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Sweetpotato Viruses: 15 Years of Progress on Understanding and Managing Complex Diseases

Sweetpotato (*Ipomoea batatas* (L.) Lam.) is a member of the *Convolvulaceae* (morning glory family) that is thought to have originated in Central or South America but also has a secondary center of diversity in the southwest Pacific islands (95). It is grown in all tropical and subtropical areas of the world and consistently ranks among the 10 most important food crops worldwide on the basis of dry weight produced, yielding about 130 million metric tons per year on about 9 million hectares. The most intensive areas of production are in China and around the Great Lakes of East Africa, but it is also important in many other countries (95).

Sweetpotato is an important crop for food security (59). It has been relied on as a source of calories in many circumstances, such as in Japan when typhoons have destroyed rice crops, during the depression of the 1930s in the United States, during famines in China in the 1960s, or when cassava was decimated by *East African cassava mosaic virus* in East Africa in the 1990s. Vines and/or storage roots can be used for direct human consumption or animal feed. Storage roots can also be used for fermentation or as a source of starch for food processing or production of chemical stocks, including organic acids (95). It is valued by subsistence farmers because it can produce a crop with few production inputs, withstands stresses such as drought, in the absence of frost it can be left in the field to harvest when needed, and it can also be sold for cash (75) (Fig. 1). In the United States, sweetpotato is considered to have high production costs, largely due to the cost of labor, but also has the potential for good profitability, as the price for fresh market sweetpotatoes is usually relatively high (33).

Growing awareness of health benefits attributed to sweetpotato has stimulated renewed interest in the crop. Orange-fleshed cultivars, a source of vitamin A, were introduced to developing countries with hope that they would replace the white-flesh varieties and help alleviate vitamin A deficiencies (95). Expanded availability of new sweetpotato products, such as chips, French fries, or other frozen products, has further enhanced demand in industrial countries. As a result, there is greater international trade in sweetpotatoes, especially to European destinations.

Sweetpotato can thus be viewed from very different perspectives: in some regions as a crop valued because it requires few inputs to support subsistence, and in others as a health food with good potential for financial profit despite high production costs. Subsistence growers cannot afford to lose to viruses yield that would feed their families, and large-scale commercial vegetable growers must maximize return on investment to make a profit.

The sweetpotato plant is an indeterminant perennial, but it is grown as an annual. It is produced by vegetative propagation using vine cuttings from production fields or from sprouted storage roots ('seed') (95). As for other crops produced by vegetative propagation, it may accumulate pathogens, particularly viruses, in the planting stock, resulting in decline in yield and sometimes quality

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of the crop. While this is a common feature of production, the nature of the virus complexes that occur in different regions and the approaches taken to manage these problems have been quite varied. In temperate zone production, the crop is generally affected by a complex of potyviruses and possibly other unknown viruses that typically cause yield reductions on the order of 20 to 40% (31,33). In East Africa, sweetpotato virus disease (SPVD), which is caused by the synergistic interaction of the whitefly-transmitted crinivirus, *Sweet potato chlorotic stunt virus* (SPCSV), and the aphid-transmitted potyvirus, *Sweet potato feathery mottle virus* (SPFMV), can cause losses of 80 to 90% in many high-yielding genotypes (75) (Fig. 2). Up until the mid-1990s, most work on sweetpotato viruses focused on SPFMV, relying on biological methods for detection and identification, with little appreciation for the complex interactions occurring among multiple viruses infecting the crop. During the past 15 years, as molecular methods have been adopted, much has been learned about the composition of the sweetpotato virus complexes, the effects of virus diseases on production systems, the biology of the virus–plant interaction, and management approaches to sweetpotato virus diseases. This article is intended to summarize what has been learned since earlier reviews (75,111,159), integrate knowledge gleaned from experiences in tropical and temperate production systems, and suggest courses of action to develop sustainable management programs for these diseases.

Recent Progress in Virus Characterization

Over 30 viruses infecting sweetpotato, assigned to 9 families, have been identified (Table 1): *Bromoviridae* (1 virus), *Bunyaviridae* (1), *Caulimoviridae* (3), *Closteroviridae* (1), *Comoviridae* (1), *Flexiviridae* (1), *Geminiviridae* (15), *Luteoviridae* (1), and *Potyviridae* (9). Half of them are recently described DNA viruses belonging to families *Geminiviridae* and *Caulimoviridae*. Most of these viruses are associated with symptomless infections in sweetpotato and in some cases even in the indicator plant *Ipomoea setosa.* Some are synergized by SPCSV, the mediator of severe virus diseases in sweetpotato, while others apparently are not. Therefore, sweetpotato DNA viruses appear to be as diverse in sequence and in their interactions as has been previously observed with RNA viruses. Although new, improved generic virus detection methods based on new DNA sequencing technologies (103) and the use of rolling circle amplification (RCA) are advancing the process of virus characterization at the molecular level, the biological characterization and validation of newly identified isolates is lagging behind. Until recently, most surveys of sweetpotato viruses paid little attention to DNA viruses (159). Major advancements in the molecular characterization of the groups of viruses are summarized in the following sections.

Crinivirus: *Sweet potato chlorotic stunt virus.* *Sweet potato chlorotic stunt virus* (SPCSV; genus *Crinivirus*) has been detected in all sweetpotato producing areas except the Pacific region (96,112,150). Different laboratories have shown the synergistic effect that distinct isolates of SPCSV have on co-infecting unrelated viruses (43,78,113,156), and dramatic reductions in yield have been reported associated with mixed infections that include SPCSV (46,57,160).

SPCSV can be differentiated into two distantly related strains, East African (EA) and West African (WA), based on serology and nucleotide sequence data (66,150,162). The WA strain occurs worldwide except apparently in East Africa, where only the EA strain has been found. The EA strain is also found in Peru, where it cooccurs with the WA strain (61). A third strain, more closely related to WA than to EA, is found predominantly in South America, but also in Rwanda and Burundi (International Potato Center [CIP], *unpublished results*). Only two complete SPCSV genomes have been reported and both correspond to isolates of the EA strain (40,86).

The first sequenced isolate of SPCSV revealed several novel features for criniviruses, such as nearly identical 3′UTRs on both genomic RNAs and two novel ORFs encoding a Class 1 dsRNA-specific RNase III enzyme (RNase3) and a p22, both able to suppress RNA silencing (42,85,86). RNase3 is most similar to plant RNase III enzymes and was probably acquired through recombination with host genes. Genome expansion through acquisition of host genes seems to have occurred frequently during the evolution of closteroviruses (47). Recent acquisition of the p22 gene seems likely because most isolates of SPCSV characterized lack the p22 gene, which has to date only been found in isolates from Uganda (40,43; A. K. Tugume, *unpublished*). Differences in host range between isolates containing or lacking p22 are not known, but those carrying p22 seem to be more virulent in the indicator host *I. setosa* (43).

Fig. 1. Sweetpotatoes for sale in a market in Salomon Islands. (Photo by Segundo Fuentes)

Fig. 2. Symptoms of sweetpotato virus disease (SPVD), caused by interaction of *Sweet potato chlorotic stunt virus* and *Sweet potato feathery mottle virus,* on a sweetpotato plant in Mozambique alongside an unaffected plant. (Photo by Segundo Fuentes)

Table 1. Viruses reported from sweetpotato, their family, genus, known vectors, methods of detection, genomic target region for detection, and references providing additional information on detection

Species name	Acronym	Family	Genus	Vector	Detection method	Genomic target region[a]	References
Cucumber mosaic virus	CMV	*Bromoviridae*	*Cucumovirus*	Aphid			35
Ipomoea crinkle leaf curl virus	ICLCV	*Geminiviridae*	*Begomovirus*				36
Ipomoea yellow vein virus	IYVV	*Geminiviridae*	*Begomovirus*	Whitefly	PCR, PCR/RFLP	Coat protein	91,101
Sweet potato pakakuy virus (synonyms Sweet potato badnavirus A and B)[b]	SPPV	*Caulimoviridae*	*Badnavirus*		PCR	Reverse transcriptase	84,104
Sweet potato C-3 virus		*Bunyaviridae*	*Phlebovirus?*				51,96, CIP, *unpublished*
Sweet potato C-6 virus		*Flexiviridae*	*Carlavirus?*				96, CIP, *unpublished*
Sweet potato collusive virus (synonym Sweet potato caulimo-like virus)[b]	SPCV	*Caulimoviridae*	*Cavemovirus*		PCR	Reverse transcriptase	41,45
Sweet potato chlorotic fleck virus	SPCFV	*Flexiviridae*	*Carlavirus*		RT-PCR	Coat protein, 3′UTR	9,72
Sweet potato chlorotic stunt virus	SPCSV	*Closteroviridae*	*Crinivirus*	Whitefly	RT-PCR, IC-RT-PCR, RT-qPCR	HSP70h	40,43,79,86,88, 105,112,113, 134,143,150
Sweet potato feathery mottle virus	SPFMV	*Potyviridae*	*Potyvirus*	Aphid	RT-PCR, RT-PCR/RFLP, IC-RT-PCR, RT-qPCR[c]	Coat protein, NIb-CP region	27,72,79,82,88, 105,113,114,12 4,128,134,141
Sweet potato Golden vein associated virus	SPGVaV	*Geminiviridae*	*Begomovirus*		[a]		132
Sweet potato latent virus	SPLV	*Potyviridae*	*Potyvirus*	Aphid	RT-PCR[c]	Coat protein	38,39,88,166
Sweet potato leaf curl virus	SPLCV	*Geminiviridae*	*Begomovirus*	Whitefly	PCR, PCR/RFLP, qPCR[a]		79,88,91,98,101, 134,145, 169
Sweet potato leaf curl Canary virus	SPLCCaV	*Geminiviridae*	*Begomovirus*	Whitefly	[a]		101
Sweet potato leaf curl China virus	SPLCV-CN	*Geminiviridae*	*Begomovirus*		[a]		102
Sweet potato leaf curl Georgia virus	SPLCGV	*Geminiviridae*	*Begomovirus*	Whitefly	PCR, PCR/RFLP[a]	Coat protein	98
Sweet potato leaf curl Lanzarote virus	SPLCLaV	*Geminiviridae*	*Begomovirus*	Whitefly	[a]		101
Sweet potato leaf curl Spain virus	SPLCESV	*Geminiviridae*	*Begomovirus*	Whitefly	[a]		101
Sweet potato leaf curl South Carolina virus	SPLCSCV	*Geminiviridae*	*Begomovirus*	Whitefly	qPCR		94,175
Sweet potato leaf curl Uganda virus	SPLCUV	*Geminiviridae*	*Begomovirus*	Whitefly	RT-PCR, RT-qPCR[a]	Coat protein	169
Sweet potato leaf speckling virus	SPLSV	*Luteoviridae*	*Polerovirus?*	Aphid	PCR	Coat protein	50
Sweet potato mild mottle virus	SPMMV	*Potyviridae*	*Ipomovirus*		RT-PCR, IC-RT-PCR, RT-qPCR		37,88,112,113
Sweet potato mild speckling virus	SPMSV	*Potyviridae*	*Potyvirus*	Aphid	RT-PCR[c]		88
Sweet potato mosaic associated virus	SPMaV	*Geminiviridae*	*Begomovirus*				132
Sweet potato ringspot virus		*Comoviridae*	*Nepovirus*				21
Sweet potato symptomless virus 1	SPSMV-1	*Geminiviridae*	*Mastrevirus*		PCR	Movement, coat protein gene	84,104
Sweet potato vein mosaic virus	SPVMV	*Potyviridae*		Aphid			126
Sweet potato virus 2	SPV2	*Potyviridae*	*Potyvirus*	Aphid	RT-PCR, RT-qPCR[c]	Coat protein	13,71,79,94,134
Sweet potato virus C	SPVC	*Potyviridae*	*Potyvirus*	Aphid	RT-PCR/RFLP, RT-qPCR[c]		79
Sweet potato virus G	SPVG	*Potyviridae*	*Potyvirus*	Aphid	RT-PCR, RT-qPCR[c]	Coat protein	39,79,88,94,134, 136,147,173
Sweet potato vein clearing virus	SPVCV	*Caulimoviridae*	*Solendovirus*[b]		PCR	Reverse transcriptase	41
Sweet potato yellow dwarf virus	SPYDV	*Potyviridae*	*Ipomovirus*				92

[a] Broad-spectrum detection methods for sweepoviruses have also been used for several sweetpotato sweepoviruses using polymerase chain reaction (PCR), PCR/restriction fragment length polymorphism (RFLP), or rolling-circle amplification (RCA)/RFLP and have targeted a conserved region in open reading frame (ORF) AC2 and AC1 or coat protein (3,64,91,101,132,169).

