Coat Protein Properties Indicate That Maize Dwarf Mosaic Virus-KS1 Is a Strain of Johnsongrass Mosaic Virus

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ABSTRACT

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It has recently been shown that 17 potyvirus isolates infecting maize, sorghum, and sugarcane in Australia and the United States, and previously classified as strains of sugarcane mosaic virus (SCMV), belong to four distinct potyviruses. However, the taxonomic status of other SCMV strains is uncertain at present. Using amino acid composition, high-performance liquid chromatographic peptide profiling of tryptic digests, partial amino acid sequence data, and electro-blot immunoassay of coat proteins, we confirm that the oat-infecting strain of maize dwarf mosaic virus (MDMV-O) from the United States is structurally and serologically closely related to the Australian johnsongrass strain of johnsongrass mosaic virus (JGMV-JG). Our results further show that the coat protein of the Kansas strain of maize dwarf mosaic virus (MDMV-KS1) is nearly identical to that of MDMV-O. Coat protein from both MDMV strains differs from the JGMV-JG coat protein by about 20 amino acid residues. The majority of amino acid substitutions occurred in the surface-exposed amino terminal region. By analogy with the observations that strains of potyviruses exhibit high-coat protein sequence identity, these results demonstrate that MDMV-O and MDMV-KS1 are strains of JGMV.

Additional keywords: amino acid composition, amino acid sequence, high-performance liquid chromatography, electro-blot immunoassay, maize dwarf mosaic virus, sorghum mosaic virus, sugarcane mosaic virus, tobacco etch virus.

Recent molecular and immunochemical approaches to the identification and classification of potyviruses and their strains have shown that reliance on biological properties alone can lead to incorrect taxonomic assignments (25). Some strains identified in the past on the basis of host range and symptomatology have been reclassified as strains of different viruses, and some viruses which were believed to be distinct are now considered strains of the same potyviruses (4,8,14,23–25,28). One such example is sugarcane mosaic virus (SCMV). A large number of strains of this virus has been reported to infect maize, sorghum, sugarcane, and other poaceous plant species (11). Until recently, the taxonomic status and the serological interrelationships of these strains were uncertain. Although results from cross-protection (10,12,26) and cytoplasmic inclusion morphology (2,3) indicated that the taxonomic assignments of several of these strains may be incorrect (25), suitable criteria were not available for accurate identification and classification of these strains. Recently, Shukla et al (18) demonstrated that antibodies directed to the surface-exposed N terminus of potyvirus coat proteins are virus-specific. On the basis of this and other information (22), a simple chromatographic procedure was developed to isolate virus-specific antibodies from crude antisera to intact potyvirus particles (15). Use of such virus-specific antibodies with 17 SCMV strains from Australia and the United States conclusively demonstrated that these strains represent four distinct potyviruses, namely, maize dwarf mosaic virus (MDMV), johnsongrass mosaic virus (JGMV), sorghum mosaic virus (SrMV), and SCMV (21). The taxonomic status, however, of other SCMV strains not included in the above studies is uncertain at present. One such case is the Kansas 1 (KS1) strain of MDMV. Jarjees and Uyemoto (6) have presented serological evidence indicating that MDMV-KS1 is distinct from strains of MDMV (MDMV-A), SrMV (SCMV-H, I, and M), and SCMV (MDMV-B, SCMV-A, B, and D) (21). Hence, the only possibilities are that MDMV-KS1 is either a strain of JGMV or it represents a fifth distinct virus in the SCMV subgroup of potyviruses.

Strains of JGMV were previously known to occur only in Australia (19). Recently, the oat-infecting MDMV strain (MDMV-O) from the United States (9) was shown to be a strain of JGMV on the basis of virus-specific N-terminal serology (21). Therefore, if MDMV-KS1 is a strain of JGMV, then it should also be closely related to MDMV-O.

Since correct identification of a pathogen is the first step toward studies aimed at its eventual control, we have attempted to establish the taxonomic status of MDMV-KS1 by comparing its coat protein with that of MDMV-O and the johnsongrass (JG) strain of JGMV on the basis of serological properties, amino acid composition, high-performance liquid chromatographic (HPLC) peptide profiling of tryptic digests, and amino acid sequence analysis of some peptides. To provide reference points for these comparisons, coat proteins of an Australian strain of SCMV (SCMV-SC) (13), and the HAT strain of tobacco etch virus (TEV) (1), both considered unrelated distinct potyviruses (21), were also compared.

