogenic dominant inheritance of Krish, is complexly inherited (11). Q7539 has also been shown to be resistant to MDMV in Texas (20).

In both of two mechanical inoculation tests of 183 mini-core-collection sorghum accessions (23) with a Kansas isolate of MDMV, two accessions failed to become systemically infected. This apparent immunity was also observed in subsequent experiments when IS 7679 and IS 20740 (tall with lax type panicle and photoperiod sensitive) were tested with MDMV and other *Potyvirus* spp. This study reports the results of systematic examinations of how these two sorghum accessions responded to mechanical inoculation with MDMV, SCMV-MDB, JGMV, SrMV, and ZeMV and advances the case for their use as sources of *Potyvirus* spp. resistance.

Materials and Methods

Virus source and maintenance. A Kansas isolate of MDMV (formerly MDMV-A) was used in the present study (16). The Ne-

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This research has been assigned contribution number 12-333-J from the Kansas Agricultural Experiment Station.

Accepted for publication 12 June 2012.

http://dx.doi.org/10.1094/PDIS-03-12-0224-RE

braska source of SCMV-MDB (formerly MDMV-B) was obtained from Stan Jensen (University of Nebraska, Lincoln), and the ZeMV isolate was the original used in 2000 to describe the virus (18). Three isolates of JGMV were used: a Kansas isolate (JGMV-KS) (17); the Nigerian isolate of JGMV (JGMV-N), which was used to describe the virus (17); and the Australian isolate (JGMV-Aus, obtained from Dennis Persley, Indooroopilly, Australia) that infects sorghum with the Krish gene for resistance (11,15). The PV 51 isolate of SrMV was obtained from the American Type Culture Collection (2). All isolates were increased in 'GA TE 76' sorghum in growth chambers (Percival Model PGC-15 WC) set at 30°C with a 12-h photoperiod of fluorescent light (250 μEs^{-1} m^{-2}). Symptomatic GA TE 76 sorghum plants separately infected with each virus were harvested separately at 21 days post inoculation (DPI) and frozen at –80°C just prior to the start of these studies. All inoculations of the seedlings in the experiments were conducted using the 21 DPI frozen virus sources. GA TE 76 sorghum was used because it is susceptible to systemic infection with all the *Potyvirus* spp. examined in this study (D. L. Seifers, *unpublished*).

Indirect enzyme-linked immunosorbent assay and sources of antiserum. The third leaves from the base of individual plants were sampled separately at 14 DPI. Each leaf sample was ground in a 1.5-ml microcentrifuge tube with a wood applicator stick (Fisher Scientific) at a 1:30 (wt/vol) dilution in 0.05 M carbonate buffer, pH 9.6 (6). Extracts (200 µl) were pipetted into wells of enzyme-linked immunosorbent assay (ELISA) plates (Immulon 1; Fisher Scientific) for 1 h at 37°C. Following a rinse step, the wells were incubated for 1 h at 37°C with the appropriate antivirus antibody (200 µl) in dilution buffer (6). The wells were rinsed and blocked for 1 h in blocking buffer (5% nonfat dry milk, 0.01% antifoam A, and 0.02% sodium azide in phosphate-buffered saline, pH 7.4) at 37°C. After blocking, 200 µl of anti-rabbit antibody/alkaline phosphatase conjugate (Southern Biotechnology Associates) in dilution buffer (1:3,000 vol/vol) was added to each well. The plates were held at 37°C for 1 h. The wells were rinsed, and 200 µl of p-nitrophenyl phosphate substrate (0.714 mg/ml), in substrate buffer (6) was added to each well. The plates were then held at 20 to 22°C for 30 min. Absorbance at 405 nm was measured using an iMark plate reader (Bio-Rad Laboratories). Absorbance values were arbitrarily considered positive if they were at least twice those of the equivalent mock-inoculated control value. Plants below the arbitrary threshold were considered negative in ELISA. The terms "positive" and "negative" are used for the purposes of brevity; rather than repeatedly stating that extracts from plants reacted with a given antibody set at a two times or greater than the healthy control, we state that such a plant or plants were positive and a plant or plants below the arbitrary threshold are negative.