[b] Virus/genus name recently accepted by the ICTV, still under process of ratification.

[c] Broad-spectrum detection methods for potyviruses have also been used for several sweetpotato potyviruses using reverse transcription (RT)-PCR for the NIb to 3′NTR region (14,38,39,54,133,136,147,150,155).

Potyviruses. In the past few years, sweetpotato virus surveys have revealed a wider distribution of potyviruses than previously known. *Sweet potato virus G* (SPVG) and SPFMV, both belonging to the genus *Potyvirus*, were previously known to occur in China, Africa, and the United States but were recently found in modern sweetpotato varieties and landraces in Australia, New Zealand, and the Pacific region (136,137).

Several recent studies have indicated that recombination occurs frequently between potyviruses and thus may be a driving force in their evolution. Isolate 10-O of SPFMV seems to be a triple recombinant between O (ordinary), EA (East African), and RC (russet crack) strains of SPFMV (157). It has also become clear that viral genotypes, such as the EA strain of SPFMV (82), is geographically more widespread than previously thought (152).

Sequence analyses suggest that interviral recombination has occurred between SPFMV and *Sweet potato virus* C ([SPVC] originally known as the C (common) strain of SPFMV). SPFMV, SPVC, SPVG, *Sweet potato virus 2* (SPV2), and an as yet unclassified sequence from Zimbabwe form a well-supported phylogenetic lineage (designated the 'SPFMV-group') of *Ipomoea*-infecting potyviruses with a probable common origin (155). An extensive study of SPFMV isolates from cultivated sweetpotato and wild relatives in Uganda indicated that recombination between virus isolates is frequent, but interspecific recombination was not observed (152). Results also showed that SPFMV populations from wild or cultivated species in Uganda were genetically undifferentiated, indicating frequent interhost transmission (152). Similar results were found for *Sweet potato mild mottle virus* (SPMMV) (153).

SPFMV and SPMMV have an N-terminal part of the P1 protein (P1-N) that is similar in the sense that it contains a region of high genetic diversity (153,158). P1-N of SPFMV is similar only to the homologous region in SPMMV, whereas the C-proximal part of P1-pro of SPFMV is similar to the P1 of many other potyviruses (157). Sequencing of the complete genomes of SPVG, SPV2, and *Sweet potato latent virus* (SPLV) has confirmed that P1-N is unique to the 'SPFMV-group' of potyviruses and SPMMV, but is not found in other potyviruses including SPLV (CIP, *unpublished*). We speculate that the P1-N domain provides some specific benefit to potyviruses infecting *Ipomoea* spp. The SPMMV P1-N is an RNA silencing suppressor which mediates its action by binding Argonaute through conserved WG/GW motifs (60,153). Although the 'SPFMV-group' potyvirids also contain less conserved WG/GW motifs in their P1-N, another ORF (PISPO) found nested within the P1-Pro domain and preceded by similar nucleotide motifs as the frameshift protein PIPO of potyviruses (28) has much more conserved WG/GW motifs and presents another unique feature of this group of potyvirids (CIP, *unpublished*). How such a big nested ORF could evolve within the existing ORF of P1-Pro is fascinating, since the same region is riddled with stop codons in non-'SPFMV-group' potyviruses (Fig. 3).

Geminiviruses. Occurrence of begomoviruses in sweetpotato is widespread, and as awareness of these viruses increased, many new species were discovered (99–102,132) (Table 1). Furthermore, at CIP, 10 to 20% of sweetpotato accessions originating in different parts of the world test positive for begomoviruses (CIP, *unpublished*). Several tentative new begomovirus species have been re-

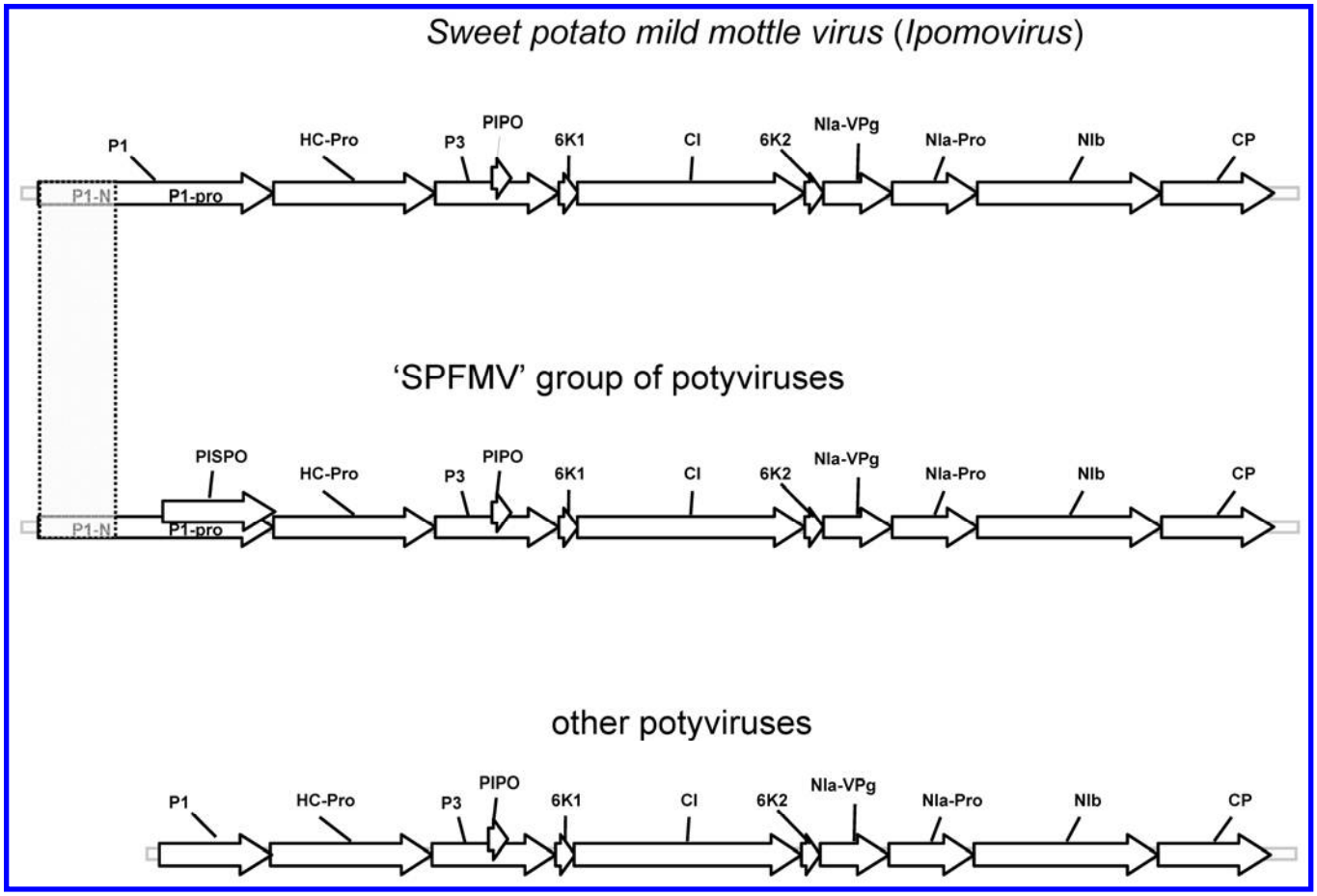

Fig. 3. Comparison of genome organization of the *Sweet potato feathery mottle virus* (SPFMV) group of potyviruses with *Sweet potato mild mottle virus* (SPMMV) and other potyviruses. The genome contains short noncoding regions at both ends. Open box arrows indicate various mature proteins initially produced as parts of a single polyprotein encoded by the open reading frame (ORF) of the virus. Names of proteins are indicated above or in arrows. The P1 protein in the 'SPFMV-group' of potyviruses and SPMMV can be divided into an N-terminal region (P1-N), which is absent from other potyvirids, and P1-pro (proteinase), which is in common with other potyvirids. In addition, 'SPFMV-group' potyviruses contain an internal nested ORF (Pretty Interesting Sweetpotato Potyvirus ORF: PISPO) within the P1-pro region, in contrast to any other known potyvirid. Other abbreviations: P1: protein 1; HC-Pro: helper component proteinase; P3: protein 3; PIPO: pretty interesting potyvirus ORF; 6K1: 6K protein 1; CI: cylindrical inclusion protein; 6K2: 6K protein 2; NIa: nuclear inclusion protein a; -VPg: viral protein genome linked; -Pro: proteinase; NIb: nuclear inclusion protein b; CP: coat protein.

ported (Table 1), as have defective begomovirus sequences, composed of incomplete genome copies, most containing the *rep* gene, a region commonly used in diagnosis and comparison of different isolates (132). Biological properties of many of these isolates are still unknown. For SPLCV, efficient whitefly transmission (145,161), experimental host range, and the presence of natural weed plants (93) have been characterized.

Sweetpotato-infecting begomoviruses are phylogenetically distinct from the new and old world begomoviruses, and are called 'sweepoviruses' as a group (20,48,169). Sequence analysis of complete genomes shows that recombination is frequent among distinct species (101,132,175). Nevertheless, *Ipomoea yellow vein virus*, which can infect sweetpotato, has been isolated only from *I. indica* in nature (17,101), suggesting some adaptation to specific hosts may be occurring. Because the sweepoviruses are mostly symptomless in sweetpotato, even in double infection with SPCSV (169), but are able to attain high titers, they can spread undetected with sweetpotato germplasm to new areas.

The putative sweetpotato-infecting mastrevirus, Sweet potato symptomless virus 1 (SPSMV-1), appears quite unique compared to other mastreviruses in that it has a much smaller replicase gene (Fig. 4). The virus has been detected by polymerase chain reaction (PCR) from sweetpotatoes collected in Peru (84), Tanzania (104), and several Central American and Asian countries (CIP, *unpublished*) but seems far less common than badnaviruses. Comparison of a Tanzanian and a Peruvian isolate of SPSMV-1 showed 100% identity over the sequenced CP-MP region.

Caulimovirids (Pararetroviruses). Sweetpotato badnaviruses have been characterized recently from mixed infected and nonsymptomatic plants (84). Sim et al. (144) also reported badnavirus-like particles from sweetpotato, but whether these are closely related has not been confirmed (159). Currently, sequences corresponding to related, but genetically distinct groups of badnaviruses have been found in sweetpotato material collected from all over the world including Africa (104) (Table 1) (CIP, *unpublished*), and the name Sweet potato pakakuy virus (SPPV) is in the process of ratification for the two first described variants (84).