MATERIALS AND METHODS

Virus strains and isolation of coat protein. JGMV-JG (13), MDMV-KS1 (6,7), MDMV-O (7,9), and SCMV-SC (13) were

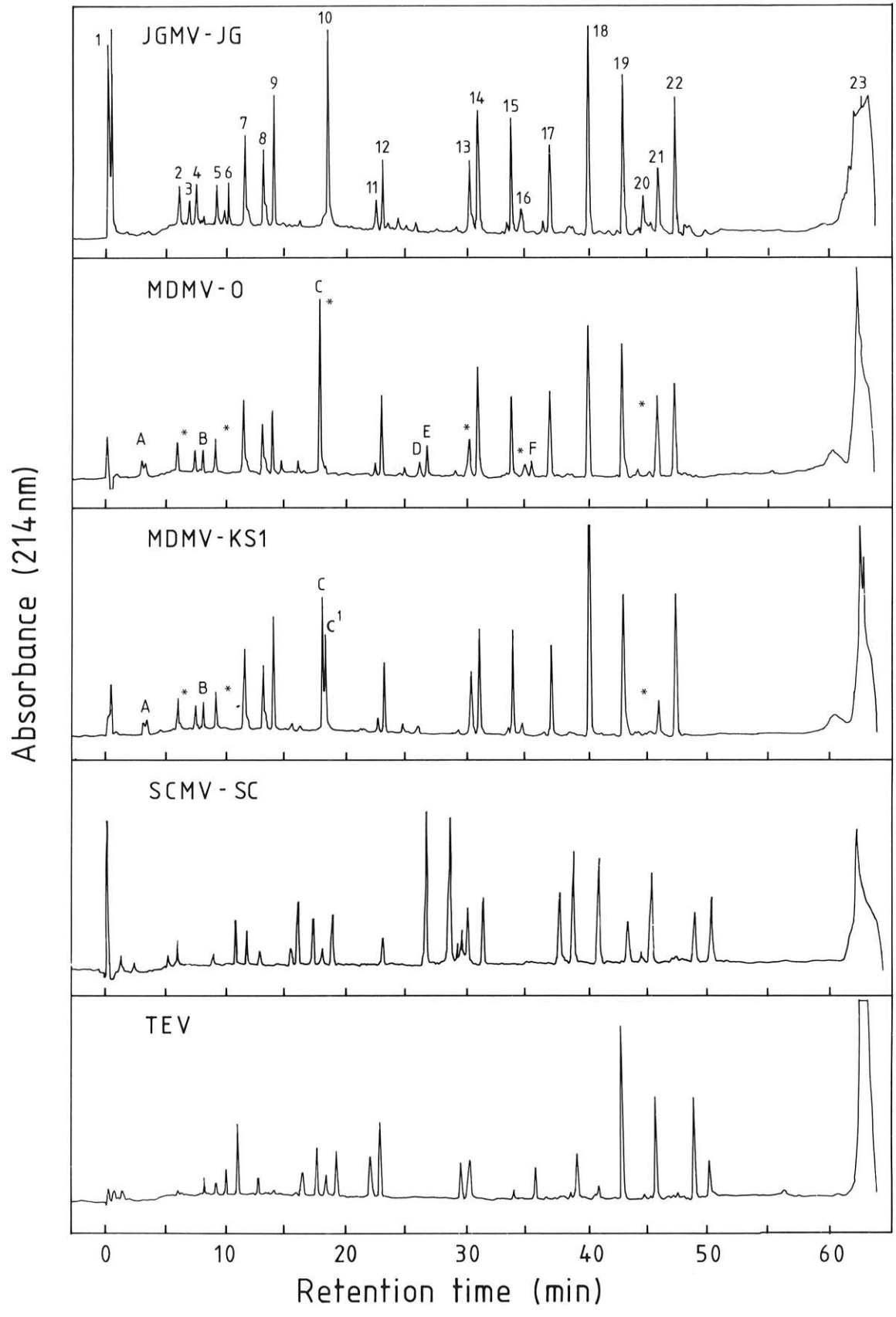

Fig. 1. Reversed phase high-performance liquid chromatography of tryptic digests of coat proteins from the johnsongrass strain of johnsongrass mosaic virus (JGMV-JG), the oat-infecting strain of maize dwarf mosaic virus (MDMV-O), the Kansas 1 strain of maize dwarf mosaic virus (MDMV-KS1), the sugarcane strain of sugarcane mosaic virus (SCMV-SC), and the tobacco etch virus (TEV). Peptides bound to the column were eluted with a linear gradient of 0–33% acetonitrile in 0.1% aqueous trifluoroacetic acid over 60 min at a flow rate of 1 ml/min and temperature of 45 C. Numbered peaks in the JGMV-JG profile and equivalent peaks in the MDMV-O and MDMV-KS1 profiles were collected and in some cases analyzed. Lettered peaks in the MDMV-O and MDMV-KS1 profiles have retention times distinct from major peaks of the JGMV-JG profile. Asterisks mark retention times of peaks present in JGMV-JG but absent in the MDMV-O or MDMV-KS1 profiles.

propagated in corn, cultivars Gold cup or Iochief, whereas TEV-HAT (1,21) was propagated in tobacco, cultivar Samsun. The viruses were purified by clarification of sap with chloroform and carbon tetrachloride, one cycle of differential centrifugation, and one or two cycles of sucrose-cesium sulphate density gradients (15). Coat protein was isolated by procedures described elsewhere (13).