The MDMV antiserum (PVAS 55) was obtained from the American Type Culture Collection and the ZeMV antiserum was obtained as previously described (18). A single antiserum prepared to a Kansas isolate as previously described was used for analyses of all JGMV isolates (17). The SCMV-MDB antiserum was from Ray Louie (United States Department of Agriculture–Agricultural Research Service, Wooster, OH) and the SrMV antiserum was from K.-B. G. Scholthof (Texas A&M, University, College Station). The MDMV, JGMV, SrMV, and ZeMV antisera were used at a 1:1,000 (vol/vol) dilutions and the SCMV-MDB antiserum at a 1:400 (vol/vol) dilution. The dilution for each antiserum was prepared from a stock solution of protein at 1 mg/ml (absorbance at 280 nm = 1.4).

Evaluating host responses in different temperature regimes. The 183 mini-core-collection accessions were analyzed in two sets in each of two experiments due to limited growth chamber space. The seed of each line were planted in separate rows in soil-filled (Harney clay loam soil, fine montmorillonitic, mesic type Argiustoll) flats (30 by 50 cm) with 14 rows each divided in half, so that 28 rows (150 by 35 mm) were available. The plants were maintained in a greenhouse for 7 days until the two-leaf growth stage. The plants were mechanically inoculated using a DeVilbiss

Table 1. Sorghum accessions that developed only a mosaic symptoms 19 days after mechanical inoculation with *Maize dwarf mosaic virus* in a growth chamber[a]

IS number	Country[b]	Phenotype[c]
608	United States	M2
995	United States	M2
1041	India	M2
1212	China	M2
1219	China	M2
2205	India	M2
2382	South Africa	M2
2397	South Africa	M2
2864	South Africa	M2
2872	Egypt	M2
3121	United States	M2
3971	India	M2
4360	India	M2
4515	India	M2
4613	India	M2
4631	India	M2
4698	India	M2
5094	India	M2
5386	India	M2
5529	India	M2
5667	India	M2
5919	India	M2
5999	India	M2
6351	India	M2
6354	India	M2
6421	India	M2
8012	Japan	M2
8348	Pakistan	M2
8777	Uganda	M2
8916	Uganda	M2
9108	Kenya	M2
9745	Sudan	M2
9830	Sudan	M2
10302	Thailand	M2
10867	Chad	M2
10969	United States	M2
11026	Ethiopia	M2
11619	Ethiopia	M2
11919	Ethiopia	M2
12447	Sudan	M2
12706	United States	M2
12735	Saudi Arabia	M2
12804	Turkey	M2
12883	India	M2
12937	Ethiopia	M2
13294	Venezuela	M2
13549	Mexico	M2
14010	South Africa	M2
14090	Argentina	M2
14290	Botswana	M2
15466	Cameroon	M2
15478	Cameroon	M2
15931	Cameroon	M2
15945	Cameroon	M2
16382	Cameroon	M2
17941	India	M2
18039	India	M2
19153	Sudan	M2
19445	Botswana	M2
19450	Botswana	M2
19975	Senegal	M2
20298	Niger	M2

(*continued on next page*)

[a] IS = International sorghum accession numbers are as given by Upadhyaya et al. (23).

[b] Country of origin of the sorghum is as given by Upadhyaya et al. (23).

[c] Phenotype: M1 = faint mosaic, M2 = moderate mosaic, M3 = severe mosaic, N1= necrotic spots and dashes (1 to 10% of the leaf), N2 = necrotic streaks and stripes (11 to 40% of the leaf), N3 = whole leaf necrosis (41 to 100% of the leaf), and NS = no symptoms. Following inoculation, the plants were held at 30°C for 14 days followed by 5 days at 16°C.