The caulimovirid Sweet potato collusive virus (SPCV, synonymous Sweet potato caulimo-like virus) was isolated for the first time in 1987 (15), and since then it has been reported infecting sweetpotatoes from Madeira, New Zealand, Papua New Guinea, China, Central America, and East Africa (96). Recent partial purification and PCR primers targeting the replicase region of SPCV have allowed the characterization of the virus sequence, revealing its identity as a member of genus *Cavemovirus* (45). Another caulimovirid, Sweet potato vein clearing virus (SPVCV), was isolated from a sweetpotato sample from the Dominican Republic. It has a different genome organization than SPCV (41) and falls into the recently accepted new genus *Solendovirus*. Both SPCV and SPVCV interact synergistically with SPCSV; however, they have not yet been reported in natural co-infection with SPCSV. All SPVCV isolates analyzed so far group together. They have been found to be common in Central America and the Caribbean islands. In contrast, SPCV isolates from Africa form a phylogenetic subgroup separated from isolates from the Americas (41). Insect vectors have not yet been identified for any members of the *Cavemovirus* or *Solendovirus* genera, but they are not mechanically transmitted. They could have become widespread by vertical transmission through vegetative propagation. The absence of these caulimovirids in the sweetpotato germplasm collected in South America, one of the centers of origin of sweetpotato, is intriguing.

Since sequences of these pararetroviruses have not been detected integrated into the genome of sweetpotato or *Ipomoea setosa* (CIP, I. Weinheimer, J. Valkonen et al., *unpublished*), it appears they are systemic. Their impact on the growth and yield of sweetpotato is unknown.

Carlaviruses. *Sweet potato chlorotic fleck virus* (SPCFV) was isolated in 1992 from the CIP germplasm collection, and different isolates were recently partially sequenced (9). A complete genome from a Ugandan isolate (10) has been characterized, indicating that

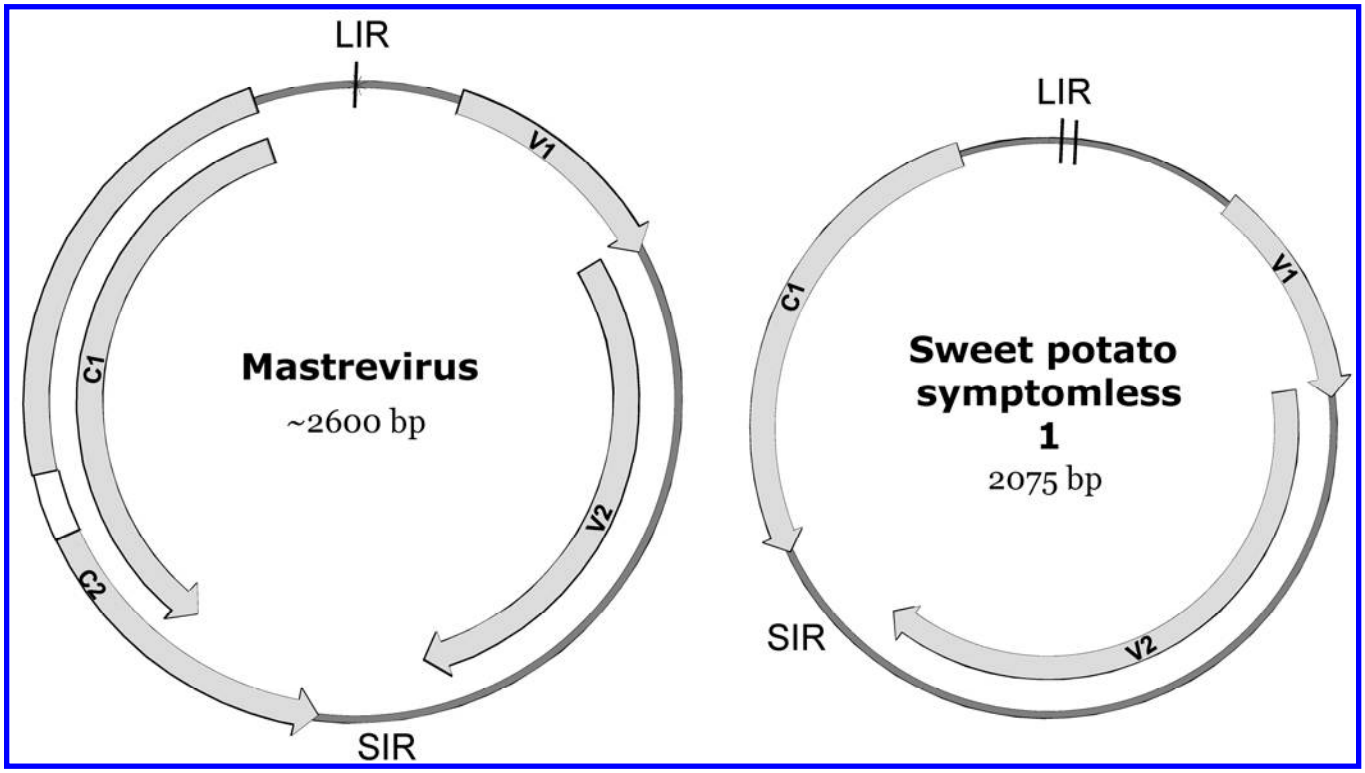

Fig. 4. Comparison of organization of Sweet potato symptomless virus 1 (SPSMV-1) genome with genome typical of other plant mastreviruses. Genomes are represented by dark gray circular lines with open box arrows indicating coding regions of various proteins with their name inside. Thin black lines crossing the genome indicate the position of a predicted hairpin loop. C1 and C2 proteins of mastreviruses are produced through alternative splicing (position of intron indicated by white open box in C2) leading to two proteins with the same N-terminus but different C-termini. SPSMV-1 genome is considerably smaller than that of other mastreviruses, resulting from lack of an alternatively spliced C2 protein. In addition, SPSMV-1 has two predicted stem-loop structures in the large noncoding region. V1 (viral sense protein 1): movement protein; V2 (viral sense protein 2): coat protein; C1 (complementary sense protein 1): replication associated protein A; C2 (complementary sense protein 2): replication associated protein.

the virus is a new member of the genus *Carlavirus* (family *Flexiviridae*). It has been reported from South and Central America and Asia (96), East Africa (9,112,149), Australia (71), and French Polynesia (137). Phylogenetic analysis showed geographical differentiation among the isolates. One major cluster was formed by isolates from East Africa and SPCFV-CIP (9).

C-6 virus has been detected in Cuba, Mexico, Peru, and the United States (34,96). The virus particles are flexuous rods that react weakly with an antiserum against *Potato virus S* (genus *Carlavirus*), suggesting that C-6 is related to carlaviruses (96). Sequence analysis of the CP region (C-6 isolate from the Dominican Republic) indicates only a low amino acid sequence similarity to other carlaviruses found in GenBank and only 55% aa sequence similarity with SPCFV, indicating that C-6 is a distinct carlavirus species (CIP, *unpublished*).

Other viruses. Virus-like particles and diseases of unknown etiology have been reported in sweetpotato collected in different parts of the world (10,112,144,149,160,164). Partial sequences of the virus C-3 indicate that this virus belongs to the *Bunyaviridae* family (CIP, *unpublished*). Only one isolate has been reported so far in a sweetpotato plant from Brazil (51). It can infect *I. setosa, I. nil,* and *Nicotiana benthamiana* (96), but no vector has been identified. *Sweet potato leaf speckling virus* (SPLSV) has been reported from Peru and Cuba (50,96). SPLSV is rarely detected, possibly due to lack of antisera for indexing.

Cucumber mosaic virus (CMV) has been found infecting sweetpotato in Israel (35). CMV only infected sweetpotato if the plants were first infected with SPCSV (36), suggesting that SPCSV acted as a helper virus. However, an isolate of CMV from *Arracacia xanthorrhiza* infected sweetpotato in the absence of SPCSV (156), and CMV was detected in sweetpotatoes infected in the field in Egypt (68) without a known helper virus. CMV is synergized by SPCSV (156), which is of epidemiological importance because it affects virus accumulation in infected plants and may enhance transmission of CMV by aphids in countries such as Egypt and Israel where both CMV and SPCSV occur in sweetpotatoes (69).

Sweetpotato ringspot virus (SPRSV) was detected in Papua New Guinea (21) and is the only sweetpotato virus sharing some physicochemical properties with members of the genus *Nepovirus*. However, the taxonomic position of SPRSV has not been determined based on serological relationships or sequence data. The virus has a wide host range including *Ipomoea*, *Chenopodium,* and *Nicotiana* species.

Contribution of Wild Species to Sweetpotato Virus Epidemiology

Wild plants can function as natural reservoirs for sweetpotato-infecting viruses. SPFMV has been detected in the perennial species *I. trichocarpa* and the annual species *I. hederacea*, *I. hederifolia*, *I. lacunosa*, and *I. wrightii* in Louisiana, USA (30). In Uganda, 24 wild plant species of family *Convolvulaceae* (genera *Ipomoea*, *Hewittia*, and *Lepistemon*) growing in different agro-ecological zones were infected with SPFMV (75,154). These plants were infected with different strains of SPFMV: EA, O, and C (now SPVC), but not RC, which has not been reported in East Africa (152). A survey of wild plants in the coastal areas of Syria suggested that 19 species belonging to mostly *Chenopodiaceae* and *Convolvulaceae* may be new natural hosts of SPFMV (2).

In Uganda, SPMMV was found in 21 of the 27 wild species tested, all of which were previously unknown natural hosts of SPMMV (153). The incidences of plants sero-positive for SPFMV and SPMMV were 17 and 9.8%, respectively. The symptoms observed on the plants were associated with SPFMV infection with a 77% probability (154). SPCSV and SPCFV have also been detected in some of the wild plant species, but their incidences seem to be much lower than SPFMV or SPMMV (A. K. Tugume et al., *unpublished*). In Spain, 60% of the tested plants of *I. alba* (synonym *I. acuminata*) were infected with different sweepoviruses, of which three were new (101). SPLCV was transmitted by whiteflies to 38 of 45 species in the genus *Ipomoea,* and several wild species were reservoirs of SPLCV in the United States (93).

In Uganda, SPFMV incidences in the wild host plants were the same in annual and perennial species (154). Because SPFMV is not known to be seed-transmitted in sweetpotato or any wild plant species (170), aphid transmission of SPFMV must occur between the wild species and sweetpotatoes. The coat protein of all characterized SPFMV isolates, regardless of host, contained the DAG motif (152) needed for aphid transmissibility of potyviruses (16). Moreover, isolates of SPFMV from wild plants and sweetpotatoes were phylogenetically indistinguishable, and were genetically undifferentiated (152). Similar observations were made for SPMMV (153), although vector transmission of SPMMV is not well understood. The lack of spatial and temporal separation between populations of wild host plants and cultivated sweetpotatoes in Uganda (Fig. 5) is an important factor enhancing virus exchange between natural and agro-ecosystems (154). Taken together, close proximity of wild plants and cultivated hosts and frequent infections of both with the same genotypes of SPFMV and SPMMV emphasize the significance of wild plants as virus reservoirs.