Antiserum. The JGMV-JG antiserum used has been described in detail (22). It is the second bleed antiserum obtained after five intravenous injections (1 mg of virus per milliliter per injection) at days 1, 8, 15, 30, and 37. This antiserum did not react with 12 other potyviruses tested including SCMV-SC, except with watermelon mosaic virus 2, with which JGMV-JG forms a paired serological relationship (18,22). The bulk of the antibodies in this antiserum recognize surface-located, virus-specific N-terminal components of the coat protein (22).

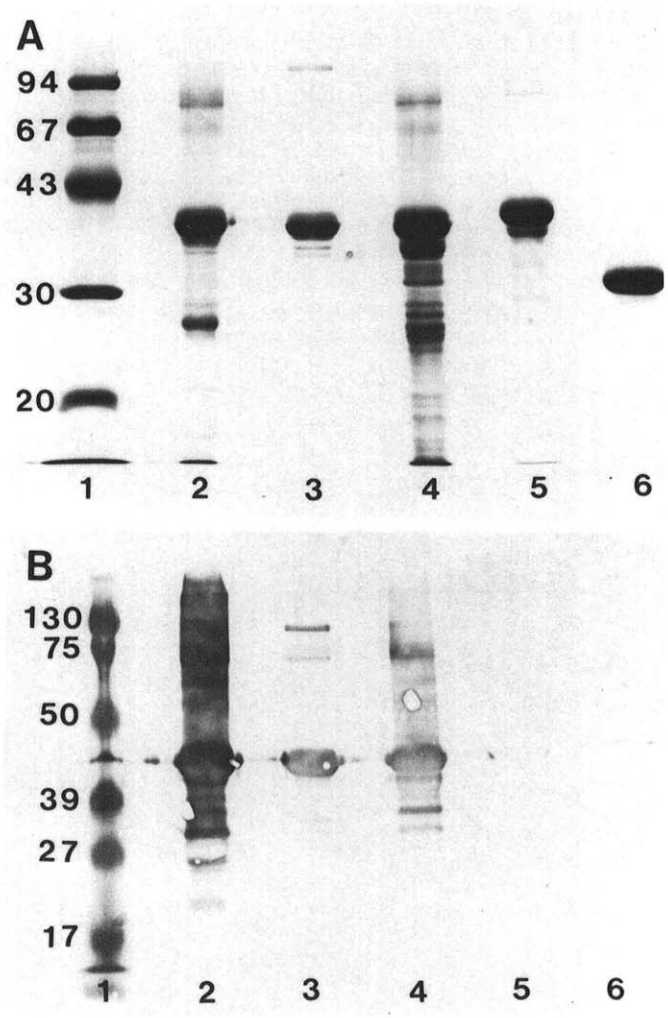

Fig. 2. A, Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (12% polyacrylamide) and **B,** electro-blot immunoassay of coat proteins of potyviruses. Lane 1 in A and B: Pharmacia and Bio-Rad (prestained) molecular weight markers, respectively. Lanes 2-6: the johnsongrass strain of johnsongrass mosaic virus (JGMV-JG), the Kansas 1 strain of maize dwarf mosaic virus (MDMV-KS1), the oat-infecting strain of maize dwarf mosaic virus (MDMV-O), the sugarcane strain of sugarcane mosaic virus (SCMV-SC), and the hat strain of tobacco etch virus (TEV-HAT), respectively. **A**, stained with Coomassie Brilliant Blue 250. **B**, reacted with a second bleed antiserum to intact particles of JGMV-JG. Approximately 5 μg of the coat protein was loaded per slot of the gel. A and B are from duplicate gels run at the same time. The lower molecular weight bands seen in lanes 2–4 of B are degradation products of the coat proteins and the higher molecular weight bands are coat protein dimers which display similar serological properties as the corresponding coat protein monomers (18). The line joining the major coat protein bands across B is an artifact.