Table 1. (*continued from preceding page*)

IS number	Country[b]	Phenotype[c]
20679	United States	M2
20697	United States	M2
20713	United States	M2
20767	United States	M2
20816	United States	M2
20956	India	M2
21083	Kenya	M2
21425	Malawi	M2
21512	Malawi	M2
22294	Botswana	M2
22609	Sri Lanka	M2
22616	Myanmar	M2
22720	Somalia	M2
23216	Zambia	M2
23514	Ethiopia	M2
23579	Ethiopia	M2
23586	Ethiopia	M2
23590	Ethiopia	M2
23644	Gambia	M2
23684	Mozambique	M2
23891	Republic of Yemen	M2
24463	South Africa	M2
24503	South Africa	M2
25249	Ethiopia	M2
25301	Ethiopia	M2
25548	Rwanda	M2
25732	Mali	M2
25836	Mali	M2
25910	Mali	M2
25981	Mali	M2
25989	Mali	M2
26025	Mali	M2
26046	Mali	M2
26222	Togo	M2
26617	Madagascar	M2
26694	South Africa	M2
26701	South Africa	M2
27557	Burkina Faso	M2
27697	Sierra Leone	M2
27786	Morocco	M2
27887	South Africa	M2
28141	Republic of Yemen	M2
28313	Republic of Yemen	M2
28389	Republic of Yemen	M2
28449	Republic of Yemen	M2
28451	Republic of Yemen	M2
28614	Republic of Yemen	M2
29091	Republic of Yemen	M2
29100	Republic of Yemen	M2
29187	Swaziland	M2
29241	Swaziland	M2
29304	Swaziland	M2
29314	Swaziland	M2
29358	Lesotho	M2
29441	Lesotho	M2
29468	Lesotho	M2
29519	Lesotho	M2
29627	South Africa	M2
29654	China	M2
29689	Zimbabwe	M2
29714	Zimbabwe	M2
29733	Zimbabwe	M2
29914	Zimbabwe	M2
30079	Zimbabwe	M2
30231	Zimbabwe	M2
30383	China	M2
30400	China	M2
30451	China	M2
30460	China	M2
30507	Republic of Korea	M2
30508	Republic of Korea	M2
30533	Republic of Korea	M2
30536	Republic of Korea	M2

(*continued in next column*)

Table 1. (*continued from preceding column*)

IS number	Country[b]	Phenotype[c]
30562	Republic of Korea	M2
30572	Cameroon	M2
31299	Uganda	M2
31706	Republic of Yemen	M2
31714	Republic of Yemen	M2
32245	Republic of Yemen	M2
32295	India	M2
32349	India	M2
32787	Somalia	M2
33353	Kenya	M2
33844	India	M2

Number 152 atomizer (4.2 kg/cm^2 air pressure) on both leaves with a 1:20 (wt/vol) dilution of extract prepared from a frozen stock of GA TE76 sorghum infected with the MDMV-KS isolate. The inoculated plants were held in a growth chamber (Percival Model PGC-15WC) set at 30°C with a 10-h photoperiod under fluorescent light (250 μEs^{-1} m^{-2}). The plants were rated at 14 days DPI for symptom phenotype as follows: M1 = faint mosaic, M2 = moderate mosaic, M3 = severe mosaic, N1= necrotic spots and dashes (1 to 10% of the leaf), N2 = necrotic streaks and stripes (11 to 40% of the leaf), N3 = whole leaf necrosis (41 to 100% of the leaf), and NS = no symptoms. Immediately following this rating, the temperature in the chamber was reduced to 16°C and the plants were rated for symptom type 5 days later at 19 DPI. On the same day as the final rating, the remaining accessions were inoculated as described above and held in the same chamber at 30°C, and the process was repeated. The experiment was repeated. The number of plants in the two experiments for each line varied from 6 to 14. Sorghum line 'KS83' and GA TE 76 served as controls, each inoculated with MDMV. When exposed to 16°C for 5 days, the former develops leaf necrosis and the latter mosaic symptoms only (D. L. Seifers, *unpublished*).

