An analysis of the complete sequence of a sugarcane bacilliform virus
genome infectious to banana and rice

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The genome of sugarcane bacilliform virus (ScBV), a
badnavirus, consists of a circular dsDNA. The complete
sequence of a cloned infective ScBV genome is reported
here. The genome is 7568 bp in size and possesses a
number of features suggesting that ScBV is a pararetro-
virus. A tRNA Met-binding site that may serve as a primer
for minus-strand synthesis is present. The plus-strand of
the ScBV genome contains three open reading frames
(ORFs) which are capable of encoding proteins with
calculated Mr values of 22K, 13K and 215K. The 215K
protein has regions with similarity to the RNA-binding
domains, aspartic proteases and replicases of retro-
elements. In addition, the 215K protein also has a region
with restricted similarity to the intercellular transport
proteins of plant viruses. Comparisons with the other
sequenced badnaviruses, Commelina yellow mottle
(CoYMV) and rice tungro bacilliform (RTBV) viruses,
indicate that the arrangement of the ORFs in these
viruses is conserved. Located next to the putative RNA-
binding domain is a cysteine-rich region that is unique to
the badnaviruses. When the molecular relationships of a
portion of the reverse transcriptases of plant pararetro-
viruses were determined, two badnaviruses, CoYMV and
ScBV, form one distinct cluster, whereas three caulimo-
viruses, cauliflower mosaic virus, carnation etched ring
virus and figwort mosaic virus, form a second cluster.
The badnavirus RTBV and the caulimovirus soybean
chlorotic mottle virus occupy intermediate positions
between the clusters. When introduced by Agro-

bacterium-mediated inoculation, a construct containing
1:1 copies of the cloned ScBV genome is infectious
to both rice and banana.

Introduction

Badnaviruses are plant pararetroviruses with non-
enveloped bacilliform particles with dimensions of
30 x 130 to 150 nm that contain a genome consisting of
a circular, relaxed, 7-5 to 80 kb, dsDNA molecule
(Lockhart, 1990; Medberry et al., 1990; Hay et al., 1991;
Qu et al., 1991). The genome is not covalently closed
because each strand is interrupted by a site-specific
discontinuity. Commelina yellow mottle virus (CoYMV)
is the type member for this genus. The sequences of two
badnavirus genomes, CoYMV (Medberry et al., 1990)
and rice tungro bacilliform virus (RTBV; Hay et al.,
1991; Qu et al., 1991), have recently been reported. Both
CoYMV and RTBV produce a transcript that is
terminally redundant (Medberry et al., 1990; Qu et al.,
1991). Analysis of the genomic sequences indicates that

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The nucleotide sequence data reported in this paper have been
submitted to the GenBank databank and assigned the accession
number M89923.
protein, the aspartic protease and viral replicase (reverse transcriptase and RNase H). The ORF III-derived protein of unknown function has restricted similarity to plant virus intercellular transport proteins suggesting that it may also be an intercellular transport protein (U. Melcher, personal communication). The function of the putative 46k RTBV ORF IV protein is unknown. None of the CoYMV ORFs shares significant similarity with the RTBV ORF IV protein.

The genomes of CoYMV and RTBV each contain a tRNA\textsubscript{Met} binding site, suggesting that tRNA\textsubscript{Met} serves as a primer for minus-strand synthesis during replication of the genome. The location of the minus-strand discontinuity found in CoYMV and RTBV is consistent with this hypothesis (Medberry et al., 1990; Bao & Hull, 1992). The location of the plus-strand discontinuity of CoYMV suggests that plus-strand synthesis is initiated at a polypurine region, which occurs in other retroelements (Medberry et al., 1990). Unlike other retroelements, the plus-strand discontinuity of RTBV does not map near a polypurine rich region (Bao & Hull, 1992).