Electro-blot immunoassay. Immunoassays were carried out after sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) of coat proteins using nitrocellulose strips and horseradish peroxidase-conjugated second antibodies according to the manufacturer's directions (Bio-Rad Laboratories, Richmond, CA) as described before (15). Prestained marker proteins were used to monitor transfer of protein to the nitrocellulose.

Amino acid analysis. Intact coat proteins or their peptide derivatives were subjected to vapor-phase hydrolysis at 110 C in 5.8 M HCl containing 0.01% phenol for 20–22 hr under N_2. Dried samples were analyzed on a Waters Amino Acid Analysis (Millipore Corp., Milford, MA) ion exchange column.

Enzymic digestion of viral coat protein and separation of peptides. Dried samples of 0.5–1.0 mg of coat protein were suspended in 250–500 μl of 0.05 M ammonium bicarbonate by sonication for 5 sec and then digested overnight at 37 C at a 1:50 enzyme/protein ratio with trypsin (*N*-tosyl-L-phenylalanine chloromethyl ketone [TPCK]–Worthington). Solutions were dried, vortexed with 250–500 μl of 0.1% trifluoroacetic acid, and centrifuged at 10,000 *g* in a benchtop centrifuge. Aliquots of 100–300 μl of soluble peptides were separated by reverse phase chromatography on a 5μ Vydac C18 column (The Separations Group, Hesperia, CA) using a Perkin-Elmer LC100 Series 4 liquid chromatograph (Perkin-Elmer Corporation, Norwalk, CT) and conditions as described in Figure 1. Some peptides were isolated by rechromatography of peaks on the same column at a different temperature, as described in the Results.

Peptide sequencing. Partial or complete sequences of peptide fragments up to 42 residues in length were determined using an Applied Biosystems (Foster City, CA) 470A protein sequencer.

RESULTS

Mobilities on SDS-PAGE show that the coat proteins from JGMV-JG, MDMV-KS1, and MDMV-O are very similar with an apparent M_r ~35,000 (Fig. 2) and are slightly smaller than that of SCMV-SC, but significantly larger than TEV. Following SDS-PAGE, coat proteins from a second gel were electro-blotted onto nitrocellulose and probed with the JGMV-JG antiserum. This antiserum recognized the coat protein of JGMV-JG, MDMV-KS1, and MDMV-O, but not the coat proteins of SCMV-SC or TEV (Fig. 2B).

Duplicate amino acid analyses of each of these coat proteins showed that JGMV-JG, MDMV-KS1, and MDMV-O had very similar amino acid compositions, whereas those of SCMV-SC and TEV were substantially different (Table 1).

TABLE 1. Amino acid composition of coat protein from the johnsongrass strain of johnsongrass mosaic virus (JGMV-JG), the oat-infecting strain of maize dwarf mosaic virus (MDMV-O), the Kansas 1 strain of maize dwarf mosaic virus (MDMV-KS1), the sugarcane strain of sugarcane mosaic virus (SCMV-SC), and tobacco etch virus (TEV)

	Residues/100 residues				
Amino acid[a]	JGMV-JG	MDMV-O	MDMV-KS1	SCMV-SC	TEV
Ala	9.6	9.2	9.6	10.9	9.4
Arg	6.4	6.5	6.7	5.9	7.7
Asx	15.0	14.8	14.4	9.5	12.9
Glx	10.7	10.8	11.0	12.4	11.3
Gly	7.7	6.8	6.7	12.0	6.6
His	2.3	2.1	2.4	2.4	3.0
Ile	3.7	3.8	4.0	2.6	2.7
Leu	5.3	5.4	5.2	5.5	7.4
Lys	6.9	7.1	7.2	4.8	5.2
Met	3.8	3.8	3.7	4.1	5.3
Phe	2.8	2.8	2.8	2.6	2.8
Pro	4.8	5.3	5.3	5.3	4.1
Ser[b]	6.9	7.9	7.7	4.6	4.4
Thr[b]	6.8	6.5	6.4	9.9	6.9
Tyr	3.2	3.3	3.2	3.3	3.5
Val	4.3	3.9	3.7	4.1	6.9

[a] Samples hydrolysed as described in text. Trp and Cys not determined.
[b] Uncorrected values.