Sweetpotato crops may also be a source of virus to wild plants. SPFMV incidences in wild plants from different agro-ecological zones of Uganda were negatively correlated with incidences of SPFMV in cultivated sweetpotato (154). This finding suggests that wild species may have developed resistance to SPFMV infection under continuous infection pressure from sweetpotato. Resistance to SPFMV was reported in some genotypes of wild plants from central Uganda (76), where there is a relatively low incidence of SPFMV in wild species but a high infection pressure from sweetpotato fields. Alternatively, SPFMV may reduce the survival of wild hosts as documented for some other viruses in non-domesticated plants (152,154).

Detection Methods

Precise and rapid detection of viruses is essential to their timely management. Sweetpotatoes present a special challenge since the virus titers are often very low and the concentrations of inhibitors are high, interfering especially with serological or PCR-based detection methods. Traditionally, sweetpotatoes have been indexed for the presence of viruses by grafting to the Brazilian morning glory, *I. setosa*. This consumes considerable time and greenhouse resources and does not reveal the identity of the viruses present. In recent years, viruses have been detected by a combination of serological and nucleic acid–based assays, but in many cases, virus titers are so low that these methods must be combined with prior grafting to *I. setosa* or other hosts.

Fig. 5. *Ipomoea hederifolia* (red flowers) growing next to sweetpotato crop in Kamuli, Eastern Uganda. (Photo by Arthur K. Tugume)

The well-known serological methods such as enzyme-linked immunosorbent assay (ELISA) carried out on a nitrocellulose membrane or a microtiter plate are widely used for detection of sweetpotato viruses. CIP provides a diagnostic kit for serological detection of a number of the most common sweetpotato RNA viruses. However, we will focus attention on the molecular detection methods that have been introduced recently and are used in many laboratories due to their greater sensitivity. Molecular methods also facilitate obtaining more information about the virus isolates by analysis and comparison of viral sequences with previously characterized isolates. Recent introduction of the 'fast technology for analysis of nucleic acids' (FTA) paper as a carrier that absorbs plant sap and protects nucleic acids against nucleases has provided a new, handy means for sampling (138) and enhances the use of PCR-based methods in plant virus diagnostics.

PCR-based approaches. Characterization of many viruses at the sequence level allows their detection using PCR methods (Table 1). For routine detection assays, PCR requires degenerate primers for detection of the variants and strains of the virus. Broad-spectrum PCR and subsequent restriction fragment length polymorphism (RFLP) analysis or sequencing of the amplification products have been used as the primary approach for the rapid confirmation and differentiation of potyvirus and sweepovirus infections (Table 1). Sweetpotato contains an inhibitory compound(s) that can affect sensitivity and reliability of virus detection by PCR (49). Thus, an internal standard is needed to assure the quality of the nucleic acid extracts used in PCR assays. The malate dehydrogenase (MDH) gene of host plants can be used for this purpose (91). Commercial kits for nucleic acid extraction are used commonly for PCR-based detection (3,40,43,84,91,128,132,136, 140,155,166,169,175). However, extraction kits are too expensive for use in large-scale processing of samples. Use of cetyl trimethylammonium bromide (CTAB) and a semi-automatic homogenizer (90) or LiCl (S. Fuentes, *unpublished*) for nucleic acid extraction are inexpensive and reliable alternatives. NaOH-Tris protocol is also a quick and cheap method for extracting viral DNA (41,45,165). Immuno-capture reverse transcription PCR (IC-RT-PCR) has been used to capture virus particles for PCR analysis and to avoid the laborious methods of nucleic acid isolation from plant tissue (87,112).

RT-PCR with universal degenerate primers and strain-specific primers has allowed detection and characterization of many potyviruses infecting sweetpotatoes (14,27,38,39,54,147). The crinivirus SPCSV can be detected with universal primers that amplify a portion of the gene that encodes the heat shock protein 70 homologue (HSP70h) present in all known members of the family *Closteroviridae* (143). Generic and virus-specific primers have been used to detect and identify sweepoviruses in in vitro plantlets and greenhouse-grown sweetpotato plants, and in indicator *Ipomoea* plants (17,91,99,101,132,169). PCR with virus-specific primers for the reverse transcriptase encoding region of SPCV and SPVCV allowed rapid detection of caulimovirids (41,45).

Accumulation of virus sequence information has enhanced the design of virus taxon-specific primers and probes, specific detection of viruses, verification of the results (39,114,147), and significantly improved resolution of incompletely characterized viruses, e.g., SPV2 (13), SPCFV (6), and SPVC (114). RT-PCR/RFLP has been used to detect and distinguish strains of SPFMV and SPV2 (88,141,148,152,172). Simultaneous detection of SPFMV, SPCSV, and SPMMV is possible by multiplex RT-PCR in a one-step or two-step reaction procedure (131,140). Progress is being made in the simultaneous detection of DNA viruses (badna-, begomo-, cavemo-, and solendovirus) by PCR using more than one primer pair (multiplex PCR) (CIP, *unpublished*).

Although PCR-based methods can amplify viruses present in low titers, false negative reactions with known infected plants have been found with potyviruses (147) and sweepoviruses (79,91,169).

Quantitative, real-time PCR. Quantitative, real-time PCR (qPCR) is an accurate and sensitive method for detection of viruses in plant tissues. In this method, amplification of mRNA or coding sequence of a host gene needs to be included as an internal control in detection of RNA and DNA viruses, respectively. Sweetpotato genes including the 18S (105) and 26S rRNA genes (113), the cytochrome oxidase (*COX*) gene (169), and plant mitochondrial NADH dehydrogenase (*Nad5*) gene (89) have been used as internal controls in the qPCR assays to normalize differences in RNA and DNA concentrations between samples.

Several RNA viruses (SPCSV, SPFMV, SPMMV, SPVG, and SPV2) and one DNA virus (SPLCV) have been detected and quantified directly from infected sweetpotato plants (79,105, 113,134). Interfering substances were not observed in the total nucleic acid extracts from leaves, and qPCR was more efficient and sensitive (perhaps 1,000-fold) in detecting sweepoviruses (79) compared to a conventional PCR (97). The use of *I. setosa* plants as an indicator of SPLCUV infection in sweetpotato plants was as efficient as using qPCR, while the sweepovirus was only detected at relatively low efficiency by conventional PCR (169). Efficiency of transmission and retention of SPLCV by the whitefly, *Bemisia tabaci,* has been determined by qPCR (145). To reduce the cost of the simplex qPCR assays (when only one primer pair and probe is used), Ling et al (94) modified and improved qPCR efficiency to detect three potyviruses (SPFMV, SPVG, and SPV2) in a multiplex assay by mixing primers and using probes labeled with distinct fluorophores and also developed generic primers and TaqMan probes to detect all sweepoviruses whose sequences were available from GenBank.

Because RT-qPCR may offer a more rapid alternative as a screening method for detection of SPFMV and differentiation of SPCSV strains, primers and TaqMan probe sets were designed for the SPCSV *Hsp70h* gene and SPFMV CP encoding sequence by CIP in collaboration with the Food and Environment Research Agency, UK. The universal assays for both SPCSV and SPFMV and the strain-specific assays developed for SPCSV performed well in a simplex one-tube step (RT and PCR combined in a single reaction) (S. Fuentes, J. Kreuze, and N. Boonham, *unpublished*). However, inhibition of SPCSV detection may be observed in a multiplex assay with COX, unless the concentrations of MMLV and primers and probe for COX are optimized. The USDA-APHIS National Plant Germplasm and Biotechnology Laboratory at Beltsville has developed RT-qPCR assays for SPCSV strains, SPMSV, SPCFV in a single- and two-step (reverse transcription and PCR separately) reaction. These assays are multiplexed with primers and probe for *Nad5* as an internal control and are used for detecting the target viruses in introduced germplasm accessions.

qPCR can save time, money, and labor, and offers extremely high detection sensitivity. However, its use in routine virus detection may be limited by the sequence specificity of the primers and TaqMan probes and the expensive reagents and instruments.

Generic, novel molecular detection methods: rolling-circle amplification (RCA) and deep-sequencing of small RNAs (siRNA). Rapidly evolving viruses cause problems for design of PCR primers to detect all viral strains (175). Thus, RCA, sometimes combined with RFLP, is emerging as a useful tool for sweepovirus diagnosis (132,175). RCA/RFLP combined with sequencing has allowed the identification of novel species, strains, and variants of sweepoviruses (3,64,101,132) and also has unrealized potential for detection of badnaviruses (70).

In 2009, Kreuze et al. (84) described the use of siRNA deep-sequencing and sequence analysis as a novel means to detect viruses. SPFMV and SPCSV co-infecting sweetpotato 'Huachano' were used as an experimental model. Virus-derived siRNA results from degradation of the viral genomic RNA (RNA viruses) or gene transcripts (DNA viruses) by the antiviral defense mechanism of the host, which recognizes double-stranded RNA (dsRNA) (42,83, 84,85). Assembly of the overlapping 21- or 22-nucleotide-long siRNA sequences to contigs was used to assemble the complete genome of a new strain of SPFMV (ca. 10,500 nucleotides) (84). Surprisingly, two previously unknown putative badnaviruses and a mastrevirus were also detected. They were revealed by comparing (blasting) contigs made from siRNA sequences against the se-

quences available in common gene bank databases. The greatest similarity was observed with some of the previously described badna- and mastreviruses (84). Accuracy of the assembled viral sequences was confirmed by PCR amplification and Sanger sequencing of the products (84). The primers were designed using the sequences obtained by the assembly of siRNA sequences. Hence, development of new PCR-based detection tools for viruses is a useful outcome of the siRNA deep-sequencing approach and was utilized to design new primers for detection of SPSMV-1 and SPPV in landrace sweetpotatoes in Tanzania (104).

The technique has since been applied to various samples to determine the complete genome sequences of SPVG, SPV2, SPLV, and SPFMV-RC and also to detect new sweepoviruses, badnaviruses, a cavemovirus, and a solendovirus, and new strains of SPFMV in samples from Africa (CIP, *unpublished*) and of SPCSV in samples from South America (40). A number of unknown viruses maintained in CIP's collection were also characterized using this method, including C-3 and C-6, which were identified as a novel bunyavirus and carlavirus, respectively (CIP, *unpublished*). The siRNA deep-sequencing approach is becoming more affordable as the output of new DNA sequencing technologies is continuously increasing and more samples can be bulked together. Current bottlenecks are the time-consuming sample preparation and the tools available for bio-informatic analysis of the data, but techniques are improving and acceleration should be expected as the method finds its way into human disease diagnostics (67). The method is currently being applied to several virus surveys in Africa, which will likely provide new interesting data.

Sweetpotato as a Model System for Virus–Virus and Virus–Host Interactions

The unusual synergistic sweetpotato virus disease (SPVD) caused by co-infection of SPCSV and SPFMV has resulted in an interesting line of research regarding host–virus interactions, with sweetpotato as a model. After the etiology of SPVD was described (57), studies concentrated on characterizing the molecular basis of the disease. SPFMV titers were found to be several hundred-fold greater in SPVD compared to SPFMV-infected plants, whereas titers of SPCSV were hardly affected or slightly reduced in the plants affected by SPVD (43,77,113). This led to the hypothesis that SPCSV was somehow interfering with a virus resistance mechanism acting against SPFMV and perhaps other viruses in sweetpotato. Indeed, other than some sweepoviruses (104), SPCSV was observed to synergize every other possible virus that has been tested including various strains of SPFMV, SPVC, SPMMV, SPVG, SPV2, SPCV, SPVCV, SPCFV, C-6, and CMV (41,43,80, 113,156), representing RNA and DNA viruses from very different evolutionary lineages. Learning why some sweepoviruses are not affected may present an opportunity to understand the mechanisms involved.