The caulimoviruses are the only other known plant pararetroviruses (for reviews see Guilfoyle, 1987; Mason et al., 1987). Like the badnaviruses, these viruses employ tRNA\textsuperscript{Met}-primed reverse transcription of a transcript that is greater than genome length to replicate the minus-strand of the genome. The sequences of four caulimoviruses, cauliflower mosaic virus (CaMV; Gardner et al., 1981), carnation etched ring virus (CERV; Hull et al., 1986), figwort mosaic virus (FMV; Richins et al., 1987) and soybean chlorotic mottle virus (SoyCMV; Hasagawa et al., 1989), are known.

In this report, the complete sequence of a sugarcane bacilliform virus (ScBV) clone that is capable of infecting rice and banana is described. Analysis of the sequence indicates that the structure of the genome is similar to that of CoYMV, and that ScBV is a pararetrovirus.

**Methods**

**Construction of genomic clones.** ScBV virions and virion DNA were purified from sugarcane, *Saccharum officinarum*, as previously described (Lockhart, 1990). Genomic clones were constructed by ligating ScBV DNA that had been digested with *Bam*HI into the *Bam*HI site of pBluescript II KS+ (Stratagene). One clone containing the entire ScBV genome, pScBV20 (Fig. 1 and 6), was identified and both strands of the ScBV DNA insert were sequenced.

**DNA sequencing.** A series of sequencing templates (plasmids) were prepared from pScBV20 using the exonuclease III method of Henikoff (1984). Sequencing of plasmid DNA was performed using the dideoxyribonucleotide chain termination method of Sanger et al. (1977). Minipreps of plasmid DNA were prepared for sequencing as described by Medberry et al. (1990). Sequencing was carried out using Sequenase II (US Biochemical) as recommended by the manufacturer. In some cases, gaps in the sequence were filled in by sequencing from oligonucleotide primers that were complementary to a region adjacent to the area to be sequenced.

**Analysis of the DNA sequence.** Computer analyses of the DNA sequence and the deduced amino acid sequence of proteins encoded by the ScBV ORFs were performed using version 5.4 of the IntelliGenetics suite of programs. In addition, some comparative studies involving the putative ScBV proteins were performed at the John Innes Institute (Norwich, U.K.) with the assistance of R. Hull using the University of Wisconsin Genetics Computer Group package (Devereux et al., 1984), the Staden program (Staden 1982) and the SOMAP program (Parry-Smith & Atwood, 1991).

The molecular relationship of the regions encompassing amino acids 1350 to 1579, 1498 to 1727, 1274 to 1502, 336 to 564, 317 to 547, 328 to 556 and 291 to 524 of ScBV ORF III, CoYMV ORF III (Medberry et al., 1990), RTBV ORF III (Hay et al., 1991; Qu et al., 1991), CaMV ORF V (Gardner et al., 1981), CERV ORF V (Hull et al., 1986), FMV ORF V (Richins et al., 1987) and SoyCMV ORF V (Hasagawa et al., 1989), respectively, were determined. Trees illustrating the relationship between these regions were generated by using the neighbour-joining method (Nei, 1987) to analyse distance matrices based on the DNA sequences encoding these regions as well as the protein sequences of these regions. A distance matrix based on the DNA sequences encoding these regions was generated using the DNADIST program of version 3.4 of the PHYLIP suite of programs (J. Felsenstein, Department of Genetics, University of Washington, unpublished). The distance matrix based on the protein sequences was generated as described by Xiong & Eckbush (1990).