Response of sorghum accessions IS 7679 and IS 20740 to different *Potyvirus* spp. Metal (30 by 50 cm) flats that were filled with a Harney clay loam soil (soil as described above) having 28 rows were prepared. On three consecutive days, seed of IS 7679, IS 20740, GA TE 76, and 'ICI 5616' were planted into four separate rows constituting a set and the same set was planted seven times in the flat, creating a total of 28 rows. Following each planting, the flats were held in a growth chamber (Percival Model PGC-15WC) at 30°C with a 10-h photoperiod under fluorescent light (250 μEs^{-1} m^{-2}). When plants were at the two-leaf growth stage, plants in each of the seven sets were mechanically (finger-rub) inoculated separately with a 1:10 (wt/vol) extract of the appropriate virus prepared from GA TE 76 inoculated 14 days earlier with the respective virus. The seven viruses used in this study were MDMV-KS, SCMV-MDB, PV 51 isolate of SrMV, ZeMV, JGMV-KS, JGMV-N, and JGMV-Aus. The experiment was repeated two more times on consecutive days. The plants in each of the three experiments were held in the same growth chamber at 30°C under the same lighting conditions as described above. The plants were rated for symptoms at 14 DPI followed by sampling of the third leaf of each plant individually and processed for analysis in ELISA as described above. The virus controls were sorghum GA TE 76, which is susceptible to all viruses used in this study, and ICI 5616, which is systemically infected by the JGMV-Aus virus isolate (D. L. Seifers, *unpublished*).

Results

Evaluation of phenotypes maintained in different temperature regimes. Examination of the 183 mini-core-collection accessions after inoculation with MDMV showed that they expressed a diverse range of disease phenotypes (Tables 1, 2, and 3). Of the 183 accessions, 146 had plants that developed only mosaic symptoms (Table 1). Sixteen IS accessions had plants which developed necrosis (Table 2). Interestingly, 19 accessions had plants that did

Table 2. Sorghum accessions that developed necrosis 19 days after mechanical inoculation with *Maize dwarf mosaic virus* in a growth chamber[a]

IS number	Country[b]	Phenotype[c]
1233	China	M2N1
2389	South Africa	M2N3
2413	Iran	M2N2
2426	Afghanistan	M2N3
3158	South Africa	M2N3
7310	Nigeria	M2N2
9177	Kenya	M2N3
13782	South Africa	M2N3
13893	South Africa	M2N2
20762	United States	M2N2
22986	Sudan	M2N2
24462	South Africa	M2N3
24953	Zambia	M2N1
26749	South Africa	M2N2
29335	Swaziland	M2N1
29582	Lesotho	M2N2

[a] IS = International sorghum accession numbers are as given by Upadhyaya et al. (23).
[b] Country of origin of the sorghum is as given by Upadhyaya et al. (23).
[c] Phenotype: M1 = faint mosaic, M2 = moderate mosaic, M3 = severe mosaic, N1 = necrotic spots and dashes (1 to 10% of the leaf), N2 = necrotic streaks and stripes (11 to 40% of the leaf), N3 = whole leaf necrosis (41 to 100% of the leaf), and NS = no symptoms. Following inoculation, the plants were held at 30°C for 14 days followed by 5 days at 16°C. Plants of these lines developed necrosis after exposure to 16°C.

Table 3. Sorghum accessions with mixed disease phenotypes 19 days after mechanical inoculation with *Maize dwarf mosaic virus* in a growth chamber[a]

IS number	Country[b]	Phenotype[c]
7305	Nigeria	M3 & M3N1
8774	South Africa	M2 & M2N1
12697	Australia	M3 & M3N1
12945	Nicaragua	M3 & M3N2
12965	Cuba	M2 & M2N2
15170	Cameroon	M2 & M2N2
19389	Bangladesh	M3 & M3N1
20632	United States	M3 & M3N1
21863	Syrian Arab Republic	M3 & M3N1
24453	South Africa	M3 & M3N1
24492	South Africa	M3 & M3N2
26737	South Africa	M3 & M3N3
29239	Swaziland	M3 & M3N3
29326	Swaziland	M3 & M3N1
29392	Lesotho	M2 & M2N2
30450	China	M2 & M2N2
30466	China	M2 & M2N1
30838	Cameroon	M2 & M2N1
31043	Uganda	M3 & M3N1

[a] IS = International sorghum accession numbers are as given by Upadhyaya et al. (23).
[b] Country of origin of the sorghum is as given by Upadhyaya et al. (23).
[c] Phenotype: M1 = faint mosaic, M2 = moderate mosaic, M3 = severe mosaic, N1 = necrotic spots and dashes (1 to 10% of the leaf), N2 = necrotic streaks and stripes (11 to 40% of the leaf), N3 = whole leaf necrosis (41 to 100% of the leaf), and NS = no symptoms. Following inoculation, the plants were held at 30°C for 14 days followed by 5 days at 16°C.

not express uniform phenotypes but encompassed individuals whose symptoms were exclusively of the mosaic type and others that developed necrosis (Table 3). The accessions IS 7679 and IS 20740 differed from all others by uniformly failing to develop symptoms in both experiments.