Tryptic digests of the coat proteins, when separated by reversed-phase HPLC, confirmed and extended the findings of the amino acid composition data. Whereas the coat protein profiles of JGMV-JG, MDMV-O, and MDMV-KS1 were very similar, those of SCMV-SC and TEV were substantially different (Fig. 1).

TABLE 2. Comparison of amino acid compositions of related peaks from HPLC profiles of tryptic digests of the johnsongrass strain of johnsongrass mosaic virus (JGMV-JG), the oat-infecting strain of maize dwarf mosaic virus (MDMV-O), and the Kansas 1 strain of maize dwarf mosaic virus (MDMV-KS1) coat proteins[a]

	Residues/molecule			
	JGMV-JG[b]	MDMV-O[c]	MDMV-KS1[c]	
Amino acid	Peak 10	Peak C	Peak C	Peak C[1]
Ala	8	6.9 (7)	6.9 (7)	5.7 (6)
Asx	7	6.0 (6)	6.1 (6)	6.0 (6)
Glx	4	4.8 (5)	5.2 (5)	5.0 (5)
Gly	3	3.0 (3)	3.0 (3)	4.3 (4)
Lys	3	4.0 (4)	4.1 (4)	4.1 (4)
Pro	3	4.0 (4)	4.2 (4)	3.6 (4)
Ser[d]	6	6.8 (7)	6.9 (7)	7.2 (7)
Thr[d]	8	6.0 (6)	6.1 (6)	6.3 (6)
Total	(42)	(42)	(42)	(42)

[a]Peak numbers and letters refer to those of Figures 1 and 3. Peak contents were hydrolyzed and analyzed as described in the text.
[b]The composition of residues 11–52 of the known JGMV-JG sequence (13) is shown, which correlated with the amino acid analysis of peak 10 (data not shown).
[c]Values calculated on the basis of 42 residues/molecule.
[d]Values adjusted for losses during hydrolysis.

Detailed comparison of these profiles showed that 18 of the 23 major tryptic peaks observed with JGMV-JG were present in similar proportion in the MDMV-O profile and 19 in the MDMV-KS1 profile. Six peaks in the MDMV-O profile and three in that of MDMV-KS1 had retention times that did not correspond with a JGMV-JG peak. Peak A of these noncorresponding peaks was rechromatographed, and, in the case of both MDMV-O and MDMV-KS1, two separate peaks (A1 and A2) were obtained and analyzed. Peaks A1, A2, B, and C were found by amino acid analysis and sequence data to be homologous with known JGMV-JG peptides (13) derived from the amino terminal region of the molecule (Table 2 and Fig. 3). A total of 10 amino acid substitutions and one insertion (serine at residue 63) were identified when the sequences of these JGMV-JG peptides were compared with those of MDMV-O. Identical substitutions to those of MDMV-O were also found in MDMV-KS1 peptides A1, A2, B, and C. An additional substitution (peak D) was identified in MDMV-O (Fig. 1). Peaks E and F, not present in either the JGMV-JG or MDMV-KS1 profiles, were found to be subfragments of known JGMV-JG peptides (residues 107–116 and 222–225, respectively), unaltered in sequence, and presumably the products of partial substitution or partial cleavage at unusual trypsin sites.

Four major peaks common to JGMV-JG, MDMV-O, and MDMV-KS1 (peaks 12, 15, 18, and 22) were also analyzed. Amino acid compositions of corresponding peaks derived from these three proteins were indistinguishable (Table 3), except for the substitution of arginine for lysine in MDMV-O peak 12.