Because of the near universal ability of SPCSV to increase the susceptibility of sweetpotato plants to co-infecting viruses, it was suspected that a general viral defense mechanism such as RNA silencing was affected. Two RNA silencing suppressor proteins, RNase3 and p22, encoded by SPCSV were discovered (85), but it was later found that most SPCSV isolates lack the *p22* gene (43). Nevertheless, all isolates were able to cause synergistic diseases (43). The use of RNase3-expressing transgenic sweetpotato plants finally confirmed that expression of this protein alone was sufficient to eliminate antiviral resistance and generate the SPVD symptoms following infection of the plants with SPFMV alone (42). Because more severe disease and higher titers were also achieved with several other viruses, and p22 had no such effect despite being a much stronger silencing suppressor in experimental systems, it was concluded that RNase3 specifically affected a key step in antiviral defense in sweetpotato plants.

RNase3 is a dsRNA specific endonuclease that can also digest ds-siRNAs in vitro (42). It is therefore tempting to speculate that RNase3 digests a key small interfering (si)RNA molecule involved in viral defense, such as the mobile signal. What is clear from siRNA northern blots (83) and deep sequencing results (84) is that general digestion of siRNAs alone is unlikely to be the mechanism, as levels of virus-specific siRNAs are dramatically increased in SPVD-affected plants. On the other hand, a dramatic increase in overall concentration of siRNAs and the proportion of 21 to 22 nt of siRNAs can be observed in SPVD-affected plants as compared to singly infected plants (84), indicating a severely affected silencing machinery. This may not be a direct effect of RNase3, but that of increased titers of SPFMV.

The profound disturbance by SPCSV (RNase3) of the host RNA silencing machinery, which is also involved in control of host gene expression, may explain the large number of changes in gene expression detected in SPVD-affected plants (81,106). An SPVD-semi-resistant cultivar accumulating only low virus titers of both viruses displayed no disease symptoms, and gene expression was only little affected in comparison with a susceptible cultivar (106). The only exception was perhaps the strong induction of a number of genes, mostly involved in protein synthesis, early during infection before any virus could be detected in the plants. It is still unclear if any of these genes have any role in the mechanism of resistance, but understanding the mechanisms of natural resistance in sweetpotatoes may serve as a model to understand resistance in other crops. An obvious mechanism for this type of general virus resistance is efficient RNA silencing. Although the challenges with sweetpotato are daunting, due to the lack of sequence information and the complex hexaploid nature of its genome, research on virus resistance in sweetpotato may well pay off the efforts in terms of discovering the mechanisms and genes required for efficient RNA silencing and antiviral defense in this important crop plant.

Managing Sweetpotato Virus Diseases

SPFMV and other potyviruses. SPFMV is probably the most common virus infecting sweetpotato worldwide, but whether by itself it causes a serious disease is debatable. Although it can be symptomless, at least in some varieties (57,58), plants with SPFMV alone comprised nearly half of all plants with virus-like symptoms, and nearly 70% of the symptomless plants were SPFMV-infected in a virus survey in Kenya (14) (Table 2). In Ugandan surveys (11,112), plants with SPFMV alone comprised slightly more than 10% and about 6% of the diseased plants, and 4% symptomless infected plants were found. Symptoms in SPFMV-inoculated plants may gradually recede as plants get older (112). Despite the negligible or mild symptoms, plants of cv. Tanzania infected with SPFMV had half the yield of uninfected controls in one experiment in Africa (58), while other varieties varied enormously in their response in another study (125). In the United States, clones of potyvirus-infected cv. Beauregard had smaller yields in most plantings than virus-free controls (26). However, no effects on yield of single inoculation by SPFMV, SPV2, or SPVG were observed (31).

Potyviruses are managed through reducing virus inoculum by using limited generation 'seed' that was initially virus free, continually flushing out the diseased material. Virus-tested 'seed' systems have been used in some temperate countries (see below), but have not been practical in subsistence agriculture, and even introductions of new planting material are generally of non-pathogen-tested material (72,73). Yet the crop survives, with a high proportion of planting material in at least East Africa being virus-free (Table 2) (58). It does so by infected plants reverting to healthy (8), probably through the process of RNA silencing; only certain varieties of sweetpotato appear to possess this capacity, perhaps those possessing the SPFMV resistance gene identified by Mwanga et al. (117,122).

Sweet potato leaf curl and related sweepoviruses. The sweepoviruses are only recently appreciated as being potentially quite important and geographically widespread (102,132,159). They mostly also seem symptomless in sweetpotato but can reduce yield substantially (31). There are few observations on their incidence in crops; in Uganda, they seem to infect quite high percentages, at least in some cultivars (169) (Table 3).

That they are mostly symptomless, yet cause considerable individual plant yield loss (20 to 80% depending on cultivar) (31,94) and may be at high incidence in crops, suggests they could be responsible for considerable crop loss. There is, however, little information available on how to manage these virus diseases. Use of resistance seems the most practical control measure. Preliminary screening of germplasm for resistance to the U.S. strain of SPLCV has revealed a wide range of resistance/susceptibility as measured by SPLCV titers (32,94; C. Clark, *unpublished*). Germplasm accessions varied greatly in the effect of SPLCV on yield and in titers of SPLCV in infected plants. However, most genotypes did not develop discernible symptoms and there was no apparent relationship between virus titer and symptom severity or crop yield among lines evaluated. In experimental plantings with a large proportion of SPLCV-infected plants and large whitefly populations, rapid reinfection of virus-tested plants occurred by the second year of field planting (94), suggesting that management of the whitefly population should be a critical element in the management of SPLCV.

SPVD and other SPCSV-related diseases. SPVD (57) is the most important disease of sweetpotato in the tropics (25). It causes very large reductions in both foliage and root yield of individual plants, generally reducing yield by 50 to >90% (57,63,109, 115,123,125). It has been reported in East Africa since at least the early 1940s, apparently so devastating in the eastern Belgian Congo (now D. R. Congo) as to cause sweetpotato to be abandoned as a food source by the main mining company (65). Resistance has developed in the crop in areas where the disease is prevalent (7), although still 10 to 20% of plants in fields in most districts of Uganda had virus symptoms, of which about a third were infected with both SPFMV and SPCSV (112), and 70% were affected by SPVD in some western districts (56) (Table 2). In Kenya, fields in most districts also had 10 to 20% plants with virus symptoms, and just over 20% of these were infected with SPFMV and SPCSV (14). It is questionable, however, whether incidences of <10% cause much crop loss because the sprawling habit of most cultivars allows for the poor yield of diseased plants to be compensated for by increased yields of neighboring healthy plants. Instead, any yield loss may be indirect, caused by the poorer yields of the resistant landraces that are generally grown to prevent much greater infections by SPVD (56). With the advent of higher yielding SPVD-resistant varieties like NASPOT 1 (121), even this crop loss is reduced, although perhaps not completely as susceptible commercial varieties may still yield more.

SPFMV and SPCSV are both systemic in plants, and so SPVD is automatically transferred in planting material if the parent plant is infected. Planting cuttings taken from symptomless parent plants, which mostly excludes planting SPVD-affected material, is considered by farmers to be one of their most effective means of controlling SPVD (56), although not preventing the planting of cuttings from plants with late, latent infections. The practice of using apical cuttings from healthy-looking vines as planting material appears to be a means for obtaining virus-free cuttings because the landrace varieties in East Africa express remarkable resistance to most viruses, except SPCSV (58).

The spread of SPVD is determined mainly by the spread of the synergizing SPCSV, SPFMV usually spreading rapidly to SPCSV-infected plants. Whiteflies, which are the vectors of SPCSV, are therefore the driver of the spread of SPVD, and their main flight pattern in sweetpotato consists of short flights between neighboring plants, rarely flying >0.5 m above the canopy (24). Consequently, spread from an infector plot is concentrated within the first few meters and spread from outside a crop is rare (55) (Fig. 6). Rogueing affected plants to remove within-crop sources of infection is therefore an effective means of controlling the disease (Fig. 7). It is also practical because the healthy neighbors of rogued plants are able to grow more and so compensate for their absence, especially when rogueing is only done during the first month or so after planting.

Table 2. Incidence of viruses in surveys of sweetpotato in East Africa

Virus survey	Status of plant	SPFMV	SPCSV	SPFMV + SPCSV	Other viruses and combinations
Ateka et al., 2004: Kenya	448 diseased	190	61	98	61
	638 symptomless	109	15	1	36
Mukasa et al., 2003: Uganda	634 diseased	71	110	224	159
	626 symptomless	0	0	0	0
Aritua et al., 2006: Uganda	1,473 diseased	75	48	1,043	186
	200 symptomless	4	1	4	0

Table 3. Prevalence of sweepovirus(es) in different locations and cultivars in Uganda (169)

	Source of samples		Infected samples	
Cultivar	Source	District	Positives/total	%
New Kawogo	University field collection	Unknown origin	1/2	50
New Kawogo	Farmer's field	Luwero	8/25	32
New Kawogo	NaCRRI[a]	Wakiso	11/76	14
New Kawogo	Farmer's field	Luwero	3/50	8
Subtotal			23/153	15
318 L	Farmer's field	Luwero	6/15	40
1081 L	NaCRRI	Wakiso	0/15	0
NASPOT 1	NaCRRI	Wakiso	0/14	0
Dimbuka	NaCRRI	Wakiso	0/15	0
Tanzania	NaCRRI	Wakiso	0/15	0
Tanzania	Farmer's field	Soroti	0/10	0
Ejumula	NaCRRI	Wakiso	0/13	0
Ejumula	Farmer's field	Soroti	5/10	50

[a] NaCRRI = National Crops Resources Research Institute.

Resistance to SPVD. Because it has such a large effect on yield and quality, and because there are few alternatives for effective control, resistance is the main way SPVD could be controlled and it is also the greatest priority in developing sweetpotato disease resistance. Although resistance is incomplete, landraces that are somewhat resistant to SPVD tend to be grown in areas where SPCSV is particularly prevalent (7). Despite its relatively low yields, cv. New Kawogo is renowned as one of the most SPCSV-resistant sweetpotatoes known (8,78,120). Growing large amounts of this resistant landrace in an area also reduces the incidence of SPVD in nearby susceptible cultivars (12). However, the task of purposeful breeding for resistance is daunting, as there are two distinctive viruses involved, each with different insect vectors. Mwanga et al. (116,117,122) determined that two separate reces-