Responses of IS 7679 and IS 20740 to inoculation with a diverse array of *Potyvirus* spp. Plants of the accessions inoculated with the seven viruses were differentially infected depending upon isolate (Table 4). All GA TE 76 plants were systemically infected when inoculated with each virus isolate. Plants of accession IS 7679 were systemically infected with JGMV-KS and JGMV-Aus; those of IS 20740 with ZeMV, JGMV-KS, and JGMV-Aus; and those of ICI 5616 with only the JGMV-Aus isolate. In the three experiments, only symptomatic plants reacted with their respective homologous antibody, indicating that immunity, rather than latent infection, accounted for the absence of symptoms.

Discussion

The mini-core collection of sorghum accessions represents 1% of a core collection of 2,247 sorghum accessions that are representative of the species diversity in sorghum (23). In the initial experiment in which we analyzed the responses of 183 mini-core-collection accessions to inoculation with MDMV, the diverse array of host responses (Tables 1, 2, and 3) encompassed a subset of accessions (8%) which developed some level of necrosis and a much larger subset (79%), which developed only a mosaic symptom in a 16°C maintenance regime. If we had failed to discover the resistance expressed by accessions IS 7679 and IS 20740, convention would have led us to advance the numerous accessions that responded with mosaic symptoms, because the mosaic reaction has been considered the resistant, tolerant, or preferred reaction to infection (1,3,5,10,22). This preference, however, has lessened since sorghum with higher levels of resistance such as Krish, Q7539, and Wiru (IS 8789) have been identified (10,13,15). We do not know why plants in 19 of the accessions had a variable response, with some individuals developing only mosaic symptoms while others developed mosaic combined with necrosis (Table 3). This variation in phenotype may be because the accessions were not genetically homogenous.

The accessions IS 7679 and IS 20740 distinguished themselves from all others by consistently failing to develop symptoms in both experiments in which the response to MDMV was used to identify candidates for resistant host responses to other *Potyvirus* spp. The question of the sources or sources of the apparent resistance needs to be addressed. We considered the possibility that IS 7679 and IS 20740, like Krish, are photoperiod sensitive (R. Perumal, *personal communication*) and might have derived their resistance from that source by crossing. This uncertainty prompted us to conduct an extended analysis of the IS 7679 and IS 20740 accessions at 30°C by separately inoculating with MDMV, SCMV-MDB, SrMV, ZeMV, and JGMV-KS, JGMV-N, and JGMV-Aus.

Different phenotypic responses to infection were observed for the IS 7679 and IS 20740 accessions depending on the inoculated virus (Table 4). The source of *Potyvirus* spp. resistance in ICI 5616 is unknown but it is assumed that Krish is the source because it is susceptible to only the JGMV-Aus isolate. In our previous studies, we have shown that QL20, a Krish source of resistance (11), was resistant to all but the JGMV-Aus virus isolate (D. L. Seifers, *unpublished*). Interestingly, the IS 7679 and IS 20740 accessions were not susceptible to infection with the Nigerian isolate of JGMV but were susceptible to the Kansas and Australian isolates of JGMV. Another difference between the JGMV-KS and the JGMV-N isolates is that the Nigerian isolate does not infect johnsongrass or oat, whereas JGMV-KS infects both species and neither isolate infects QL3 sorghum (17; D. L. Seifers*, unpublished*). Further characterization of the resistance in the two accessions was obtained using the ZeMV isolate that systemically infected IS 20740 but not IS 7679, indicating that the two sources of resistance are not identical. Sorghum GA TE 76 responded with systemic symptoms to inoculation with each of the tested *Potyvirus* spp., which is consistent with previous work (D. L. Seifers, *unpublished*). Taken together, these data are most consistent with the resistance conferred by IS 7679 or IS 20420 not being a Krish source.