**Agrobacterium-mediated inoculation of banana and rice.** The construct called pScBV1 (Fig. 6a) contains 1:1 copies of the ScBV genome in the binary vector pOCA28 (Medberry et al., 1990). This plasmid was constructed by ligating the 7.3 kb *Bam*HI--*Age*I fragment from pScBV20 and the 1:1 kb *Bam*HI--*Sac*I fragment from pScBV20 between the *Xmn*I and *Sac*I sites of pOCA28. Because this construct was unstable in *Escherichia coli* (data not shown), it was necessary to introduce the ligation reaction products directly into *A. tumefaciens*, where the construct was stable, by electroporation (Mersereau et al., 1990). Agroinoculation of rice (*Oryza sativa*) var. F.K.135 and ASD7, and banana (*Musa acuminate*) was performed by the method of Dasgupta et al. (1991). Infection was scored 45 days post-inoculation using immunosorbent electron microscopy (ISEM). Partially purified extracts of individual inoculated plants were first prepared by extracting 5 g fresh leaf tissue in 250 mm-Tris-HCl pH 7.4 containing 0.5 % Na\textsubscript{2}SO\textsubscript{3} and 0.5 % (w/v) 2-mercaptoethanol. The crude extract was filtered and then clarified by centrifugation at 18000 g (max) for 15 min. Triton-X-100 was added to the supernatant to a final concentration of 2 %, and the mixture was layered over 6 ml of 30 % sucrose in extraction buffer and centrifuged for 40 min at 148000 g (max) in a Beckman type 50.2 Ti rotor. Each pellet was resuspended in 200 μl of 25 mm-Tris--HCl pH 7.4, shaken with an equal volume of chloroform and microfuged for 6 min. The clarified aqueous phase constituted the partially purified extract. Carbon-coated Formvar grids were floated for 15 min on anti-ScBV rabbit whole serum (Lockhart & Autrey, 1988) diluted 500-fold in 10 mm-Tris--HCl pH 7.2. The grids were rinsed with 30 drops of the same buffer, incubated on drops of partially purified sample extract for 2 h, rinsed with 20 drops of buffer and negatively stained by rinsing with 20 drops of 2 % sodium phosphotungstate pH 7.0. Twenty squares of each of two duplicate grids were examined for each sample.

**Detection of discontinuous virion DNA.** Virion DNA was isolated as described by Lockhart (1990). Purified virion DNA and pScBV20 DNA were digested with either *Bam*HI or *Bam*HI and *Xba*I. Following the addition of 1:5 volumes of a solution containing 95 % formamide, 20 mm-EDTA, 0.05 % bromophenol blue and 0.05 % xylene cyanol FF, the DNA was denatured by heating at 85 °C for 3 min and loaded...
Complete sequence of ScBV

Results

DNA sequence

The complete nucleotide sequence of both strands of pScBV20 (Fig. 1 and 6a), a complete 7568 bp genomic clone of ScBV, was determined. This sequence has been deposited in the GenBank databank under accession number M89923. Following the precedent of other published badnavirus and caulimovirus sequences, numbering of the sequence refers to the plus-strand of the genome and begins at the 5' end of the putative minus-strand replication priming site (see below). The ScBV genome had a G+C content of 43.3%, which is higher than that of CoYMV (39.6%; Medberry et al., 1990) and RTBV (33.7%; Hay et al., 1991; Qu et al., 1991).

Similar to other characterized plant retroelements, the plus-strand of the ScBV genome contained a region, located between nucleotides 1 and 19, that can potentially form base pairs with the 3' end of initiator tRNA^Met (Fig. 1 and 2). These tRNA^Met-binding sites are believed to play a role in caulimovirus and badnavirus replication (Guilfoyle, 1987; Mason et al., 1987; Medberry et al., 1990; Hay et al., 1991). During the replication of these viruses, tRNA^Met is believed to anneal to the tRNA^Met-binding site of the viral transcript and prime the synthesis of the minus-strand. The ScBV primer binding site orientation was similar to those of CoYMV, RTBV and the caulimoviruses, suggesting that it serves a similar function in ScBV.

Coding region

The plus-strand of the ScBV genome contained three ORFs that were capable of encoding proteins with a calculated \( M_r \) larger than 10K (Fig. 1 and Table 1). The minus-strand contained no ORFs capable of coding for proteins larger than 10K. The number, size and arrangement of the ScBV plus-strand ORFs were similar to those of CoYMV. The RTBV genome contains three similar ORFs followed by an additional ORF (ORF IV).