Amino acid analysis of peak 23 indicated that it contained a large, hydrophobic fragment of JGMV-JG protein (residues 147–211) homologous in size and composition with a core frag-

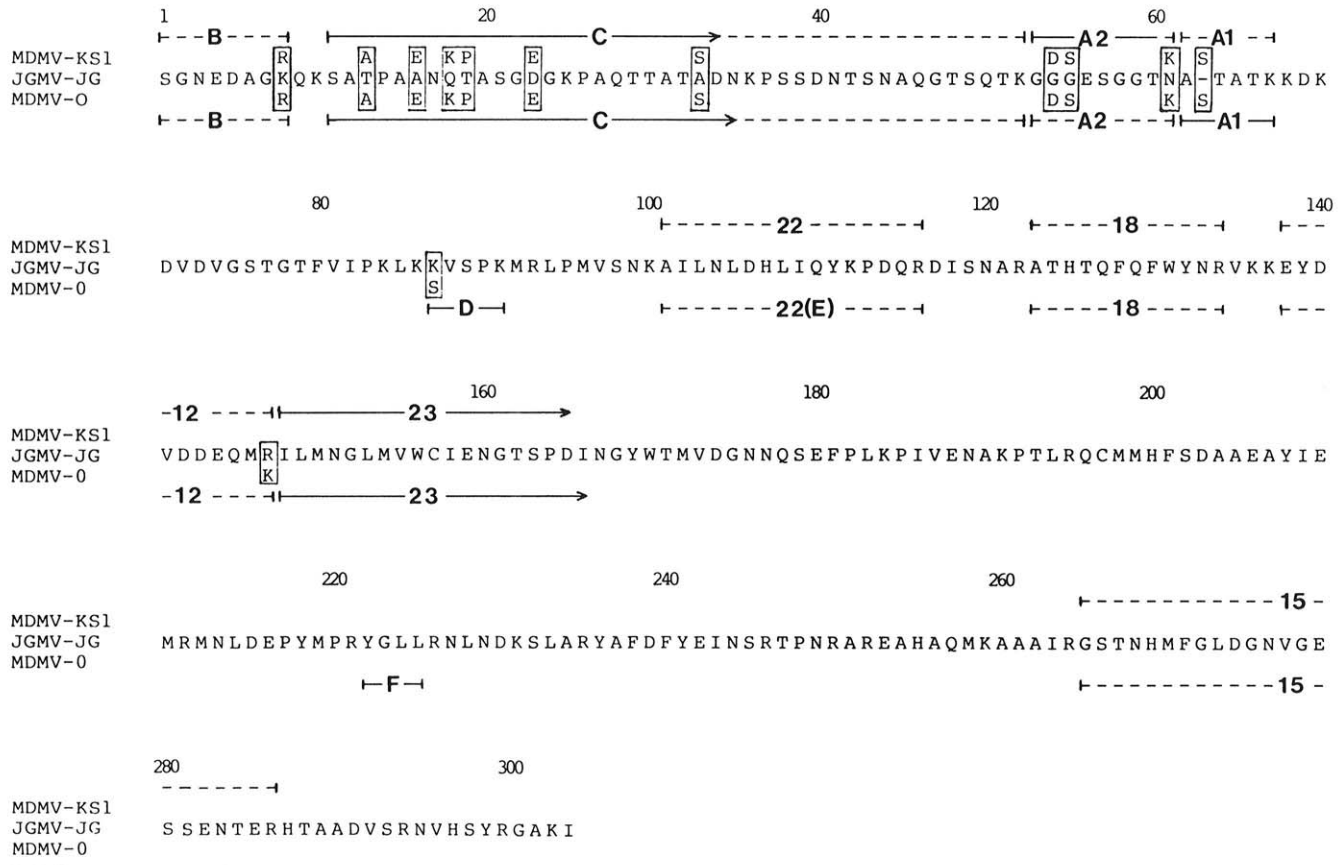

Fig. 3. Primary structure of the johnsongrass strain of johnsongrass mosaic virus (JGMV-JG) coat protein (13) showing known differences in the oat-infecting strain of maize dwarf mosaic virus (MDMV-O) and the Kansas 1 strain of maize dwarf mosaic virus (MDMV-KS1) as determined by high-performance liquid chromatograph peptide profiles, amino acid analysis, and sequencing of tryptic peptides. Dashed lines show MDMV-KS1 (above) or MDMV-O (below) tryptic peptides found by amino acid analysis to be homologous to the known JGMV-JG sequence at that region. Solid lines show peptides of MDMV-KS1 (above) or MDMV-O (below) found to have amino acid sequences homologous to the corresponding JGMV-JG sequence. Differences in sequence of MDMV-KS1 and MDMV-O peptides with the sequence of JGMV-JG coat protein are shown and boxed. Peptide numbers and letters refer to the labeled peaks of Figure 1.