In addition to providing sources of resistance to MDMV, the IS 7679 and IS 20470 accessions may well serve as effective new sources of resistance to other *Potyvirus* spp., including SCMV-MDB and SrMV. For IS 7679, resistance extends to ZeMV as well. These two sources do not confer resistance to all the isolates of JGMV we tested, and variant isolates of other viruses yet to be tested might also attenuate the case we make here for their versatil-

Table 4. Enzyme-linked immunosorbent assay (ELISA) values and numbers of infected sorghum plants mechanically inoculated with different *Potyvirus* spp. after being held in a growth chamber at 30°C for 14 days[a]

	Sorghum entry and number of plants (*N*) analyzed for each entry[b]								
Virus[c]	***N***	**IS 7679**	***N***	**IS 20740**	***N***	**ICI 5616**	***N***	**GA TE 76**	***N***	**Healthy**
MDMV	0/15	0.005 (±0.001)	0/22	0.004 (±0.000)	0/22	0.004 (±0.000)	3/3	**0.445** (±0.068)	0/3	0.008 (±0.001)
SCMV-MDB	0/19	0.005 (±0.000)	0/22	0.004 (±0.001)	0/25	0.004 (±0.001)	3/3	**0.201** (±0.043)	0/3	0.013 (±0.004)
SrMV	0/14	0.003 (±0.001)	0/22	0.003 (±0.001)	0/28	0.003 (±0.001)	3/3	**0.465** (±0.091)	0/3	0.010 (±0.003)
ZeMV	0/14	0.032 (±0.013)	23/23	**0.771** (±0.034)	0/25	0.076 (±0.009)	3/3	**0.955** (±0.067)	0/3	0.074 (±0.024)
JGMV-KS	13/13	**0.641** (±0.090)	18/18	**0.560** (±0.018)	0/23	0.031 (±0.002)	3/3	**0.667** (±0.015)	0/3	0.022 (±0.003)
JGMV-N	17/17	0.012 (±0.003)	0/23	0.018 (±0.003)	0/27	0.016 (±0.003)	3/3	**0.324** (±0.093)	0/3	0.019 (±0.003)
JGMV-Aus	13/13	**0.408** (±0.031)	22/22	**0.309** (±0.043)	22/22	**0.372** (±0.031)	3/3	**0.383** (±0.028)	0/3	0.018 (±0.001)

[a] Values (405 nm) are the mean from three experiments followed by ± the standard error.

[b] IS 7679 and IS 20740 are sorghum accessions and 'GA TE 76' and 'ICI 5616' are cultivars. Healthy = mock-inoculated GA TE 76 sorghum. For each value *N*, the numerator represents the number of sorghum plants with symptoms and positive in ELISA for a respective antiserum and the denominator the total number of plants inoculated with a respective virus in the three experiments. For the GA TE 76 positive controls, a single infected plant was sampled from a set of 6 to 10 symptomatic control plants for a given virus treatment in each experiment, so that three plants were analyzed. ELISA values in bold are considered positive, indicating that virus-specific antibodies reacted with extracts from plants above the arbitrary positive threshold of twice the healthy control value. Only symptomatic plants were positive.

[c] MDMV = *Maize dwarf mosaic virus*, SCMV-MDB = *Sugarcane mosaic virus* strain maize dwarf mosaic B, SrMV = *Sorghum mosaic virus*, ZeMV = *Zea mosaic virus*, JGMV = *Johnsongrass mosaic virus* (KS = Kansas, N = Nigerian, and Aus. = Australian isolates).

ity in conferring protection against a diverse array of *Potyvirus* spp. These findings point to a need for further studies to determine how the resistance conditioned by these accessions is inherited, and how effectively they protect yield of elite sorghum germplasm when under pressure from infection with a range of *Potyvirus* spp. Fresh crosses were made using IS 7679 and IS 20420 separately as donor parents with 'RTx 430', a universal adaptable sorghum susceptible to these *Potyvirus* spp. Evaluation of the F_1 to study the gene action and subsequent segregating generations will be evaluated to develop recombinant inbred lines for marker-assisted selection to MDMV and other *Potyvirus* spp. resistance.

Acknowledgments

We thank J. Ackerman for his assistance in all phases of the experiments and ELISA analyses during these studies and S. Haber for critically reading the manuscript.

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