The putative ScBV ORF I protein shared similarity with the putative ORF I proteins of CoYMV and RTBV, and the putative ORF II proteins of all three viruses also shared similarity (Table 2). Comparisons between the deduced amino acid sequences of the proteins encoded by ORFs I and II of ScBV and those contained in the

<p>| Table 1. Protein-coding regions of the ScBV genome and sizes of proteins encoded by CoYMV and RTBV genomes |
|-------------------------------------------------|-----------------|-----------------|-----------------|</p>
<table>
<thead>
<tr>
<th>ORF</th>
<th>ScBV</th>
<th>CoYMV*</th>
<th>RTBV*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Starting ending</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>nucleotide†</td>
<td>nucleotide‡</td>
<td>( M_r )</td>
</tr>
<tr>
<td>I</td>
<td>611 1168</td>
<td>21730</td>
<td>23335</td>
</tr>
<tr>
<td>II</td>
<td>1168 1539</td>
<td>13448</td>
<td>14787</td>
</tr>
<tr>
<td>III</td>
<td>1536 7151</td>
<td>215323</td>
<td>215673</td>
</tr>
<tr>
<td>IV</td>
<td>— —</td>
<td>—</td>
<td>—</td>
</tr>
</tbody>
</table>

* Data on the ORFs of CoYMV and RTBV data are from Medberry et al. (1990), Hay et al. (1991) and Qu et al. (1991).
† Starting nucleotide of the first ATG codon.
‡ Last base of the stop codon.
Table 2. Similarity between the ORFs of ScBV, CoYMV and RTBV

<table>
<thead>
<tr>
<th>Virus</th>
<th>ORF</th>
<th>ScBV</th>
<th>CoYMV</th>
<th>RTBV</th>
</tr>
</thead>
<tbody>
<tr>
<td>ScBV</td>
<td>I</td>
<td>37</td>
<td>26</td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>I</td>
<td>27</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>II</td>
<td>27</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>II</td>
<td>27</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>III</td>
<td>47</td>
<td>26</td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>III</td>
<td>26</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>III</td>
<td>26</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>III</td>
<td>25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>III</td>
<td>24</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>III</td>
<td>23</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>III</td>
<td>18</td>
<td>17</td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>III</td>
<td>17</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>III</td>
<td>49</td>
<td>36</td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>III</td>
<td>49</td>
<td>36</td>
<td></td>
</tr>
<tr>
<td>ScBV</td>
<td>III</td>
<td>34</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CoYMV</td>
<td>III</td>
<td>34</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Percentage identity equals (number of identical amino acids/total number of amino acids) x 100 for the comparable ORF or region.

PIR 31 and Swiss-Prot 20 databanks have identified no significant similarities (data not shown).

Analysis of the ScBV ORF III protein sequence indicated that it was similar to the CoYMV and RTBV ORF III proteins as well as to the ORF I, IV and V proteins of the caulimoviruses (Fig. 3). The first 350 amino acids of the putative ORF III protein contained a region of restricted similarity to the ORF I-encoded intercellular transport proteins of the caulimoviruses (U. Melcher, personal communication; Fig. 3). The most distinctive similarity between the coat protein (ORF IV) of the caulimoviruses and the ORF III proteins of CoYMV, RTBV and ScBV was the presence of a zinc finger-like RNA-binding domain that is common to retroelements (Covey, 1986; Prats et al., 1988). An alignment of this region indicated that the plant pararetrovirus consensus RNA-binding sequence (C-X-C-X-C-X-H-X-C) was larger than the retroelement consensus RNA-binding sequence (C-X2-C-X2-H-X2-C) and included an additional cysteine at the -2 position (Fig. 3).

The ScBV ORF III protein shared similarity with the aspartic protease, reverse transcriptase and RNase H regions of CoYMV, RTBV and the caulimoviruses (Fig. 3). These regions were arranged similarly in all of the plant pararetroviruses (Fig. 4).

The presence of a second cysteine-rich region that did not conform to the RNA-binding region consensus was also noted (Fig. 3 and 4). This cysteine-rich region was located between the RNA-binding and aspartic protease regions. A computer search of the retroelement sequences contained in the PIR 31 and Swiss-Prot 20 databanks indicated that a similar cysteine-rich region was absent from other characterized retroelements (data not shown).