ment obtained from tryptic digests of other potyvirus coat proteins. Analysis of the corresponding MDMV-O and MDMV-KS1 peaks indicated that each had a very similar amino acid composition to that of the JGMV-JG peptide (data not shown), suggesting few differences likely in sequence. This assumption was partially confirmed by sequence determination of the first 18 or 19 residues of MDMV-KS1 and MDMV-O, which yielded in each case a sequence corresponding to that known for JGMV-JG (13). The MDMV-KS1 tryptic peak adjacent to peak C (designated C^1) and corresponding in retention time to peak 10 of JGMV-JG was found by amino acid analysis (Table 2) and amino acid sequencing to be very similar to peak C of MDMV-KS1. The first 30 amino acid residues were sequenced and found to be identical to those of peak C. Amino acid analysis indicated that peak C^1 had one less alanine residue and an extra glycine residue when compared with peak C of MDMV-KS1. This change, together with other possibly undetected differences at the distal end of the C^1 peptide, may account for its altered retention time compared to that of C. Peptide C^1 may indicate the presence of a closely related mutant of the MDMV-KS1 that has copurified with that strain.

DISCUSSION

Although it is well recognized that some strains of SCMV and MDMV are serologically interrelated, it was only recently shown that a relatively clear-cut taxonomic grouping of these viruses could be made (21). The recognition that JGMV-JG and MDMV-O were strains of the same virus was based on serological studies (21), as was the finding that MDMV-KS1 was distinct from the various MDMV, SrMV, and SCMV strains (6). The results of this paper show that not only is MDMV-KS1 related to JGMV-JG, but also that MDMV-O and MDMV-KS1 have nearly identical coat protein structures (Figs. 1 and 3, Tables 1 and 2).

Electro-blot immunoassay of the coat proteins of these three strains showed their close interrelationship, in contrast to the lack of reactivity of SCMV-SC and TEV coat proteins to the second bleed antiserum to JGMV-JG (Fig. 2). Consistent with these findings were the results of amino acid analysis, which demonstrated the overall similarity of JGMV-JG, MDMV-KS1, and MDMV-O (Table 1). HPLC profiling of tryptic digests of all five proteins used in this study (Fig. 1) rapidly and precisely demonstrated the close relationship of the viral strains, confirming the serological and amino acid analysis data. Clearly, the profiles of JGMV-JG, MDMV-O, and MDMV-KS1 are similar to each other, both in height and retention time of the major, labeled peaks. Although minor differences were observed between the JGMV-JG profile and those of MDMV-KS1 and MDMV-O, these were shown to be due to a total of only 13 observed sequence substitutions. All but two of these substitutions occurred within the first 70 residues of the molecules, and, with the exception of two changes, were identical for MDMV-KS1 and MDMV-O. The presence of these substitutions was manifested by a change in the HPLC profile compared to that of JGMV-JG. Four sets of peaks common to all three HPLC profiles were analyzed to test whether identical retention times indicated identical amino acid composition. This was shown to be true for three of the four sets of peaks analyzed (Table 3). A single conservative change (lysine to arginine) was observed across the fourth set of peaks.

Some small differences in the peptide profiles of MDMV-KS1 and MDMV-O were observed, but except for peak D of MDMV-KS1, these peaks (E and F) were found to consist of fragments of peptides that were otherwise common to all three coat proteins.

In contrast to the similarity of the above three profiles, those of SCMV-SC and TEV were found to have substantially different patterns of retention time and relative peak height. This is consistent with the fact that TEV is biologically very different from SCMV or MDMV, and that SCMV is a potyvirus distinct from that of JGMV (Fig. 2) (21).

Although only a few MDMV-O and MDMV-KS1 amino acid sequence fragments were determined in this study, comparison of the peptide profiles with that of JGMV-JG enabled us to predict that both the former proteins are likely to be of similar size to that of JGMV-JG and that their sequences possibly differ from that of JGMV-JG by less than 20 residues. Furthermore, we would anticipate that the majority of these changes occur in the amino terminal regions of MDMV-KS1 and MDMV-O.

From comparison of the peptides we predict that the sequences of intact coat protein of MDMV-O and MDMV-KS1 are virtually identical, having at most only a few amino acid differences between them. These results suggest that MDMV-KS1 and MDMV-O are distinct strains of the same virus (5). However, a systematic comparison is required to determine if these genetic differences are reflected in their biological properties.