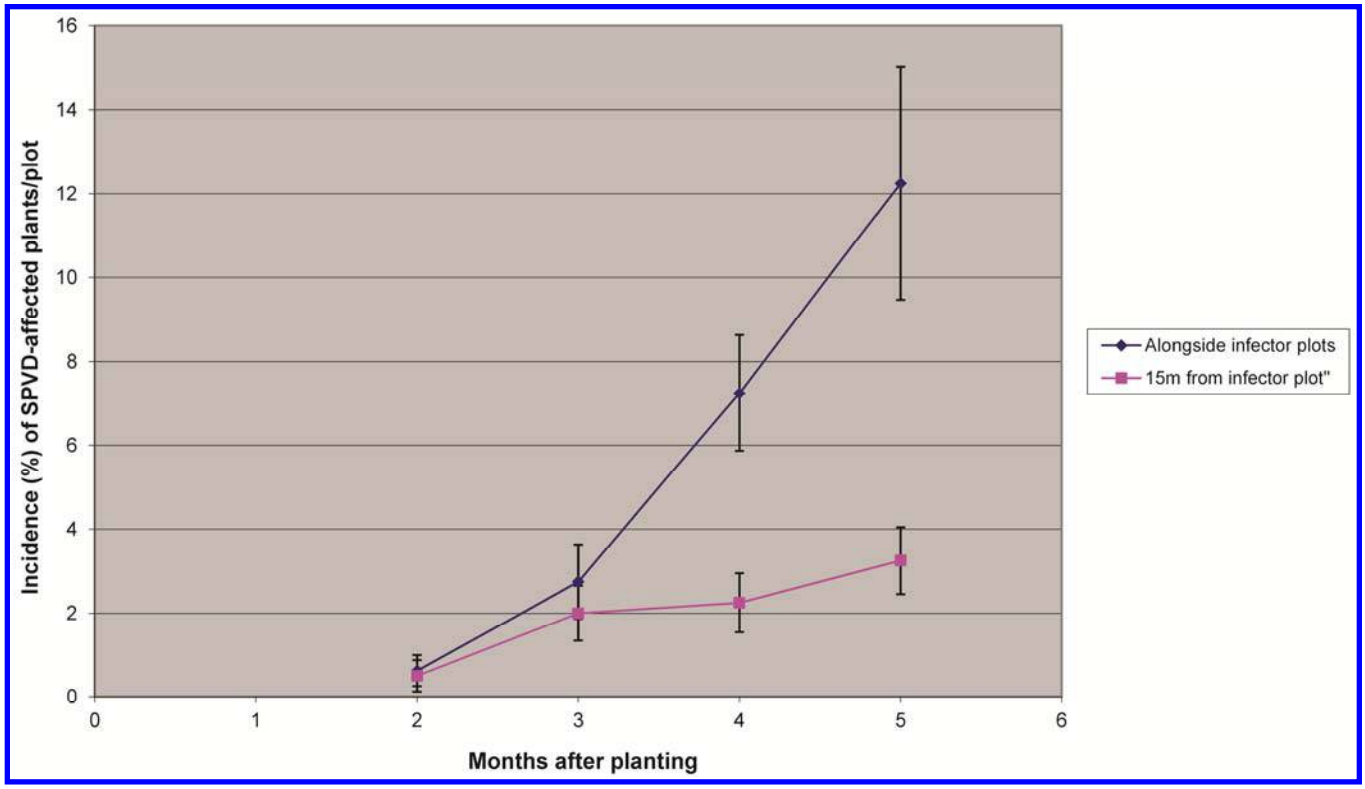

Fig. 6. Spread of sweetpotato virus disease (SPVD) to plots adjacent to or 15 m away from an SPVD-affected plot (±SE). (Reproduced from 55)

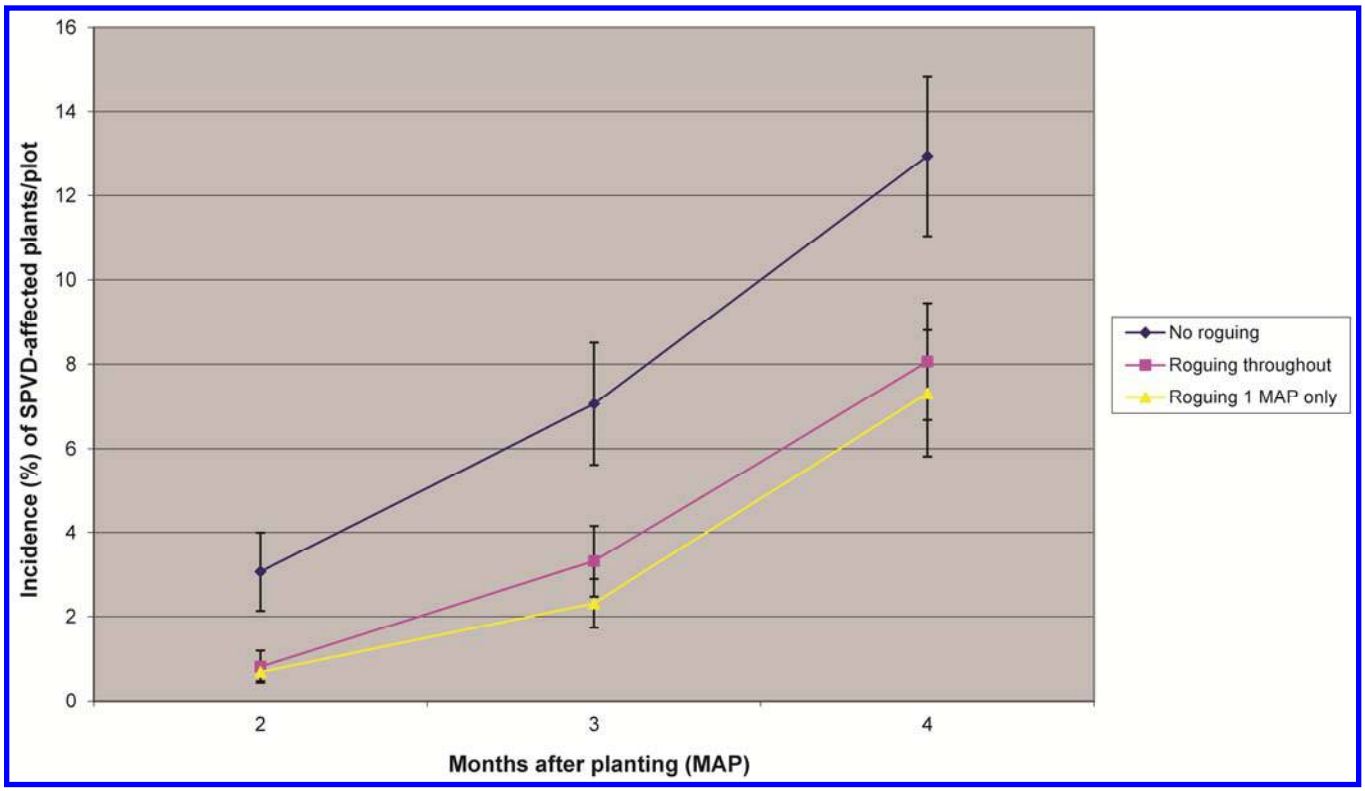

Fig. 7. Effect of roguing on spread of sweetpotato virus disease (SPVD). (Reproduced from 55)

sive genes conditioned resistance to SPFMV and SPCSV and that heritability was moderate to high. This led to the eventual release in Uganda of a series of varieties that incorporate high yield and SPVD resistance, of which NASPOT 1 and NASPOT 11 are particularly popular (118,119,121). The main feature of these resistant cultivars is that they become infected less frequently, although symptom severity, virus titers, and yield losses may also be less. The resistant cultivars also tend to recover from infection with SPFMV alone (see later), and farmers have found that cuttings taken from these cultivars are often apparently virus free (58,116).

Selection for resistance to SPVD has so far been based only on resistance to the viruses themselves (8). However, sweetpotato germplasm may yet include genotypes that affect vector behavior and possibly provide a means to enhance existing resistance. Wild species of *Ipomoea* have also been found with high levels of resistance to SPFMV, SPCSV, and SPVD (76). Molecular markers for SPVD resistance (74,107,108) and some of the genes differentially expressed in resistant cultivars in response to infection (106) have been identified and should enable more efficient and effective selection of resistance in the future.

Engineered resistance. Due to the difficulty of breeding for virus resistance in sweetpotato, genetic engineering has been attempted since the mid-nineties. Initially, the target was only SPFMV, as the relevance of SPCSV in the pathology of sweetpotato virus diseases was not well understood. Thus, resistance was obtained to SPFMV using expression of the coat protein (CP) by Monsanto scientists, and this led to the first field trials for virus resistance in Kenya. However, the resistance to SPFMV observed in greenhouses broke down in the field in Kenya (New Scientist, 7 February 2004, p. 7), possibly because the transgene was not from a locally prevalent SPFMV strain and/or because the plants became infected with SPCSV, resulting in breakdown of resistance. CP-mediated resistance to SPFMV was also achieved by Okada et al. (127,129), although durability of the resistance in the field was not reported.

Other strategies to generate virus resistance in sweetpotato to both viruses have also been tested. Rice cystein proteinase inhibitor (*OCI*) mediated resistance to potyviruses is thought to act through inhibiting the viral cystein proteinase NIa that processes the potyviral polyprotein (62). Closteroviruses also encode cystein proteinases to modify some of their proteins, and it was considered that expression of *OCI* in transgenic plants might confer resistance to both SPFMV and SPCSV. Cipriani et al. (29) reported increased resistance to SPFMV in plants of cv. Jonathan transformed with the *OCI,* but the resistance was not efficient and typical symptoms of SPVD developed in plants infected with SPCSV and SPFMV.

An RNA silencing approach was also used to target both viruses simultaneously. A construct was designed to produce transcripts that generate a double-stranded RNA structure that was homologous to the polymerase genes of each virus, thereby efficiently inducing RNA silencing defense system against both (83). Many transgenic lines accumulated only low titers of SPCSV and no symptoms developed. These results showed that sweetpotato could be protected against the disease caused by SPCSV using RNA silencing. Nevertheless, the low concentrations of SPCSV in the transgenic plants were still sufficient to break down the natural high levels of resistance to SPFMV, and SPVD developed. The authors hypothesized that even minimum amounts of RNase3 produced by SPCSV were sufficient to break down resistance to SPFMV, and immunity to SPCSV may be required for prevention of the severe virus diseases.

'Seed' Programs for Producing Healthy Planting Materials

The ability to produce plants free of detectable viruses through meristem-tip culture and to maintain and increase these plants in tissue culture has for several decades offered the potential of improving production. Recently, cryotherapy of shoot tips has been introduced as a novel and very efficient alternative for virus therapy (168). However, logistical problems of delivering the benefits of these systems to the farm level have delayed the implementation of this technology in most places. In the United States, sweetpotato foundation 'seed' programs were developed long before the advent of technology to produce virus-tested propagating material, and were designed primarily to reduce the incidence of mutations (primarily those causing loss of the desired orange storage root flesh color) and to maintain high yield through hill selection. One of the earliest seed programs to include virus testing began in California in the 1960s as a means of managing russet crack disease (44). In the past 10 to 20 years, virus testing has become an integral part of foundation seed programs in other states and has been implemented by governmental agencies in many temperate zone countries but has not generally been adopted in tropical production areas. Although they are directed at reducing effects of viruses, these programs may have additional benefits; for example, nodal propagation has the added advantage that by avoiding generation of *de novo* meristems, which are produced on storage roots used as 'seed', the incidence of somatic mutations is minimized (163). Incidence of other pathogens, such as the soft rot bacterium, *Dickeya dadantii*, may also be reduced.

There has not been a systematic survey of the methods and outcomes of these programs, but an extensive economic assessment of the program in Shandong, China (52) indicated adoption of the clean 'seed' by about 80% of the province's small growers, and an overall yield increase of about 30%. It has been more difficult to assess the impact of virus-tested 'seed' programs on management of russet crack in the United States because there has been a shift to cultivars tolerant of russet crack as these programs have been implemented. Most assessments of virus-tested sweetpotato have focused on yield and quality, but one report from Japan (151) found that virus-tested plants of a Streptomyces soil rot–resistant cultivar were more susceptible to this disease than traditionally produced infected plants. Whether there are other unintended effects of eliminating viruses from sweetpotato plants has not been explored.