Molecular relationship between plant pararetroviruses

Analysis of the deduced sequences of the putative ORF III proteins of CoYMV, RTBV and ScBV identified four conserved regions that were separated by regions of
variable length (A to D; Fig. 4). Regions A, B, C and D encompass the region of the possible intercellular transport protein, the coat protein region, the cysteine-rich sequence and the reverse transcriptase/RNase H region, respectively. Comparisons between these regions as well as between the ORF I and II protein sequences indicated that the ScBV and CoYMV proteins were more similar to each other than to those of RTBV (Table 2).

Comparisons between the replicases of ScBV, RTBV, CoYMV and of the caulimoviruses suggested that RTBV might be as closely related to the caulimoviruses as it was to ScBV and CoYMV (data not shown). For this reason, the molecular relatedness of a 236 amino acid region of the viral reverse transcriptases and the DNA sequence encoding it were determined. When unrooted trees illustrating the relationship of the viruses were constructed, the DNA and protein sequence analyses produced similar trees. A representative tree based on the analysis of the DNA sequences is shown in Fig. 5. These analyses indicated that ScBV and CoYMV were closely related and formed a cluster that was distinct from the other viruses and that CaMV, FMV and CERV were also a distinct cluster of closely related viruses. RTBV and SoyCMV were quite distinct and occupied positions between the badnavirus and caulimovirus clusters.

The cloned ScBV genome is infectious in banana and rice

Constructs containing 1-3 copies of the CoYMV and RTBV genomes cause infections when introduced into plants by agroinoculation (Medberry et al., 1990; Dasgupta et al., 1991). An analogous ScBV construct called pScBV1 (Fig. 6a) was constructed and used to agroinoculate banana and rice. Banana (M. acuminata) and rice were used instead of sugarcane for three reasons. Firstly, it was difficult to determine with certainty whether any clones of sugarcane, a vegetatively propagated plant, were free of ScBV infection. It has been shown that the vast majority of S. officinarum clones world-wide are infected by ScBV (Comstock & Lockhart, 1990), and analysis by PCR (using primers directed against plant pararetrovirus replicase and tRNA^Met^-binding site) of the apparently virus-free commercial Saccharum hybrid used initially in these experiments suggested that it was also infected. Test plants of rice and M. acuminata were grown from seed and were demonstrated to be virus-free. Secondly, banana streak virus (BSV; Lockhart, 1986) and ScBV have been shown to be closely related serologically, and BSV is reported to infect sugarcane (Lockhart & Autrey, 1988). Additionally, all tested isolates of ScBV infect banana and produce symptoms resembling those of BSV (B.E.L. Lockhart, unpublished results). Finally, ScBV has been found to be transmitted to several cultivars of rice by both mechanical inoculation and mealybug transmission (B.E.L. Lockhart, unpublished results).

To determine whether the cloned ScBV genome was capable of causing an infection in rice and banana,
Table 3. Agroinoculation of banana and rice with A. tumefaciens containing a clone of ScBV

<table>
<thead>
<tr>
<th>Host plant</th>
<th>Number of plants inoculated*</th>
<th>Number of plants infected†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Banana (M. acuminata)</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Rice cv. FK135†</td>
<td>23</td>
<td>4</td>
</tr>
<tr>
<td>Rice cv. ASD7</td>
<td>7</td>
<td>2</td>
</tr>
</tbody>
</table>

* Seedlings were inoculated with A. tumefaciens strain A281 containing pScBV1.
† Infection was scored by testing for the presence of ScBV virions using ISEM.
‡ Results from three separate experiments.

Because denaturation of the virion DNA following digestion with *Bam*H1, which cuts the ScBV genome only once, produced four fragments that were shorter than the ScBV genome, each strand of the virion DNA, like CoYMV and RTBV, must be interrupted by a site-specific discontinuity.

*M. acuminata* infected with the cloned ScBV genome exhibited chlorotic leaf streak symptoms similar to those produced by all tested isolates of ScBV (B.E.L. Lockhart, unpublished results) and by BSV (Lockhart, 1986). The leaves of ScBV-infected rice seedlings exhibited broken chlorotic streaks (Fig. 7). Symptoms were only observed in plants that contained bacilliform virions and ScBV DNA. Plants that did not contain virions or detectable ScBV DNA did not exhibit symptoms. In addition, control plants inoculated with *A. tumefaciens* that did not contain the pScBV1 construct never exhibited these symptoms.