The value of peptide profiling of coat proteins as a tool for

TABLE 3. Amino acid compositions of selected peaks from HPLC profiles of coat protein tryptic digests from the johnsongrass strain of johnsongrass mosaic virus (JGMV-JG), the oat-infecting strain of maize dwarf mosaic virus (MDMV-O), and the Kansas 1 strain of maize dwarf mosaic virus (MDMV-KS1)

	Residues/molecule[a]											
	Peak 12[b]			Peak 15			Peak 18			Peak 22		
Amino acid	-JG	-O	-KS1	-JG	-O	-KS1	-JG	-O	-KS1	-JG	-O	-KS1
Ala	...	...	...	...	...	...	1.1	1.0	1.0	1.2	1.1	1.0
Arg	...	1.1	...	1.2	1.0	1.1	1.2	1.1	1.1	1.2	1.1	1.1
Asx	3.0	3.1	3.0	4.0	3.9	4.0	1.1	1.0	1.0	3.2	3.1	3.1
Glx	3.0	3.0	3.0	3.0	2.9	3.0	2.0	2.0	2.0	2.2	2.1	2.1
Gly	0.2	0.1	0.1	4.0	3.7	3.7	0.4	0.2	0.1	0.2	0.1	0.0
His	...	...	...	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Ile	...	...	...	...	...	...	...	...	...	2.0	1.9	1.9
Leu	...	...	...	1.0	1.0	1.0	...	...	...	3.2	3.1	3.1
Lys	1.1	...	1.2	...	...	...	...	...	...	1.0	1.0	1.0
Met	0.8	0.9	0.9	0.7	0.9	0.8	...	...	...	...	...	...
Phe	...	...	...	1.0	1.0	1.0	1.9	1.9	2.0	...	...	...
Pro	...	...	...	...	...	...	...	...	...	1.1	1.0	1.1
Ser[c]	...	...	...	3.0	2.8	2.7	...	0.1	...	...	...	...
Thr[c]	...	...	...	2.0	2.0	1.9	2.0	2.0	2.1	...	...	...
Tyr	0.9	0.9	0.9	...	...	...	0.7	0.8	0.8	1.0	0.9	1.0
Val	1.0	1.0	1.1	1.0	1.0	1.0	...	...	...	...	...	...
Total[d]	10	10	10	22	22	22	11	11	11	16	16	16

[a] Values calculated on basis of known peptides of JGMV-JG coat protein.
[b] Peak numbers taken from Figures 1 and 3.
[c] Values corrected for losses during hydrolysis.
[d] Individual values rounded to nearest integer.

classification of potyviruses lies in its ability to discriminate minor differences in amino acid sequences, without losing the ability to see common structure (8,16). Hence, while the coat protein structures of MDMV-O and MDMV-KS1 are somewhat different than that of JGMV-JG, the differences, when compared to established, distinct viruses such as TEV or SCMV, are relatively minor. As a consequence of these results we conclude that MDMV-O and MDMV-KS1 are strains of JGMV, a conclusion that is consistent with previous observations (13, 17, 20, 23, 28) that strains of the same potyvirus showed 90% or greater identity in their coat protein sequences, whereas distinct potyviruses exhibited sequence homology of 38–71%. This classification is consistent with earlier results from N-terminal serology (21) demonstrating that MDMV-O is a strain of JGMV and that MDMV-KS1 is distinct from a number of other MDMV, SrMV, and SCMV strains (6). This assignment is further supported by the following recent findings: 1), only JGMV-JG, MDMV-KS1, and MDMV-O infect oat cultivars, whereas strains of MDMV, SCMV, and SrMV do not (27; S. G. Jensen, *unpublished work*); 2), antisera to cytoplasmic inclusions of MDMV-KS1 and MDMV-O cross-react, but these antisera do not react with cytoplasmic inclusions from other strains of MDMV and SCMV (7); and 3), the coat proteins of JGMV-JG, MDMV-KS1, and MDMV-O are structurally as different from that of MDMV-A, the type strain of MDMV, as they are from potato virus Y, TEV, and other distinct potyviruses (25). This evidence comes from peptide profiling of MDMV-A coat protein (N. M. McKern, R. W. Toler, and D. D. Shukla, *unpublished observations*) and its amino acid sequence deduced from the coat protein gene sequence (J. Jilka and J. M. Clark, Jr., *personal communication*).

While serological comparisons in this study showed that at least one major epitope was common to these three strains, only by analysis of the peptide profiling data was it conclusively shown that the coat protein structures are highly homologous. On the basis of the above conclusions, and following the principles of nomenclature suggested by Shukla and Ward (25), we propose that MDMV-O and MDMV-KS1 should be renamed JGMV-MDO and JGMV-MDKS1, respectively.

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