Economics have dictated that foundation 'seed' (planting material) programs provide growers with a small stock of clean planting material that the growers must increase on their farms in order to plant succeeding crops (23). Although studies in the United States indicate a yield and quality benefit from use of early generation propagating material (22,23,26,33), re-infection with viruses can be very rapid. In two separate studies in the United States (22,33), 100% of virus-tested plants were re-infected with SPFMV within the first year in the field, even though decline in yield occurred gradually over several years, but yield declines did parallel slower re-infection with SPVG and SPV2 (33). This is consistent with observations that single potyvirus infections had minimal effect on yield of U.S. cultivars but that mixed infections could reduce yield 15 to 20% (31). In Brazil and China, 2 to 3 years were required for the virus-tested stock to become re-infected with SPFMV when a source of virus was in close proximity, but in Brazil and Israel, re-infection rates were slower when the virus-tested planting was isolated from sources of SPFMV (109,135,174). Thus, while re-infection with SPFMV can occur in the first year, yield decline may take 2 to 3 or more years and may be associated with accumulation of complexes of viruses that are not adequately defined.

Experiences with virus-tested 'seed' programs suggest that while they have significant benefit as currently operated, there is potential to improve the outcome if re-infection of virus-tested 'seed' can be economically reduced in growers' 'seed' programs (33). Preliminary studies in Louisiana (171) indicate that although aphid vectors are present throughout the growing season, transmission of potyviruses occurs primarily during a one- to two-month period shortly after slips are transplanted to the field. An area of future investigation will be to develop approaches growers can use, such as mineral oil sprays, barrier crops, and others, to reduce virus spread during this critical time. As meristem-tip culture has been applied to an increasing list of genotypes, some have been found that do not show a yield increase in response to therapy, suggesting the possibility that they may have resistance or tolerance to prevailing viruses (130) that might be exploited to enhance the effects of virus-tested 'seed'.

Sweetpotato 'Seed' Systems: Potential for Africa

Systems for multiplication and distribution of planting material for vegetatively propagated crops in sub-Saharan Africa are often inefficient due either to lack of national 'seed' production schemes or to low participation by the private sector. Informal 'seed' systems predominate, with the main source of planting material being farmer-saved 'seed' or 'seed' obtained by purchase or barter from local sources. The traditional approach of obtaining planting materials from old fields has a risk of rampantly spreading different diseases, particularly SPVD, despite farmers' efforts to select healthy-looking vines. The lack of well-organized 'seed' distribution systems for vegetatively propagated crops in sub-Saharan Africa (139) has contributed to the slow rate of dissemination of new cultivars, inadequate quantities of vines, and contributed to the prevalence of diseases in subsistence cropping. The development of sustainable 'seed' systems in Africa has been impeded by the inability to produce planting material during the dry season and the lack of specific regulatory control in most countries (e.g., certification), with the exception of South Africa.

A sustainable 'seed' system for vegetatively propagated crops should serve four critical roles: (i) provide a timely supply of adequate quantities of planting material for farmers, (ii) provide a channel for the dissemination of new cultivars, (iii) provide a means for controlling quality of planting material, and (iv) be affordable. The critical need to have planting material at the start of the rains to avoid late or missed planting has been considered more important than clean planting material (4). This indicates the importance of finding solutions to the problem of vine conservation through dry periods (4). In a system such as the CIP technologies deployed in Shandong, China (52), when the virus-tested plants are established in vitro, they can be rapidly multiplied using less space and potentially provide planting materials to the farmers at the beginning of the rains. While sub-Saharan Africa lacks the same infrastructure, there is a potential benefit for adopting clean 'seed' technology as indicated by Smith (146), who reported that tissue cultured sweetpotatoes planted by smallholder farmers in Zimbabwe yielded up to 25 tons compared to a national average of 6 tons per hectare.

Conventional propagation of sweetpotato in the tropics using vine cuttings has a slow multiplication rate (15:1) compared to cereal crops (200 to 300:1). However, there are different strategies for sweetpotato multiplication (18), each with merits and drawbacks, but with the potential for attaining rates of 90:1. Their management intensity requires understanding of how to optimally structure primary multiplication and what mix of secondary private and community based approaches are appropriate. For instance, primary multiplication sites were set up and well maintained by commercial farmers in Kenya and Uganda (4), and these were used to multiply and deliver large quantities of planting material. Preliminary evidence suggests that storage roots may be a viable alternative to vines for conservation and multiplication of planting material in Uganda (59), especially in areas prone to drought. There are other methods for vine multiplication and storage root preservation through the dry season (59), but much more effort is needed to find optimal systems for sub-Saharan Africa.

'Seed' systems are varied, reflecting the diversity of climate, crop types, farming systems, culture, and economics. For instance, in Uganda 'seed' systems are greatly influenced by the unimodal and bimodal rain patterns in the eastern and central regions of the country (19). The effectiveness of these systems in controlling SPVD may be augmented by farmers' practices of selecting healthy appearing planting material (55). However, without adequate 'seed' systems, this approach can also lead to the perpetuation of plants with high incidence of unrecognized single infections (112,140) with viruses such as SPCSV and SPFMV that then serve as sources of inoculum leading to SPVD development. The abundance of alternative weed hosts around sweetpotato fields also poses a challenge for maintaining healthy stock. The prevalence of SPVD has influenced the cultivars that are grown, with increasing use of cultivars that recover from SPVD but produce lower yields, such as New Kawogo, Nderera, and Munyeera (53). Some gains can be realized by extending techniques such as rogueing, rapid nursery vine multiplication, use of small roots for 'seed' that would otherwise not be consumed, and extending training from Uganda to other regions of Africa. Intervention of private plant tissue culture laboratories in this effort could lead to increased utilization of micropropagation technologies and sustainability of quality 'seed' production.

Although there is a long tradition of farmers producing their own planting material, there is clear evidence of a willingness to pay for vines under specific instances, when linked to markets and when vines are scarce (4). It is apparent that effective farmer demand for purchased vines will depend principally on the level of virus pressure, rain patterns, availability of irrigation or wetlands with adequate soil moisture throughout the year, and the existence of a significant market demand for roots. It will also be important to distinguish between developing 'seed' systems for farmers increasingly linked to markets as well as for those who remain largely subsistence farmers (55). Future interventions should create long-term economically sustainable 'seed' systems through the involvement of the private sector, including farmers located in wetlands or prepared to irrigate, as well as tissue culture laboratories, and it should take advantage of tissue culture, virus therapy, and diagnostic technologies.

Quarantine Issues

Quarantine programs play an important role in protecting the agriculture of their respective countries. Sweetpotatoes are usually banned from casual introduction in most of the countries that cultivate this crop because of quarantine restrictions. In general, quarantine programs adopted the Technical Guidelines for the Safe Movement of Sweetpotato Germplasm published by Food Agriculture Organization (FAO) and International Board for Plant Genetic Resources (IBPGR) in 1989 (110). This document provided relevant information on disease indexing available at that time. Current quarantine sweetpotato programs around the world have complemented disease indexing with new detection technologies (e.g., PCR) suited to each quarantine program (5,91).

"Seed Movement", which refers to botanical seed, is not prohibited in the United States because there is no evidence of seed transmission for any of the sweetpotato viruses described so far (170; CIP, *unpublished*). Nevertheless, since some nepoviruses are seed transmitted, efforts were initiated by the USDA-APHIS quarantine program to monitor symptoms of SPRSV (21) and others in plants generated by seeds introduced from abroad. Transport of germplasm as in vitro plantlets is still the most advisable way to meet the guideline for "Vegetative Propagating Material". Most virus-infected accessions are intercepted when introduced as cuttings or

Fig. 8. Sweetpotato accession received by the USDA-APHIS quarantine program infected with five viruses. Left: plants prior to therapy. Right: the same accession after meristem-tip culture and indexing. (Photo by Crindi Loschinkohl)

roots. Therapy by meristem-tip culture, particularly in combination with thermotherapy or cryotherapy, continues to be an excellent alternative when no virus-tested source of accessions is available (Fig. 8). Ideally, therapy should be done in the country of origin (110), but many sweetpotato-producing countries lack the resources; consequently it is an essential part of postentry quarantine. Unfortunately, intermediate quarantine centers, as called for in the guidelines, do not seem to exist. Indexing is still mainly focused on grafting onto the universal indicator plant for sweetpotato viruses, *I. setosa,* and the potyvirus preinfected TIB-8 sweetpotato clone to detect SPCSV (142). Although SPCSV is reported in the United States (1), SPCSV continues to drive quarantine decisions due to its role in SPVD and synergizing with many viruses (41,78,80,156). Quarantine programs will have to adjust to take into account the many newly discovered viruses of sweetpotato (Table 1). Sweetpotato viruses continue to be frequently detected in quarantine programs, and additional tests may be needed for these viruses (90,132; S. Fuentes and J. Abad, *unpublished data*). Fortunately, *I. setosa* is still a good indicator plant for most of those viruses. However, recently, two badnaviruses and a mastrevirus (84) were reported in sweetpotato that are not efficiently detected by *I. setosa*; consequently, new technology, such as serology or PCR, should be included in quarantine programs for their detection. At CIP, a large number of sweetpotato accessions (ca. 500) from its germplasm collection are indexed for viruses annually. Despite the progress in identifying sweetpotato viruses, many of which proved to be widespread, several lines of evidence suggest that more viruses remain to be identified (C. Clark and S. Fuentes, *unpublished*) and quarantine programs will continue to face new challenges. In Africa, sweetpotato plants are going to be moved around the continent under the guidelines of the CIP-Sweetpotato Action for Security and Health in Africa (SASHA) project. To ensure success, capacities and facilities in quarantine centers in Kenya, Mozambique, and Ghana are being improved. These centers have instant access to the Sweetpotato Knowledge Portal developed for this purpose (http://sweetpotatoknowledge.org/) in order to keep updated.

Future Directions

Recent studies indicate that control of virus diseases in sweetpotato crops is more challenging than previously realized. There are many more viruses infecting the crop than previously known. Most if not all viruses seem to be distributed worldwide as a consequence of the movement of germplasm over the past decades and centuries. Awareness of all the new viruses and the siRNA analysis-based universal approach to detect virtually any previously unknown virus provide better possibilities to enhance virus control. It can be achieved in part via virus eradication schemes in gene banks and local depositories of nuclear stocks of locally adapted cultivars. Application of the available new techniques such as cryotherapy linked to cryopreservation should be helpful to this end (167,168). There are important production areas such as western Africa, China, and some other parts of Asia where little information is available about viruses infecting sweetpotato crops. There are also indications of other undetected viruses in the Americas and Africa, so indexing sweetpotato crops for viruses is an as yet uncompleted mission that should be continued. Knowledge of vector biology and ecology has been largely neglected in sweetpotato virus epidemiology and management. Vectors of many sweetpotato viruses such as SPMMV, SPCFV, and the recently described mastre-, begomo-, cavemo-, solendo-, and badnaviruses remain to be identified and elucidated. Information on how these viruses and vectors interact and affect movement and vector biology is key in efforts to control virus spread. Methods are needed to allow rapid, inexpensive detection of viruses in the field to enable epidemiological studies and planning of proper 'seed' multiplication systems, taking both virus infection pressure and vector management into consideration. The common occurrence of sweetpotato viruses in wild species in at least the United States and East Africa is an additional dimension that needs to be considered in order to reduce reinfection of healthy propagation materials. Finally, efforts need to be continued to elucidate the mechanism by which SPCSV eliminates the antiviral defense in sweetpotato plants and to use the knowledge to engineer SPCSV-resistant cultivars. Controlling this single virus alone is expected to improve the yields of sweetpotato crops significantly in those areas where SPCSV and SPVD occur.