Discussion

The complete sequence of an infective ScBV clone has been determined. The structure of the ScBV genome is identical to that of CoYMV in that both genomes contain a tRNA<sup>Met</sup>-binding site and three ORFs which are similar in arrangement and size. This is in contrast to RTBV, the other sequenced badnavirus, which contains an additional ORF.

The sequence similarities and similar sizes of the respective ORF I-, II- and III-encoded proteins of ScBV, CoYMV and RTBV suggest that the products of comparable ORFs have similar functions. However, it is unclear whether RTBV ORF I is expressed, as it has no ATG codon (Hay et al., 1991; Qu et al., 1991). The function(s) of the putative ORF I and II proteins of these viruses is unknown. The ORF III-encoded proteins are believed to be large polyproteins that are proteolytically processed to yield a protein of unknown function, the virus coat protein, a protease and the viral replicase. The ORF III-derived protein of unknown function shares restricted similarity with plant virus intercellular transport proteins (U. Melcher, personal communication; Fig. 3) suggesting that it may be an intercellular transport protein.

All characterized badnavirus and caulimovirus coat proteins contain an RNA-binding domain that is characteristic of retroelements and is believed to play a role in replication. Interestingly, the first cysteine of the plant pararetrovirus RNA-binding region consensus (C-X-C-X<sub>2</sub>-C-H-X<sub>4</sub>-C; Fig. 3) is not present in any of the other retroelement RNA-binding domain sequences contained in the Swiss-Prot 20 or PIR 31 databanks. The presence of this extra cysteine in the RNA-binding domain of all sequenced plant pararetroviruses suggests
that it may be important to the function of this region, and that there may be functional differences between the RNA-binding domains of plant pararetroviruses and those of other retroelements.

All of the features of the ScBV genome that are described above suggest that ScBV, like CoYMV and RTBV, is a pararetrovirus.

Analysis of the sequences of the ORF III protein sequences has identified a second cysteine-rich region that is located downstream of the RNA-binding domain, and does not conform to the consensus sequence of RNA-binding domains (Fig. 3). Searches of the protein sequence databanks have identified no proteins containing cysteine-rich regions that conformed to the consensus sequence of this region, suggesting that this region may be unique to the badnaviruses. The function, if any, of this region is unknown. However the spacing of the cysteine residues in this region suggests that this region could be a second zinc finger. The RNA-binding domain of some retroviruses consists of two zinc fingers; however, both zinc fingers conform to the RNA-binding domain consensus sequence (Berg, 1986).

Both the DNA and amino acid sequences of a portion of the sequenced plant pararetrovirus reverse transcriptases have been used to determine the molecular similarity of these viruses. Within the caulimoviruses, CaMV, CERV and FMV form a distinct cluster, whereas amongst the badnaviruses, CoYMV and ScBV form a distinct cluster. Interestingly, RTBV and SoyCMV occupy positions distinct from the main badnavirus and caulimovirus clusters, and the genetic structure of both genomes also distinguishes these viruses from other members of their respective groups. The genomes of both viruses contain more ORFs than do the genomes of other group members (Hasagawa et al., 1989; Hay et al., 1991; Qu et al., 1991).

The majority of mealybug-transmitted plant badnaviruses infect a restricted range of host plants, sometimes limited to members of a single genus. ScBV and cacao swollen shoot virus (Brunt, 1970) are exceptional in infecting host plants in at least two different families. In addition to banana, rice and sugarcane, ScBV also infects sorghum (B.E.L. Lockhart, unpublished results), and it is possible that the full host range is even more extensive. Given the wide distribution of ScBV in sugarcane germplasm (Comstock & Lockhart, 1990), the ubiquitous occurrence of its mealybug vectors and the range of plant species susceptible to infection by this virus, greater attention should be given to the presence and movement of ScBV in sugarcane germplasm. It is of interest to note that the symptoms of a mealybug-transmitted chlorotic streak disease of rice in India (Anjaneyulu et al., 1980) closely resemble those produced by the ScBV clone used here.

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References


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