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Literature Cited

1. Abad, J. A., Parks, J., New, S. L., Fuentes, S., Jester, W., and Moyer, J. W. 2007. First report of *Sweetpotato chlorotic stunt virus* (genus *Crinivirus,* family *Closteroviridae*) in North Carolina. Plant Dis. 91:327.
2. Akel, E., Ismail, I. D., Al-Chaabi, S., and Fuentes, S. 2010. New natural weed hosts of *Sweet potato feathery mottle virus* in Syria. Arab J. Plant Prot. 28:96-100.
3. Albuquerque, L. C., Inoue-Nagata, A. K., Pinheiro, B., Ribeiro, S. D., Resende, R. O., Moriones, E., and Navas-Castillo, J. 2011. A novel monopartite begomovirus infecting sweet potato in Brazil. Arch. Virol. DOI 10.1007/s00705-011-1016-x.
4. Andrade, M., Barker, I., Cole, D., Dapaah, H., Elliott, H., Fuentes, S., Grüneberg, W., Kapinga, R., Kroschel, J., Labarta, R., Lemaga, B., Loechl, C., Low, J., Lynam, J., Mwanga, R., Ortiz, O., Oswald, A., and Thiele, G. 2009. Unleashing the potential of sweetpotato in sub-Saharan Africa: Current challenges and way forward. International Potato Center (CIP), Lima, Peru. Working Paper 2009-1.
5. Anonymous. 2007. *Ipomoea* (Sweetpotato/Kumara) Post-Entry Quarantine Testing Manual. Biosecurity New Zealand, Ministry of Agriculture and Forestry, Aukland. http://www.biosecurity.govt.nz/files/regs/imports/plants/high-value-crops/ipomoea-testing-manual.pdf.
6. Aritua, V., and Adipala, E. 2004. Molecular diversity in sweetpotato infecting viruses in Africa: With emphasis on sweet potato chlorotic fleck virus. Uganda J. Agric. Sci. 9:707-713.
7. Aritua, V., Adipala, E., Carey, E. E., and Gibson, R. W. 1998. The incidence of sweet potato virus disease and virus resistance of sweet potato grown in Uganda. Ann. Appl. Biol. 132:399-411.
8. Aritua, V., Alicai, T., Adipala, E., Carey, E. E., and Gibson, R. W. 1998. Aspects of resistance to sweet potato virus disease in sweet potato. Ann. Appl. Biol. 132:387-398.
9. Aritua, V., Barg, E., Adipala, E., Gibson, R. W., Lesemann, D. E., and Vetten, H. J. 2009. Host range, purification, and genetic variability in Sweet potato chlorotic fleck virus. Plant Dis. 93:87-93.
10. Aritua, V., Barg, E., Gibson, R. W., Adipala, E., and Vetten, H. J. 2007. Sequence analysis of the entire RNA genome of sweet potato chlorotic fleck virus reveals that it belongs to a distinct carlavirus species. Arch. Virol. 152:813-818.
11. Aritua, V., Bua, B., Barg, E., Vetten, H. J., Adipala, E., and Gibson, R. W. 2006. Incidence of five viruses infecting sweetpotatoes in Uganda; the first evidence of Sweet potato caulimo-like virus in Africa. Plant Pathol. 56:324-331.
12. Aritua, V., Legg, J. P. T., Smit, N. E. J. M., and Gibson, R. W. 1999. Effect of local inoculum on the spread of sweet potato virus disease: Widespread cultivation of a resistant cultivar limits infection of susceptible cultivars. Plant Pathol. 48:655-661.
13. Ateka, E. M., Barg, E., Njeru, R. W., Lesemann, D. E., and Vetten, H. J. 2004. Further characterization of 'sweet potato virus 2': A distinct species of the genus *Potyvirus*. Arch. Virol. 149:225-239.
14. Ateka, E. M., Njeru, R. W., Kibaru, A. G., Kimenju, J. W., Barg, E., Gibson, R. W., and Vetten, H. J. 2004. Identification and distribution of viruses infecting sweet potato in Kenya. Ann. Appl. Biol. 144:371-379.
15. Atkey, P. T., and Brunt, A. A. 1987. Electron microscopy of an isometric caulimo-like virus from sweet potato (*Ipomoea batatas*). J. Phytopathol. 118:370-376.
16. Atreya, P. L., López-Moya, J. J., Chu, M., Atreya, C. D., and Pirone, T. P. 1995. Mutational analysis of the coat protein N-terminal amino acid involved in potyvirus transmission by aphids. J. Gen. Virol. 76:265-270.
17. Banks, G. K., Bedford, I. D., Beitia, F. J., Rodriguez-Cerezo, E., and Markham, P. G. 1999. A Novel Geminivirus of *Ipomoea indica* (*Convolvulacae*) from Southern Spain. Plant Dis. 83:486.

18. Barker, I. 2008. Strategies for improving formal and farmer based seed systems for enhancing potato and sweetpotato production efficiency and competitiveness in LAC, SSA and Asia. Annual Output Report. International Potato Center (CIP), Lima, Peru.
19. Bashaasha, B., Mwanga, R. O. M., Ocitti, P., Obwoya, C., and Ewell, P. T. 1995. Sweetpotato in the farming and food systems of Uganda: A farm survey report. International Potato Center (CIP) and National Agricultural Research Organization (NARO), Lima, Peru.
20. Briddon, R. W., Patil, B. L., Bagewadi, B., Nawaz-ul-Rehman, M., and Fauquet, C. M. 2010. Distinct evolutionary histories of the DNA-A and DNA-B components of bipartite begomoviruses. BMC Evol. Biol. 10:97.
21. Brown, J. D., Brunt, A. A., and Hugo, S. A. 1988. Studies on viruses isolated from sweetpotato (*Ipomoea batatas*). Pages 104-108 in: Report of the Glasshouse Crops Research Institute for 1986-8. Littlehampton, UK.
22. Bryan, A. D., Pesic-VanEsbroeck, Z., Schultheis, J. R., Pecota, K. V., Swallow, W. H., and Yencho, G. C. 2003. Cultivar decline in sweetpotato: II. Impact of virus infection on yield and storage root quality in 'Beauregard' and 'Hernandez'. J. Am. Soc. Hortic. Sci. 128:856-863.
23. Bryan, A. D., Schultheis, J. R., Pesic-VanEsbroeck, Z., and Yencho, G. C. 2003. Cultivar decline in sweetpotato: I. Impact of micropropagation on yield, storage root quality, and virus incidence in 'Beauregard'. J. Am. Soc. Hortic. Sci. 128:846-855.
24. Byamukama, E., Gibson, R. W., Aritua, V., and Adipala, E. 2004. Within-crop spread of sweet potato virus disease and the population dynamics of its whitefly and aphid vectors. Crop Prot. 23:109-116.
25. Carey, E. E., Gibson, R. W., Fuentes, S., Machmud, M., Mwanga, R. O. M., Turyamureeba, G., Zhang, L., Ma, D., Abo el Abbas, F., El-Bedewy, R., and Salazar, L. F. 1999. The causes and control of virus diseases of sweetpotato in developing countries: is sweetpotato virus disease the main problem. Pages 241-248 in: CIP Program Report 1997-8 Sweetpotato. International Potato Center (CIP). Lima, Peru.
26. Carroll, H. W., Villordon, A. Q., Clark, C. A., La Bonte, D. R., and Hoy, M. W. 2004. Studies on Beauregard sweetpotato clones naturally infected with viruses. Int. J. Pest. Manag. 50:101-106.
27. Chen, L., Du, Z., Hamaguchi, T., Sugita, T., Nagata, R., Terao, H., and Tsuzuki, E. 2008. Clonal propagation and quick detection of virus-free plants of sweet potato, *Ipomoea batatas*. Bull. Minamikyushu Univ. No. 38A:1-5.
28. Chung, B. Y.-W., Miller, W. A., Atkins, J. F., and Firth, A. E. 2008. An overlapping essential gene in the Potyviridae. Proc. Natl. Acad. Sci. USA 105:5897-5902.
29. Cipriani, G., Fuentes, S., Bello, V., Salazar, L. F., Ghislain, M., and Zhang, D. P. 2001. Transgene expression of rice cysteine proteinase inhibitors for the development of resistance against sweetpotato feathery mottle virus. Pages 267-271 in: CIP Program Report 1999-2000. International Potato Center, Lima, Peru.
30. Clark, C. A., Derrick, K. S., Pace, C. S., and Watson, B. 1986. Survey of wild *Ipomoea* species as potential reservoirs of sweet potato feathery mottle virus in Lousiana. Plant Dis. 70:931-932.
31. Clark, C. A., and Hoy, M. W. 2006. Effects of common viruses on yield and quality of Beauregard sweetpotato in Louisiana. Plant Dis. 90:83-88.
32. Clark, C. A., Hoy, M. W., and Kokkinos, C. D. 2004. Yield decline of sweetpotato cultivars and virus infection. (Abstr.) Phytopathology 94:S19.
33. Clark, C. A., Smith, T. P., Ferrin, D. M., and Villordon, A. Q. 2010. Performance of sweetpotato foundation seed after incorporation into commercial operations in Louisiana. HortTechnology 20:977-982.
34. Clark, C. A., and Valverde, R. A. 2000. Viruses and sweetpotato cultivar decline in Louisiana, USA. Pages 62-69 in: Proc. Int. Workshop Sweetpotato Cultivar Decline Study. Y. Nakazawa and K. Ishiguro, eds. Kyushu National Agricultural Experiment Station (KNAES), Miyakonjo, Japan.
35. Cohen, J., and Loebenstein, G. 1991. Role of a whitefly-transmitted agent in infection of sweetpotato by cucumber mosaic virus. Plant Dis. 75:291-292.
36. Cohen, J., Milgram, M., Antignus, Y., Pearlsman, M., Lachman, O., and Loebenstein, G. 1997. Ipomoea crinkle leaf curl caused by a whitefly-transmitted Gemini-like virus. Ann. Appl. Biol. 131:273-282.
37. Colinet, D., Kummert, J., and Lepoivre, P. 1996. Molecular evidence that the whitefly-transmitted sweetpotato mild mottle virus belongs to a distinct genus of the Potyviridae. Arch. Virol. 141:125-135.
38. Colinet, D., Kummert, J., and Lepoivre, P. 1997. Evidence for the assignment of two strains of SPLV to the genus *Potyirus* based on coat protein and 3′ non-coding region sequence data. Virus Res. 49:91-100.
39. Colinet, D., Nguyen, M., Kummert, J., and Lepoivre, P. 1998. Differentiation among potyviruses infecting sweet potato based on genus- and virus-specific reverse transcription polymerase chain reaction. Plant Dis. 82:223-229.
40. Cuellar, W. J., Cruzado, R. K., Fuentes, S., Untiveros, M., Soto, M., and Kreuze, J. F. 2011. Sequence characterization of a Peruvian isolate of *Sweet potato chlorotic stunt virus*: Further variability and a model for p22 acquisition. Virus Res. 157:111-115.
41. Cuellar, W. J., De Souza, J., Barrantes, I., Fuentes, S., and Kreuze, J. F. 2011. Distinct cavemoviruses interact synergistically with *Sweet potato chlorotic stunt virus* (genus *Crinivirus*) in cultivated sweet potato. J. Gen. Virol. 92:1233-1243.
42. Cuellar, W. J., Kreuze, J. F., Rajamaki, M.-L., Cruzado, K. R., Untiveros, M., and Valkonen, J. P. T. 2009. Elimination of antiviral defense by viral RNase III. Proc. Nat. Acad. Sci. USA 106:10354-10358.
43. Cuellar, W. J., Tairo, F., Kreuze, J. F., and Valkonen, J. P. T. 2008. Analysis of gene content in sweet potato chlorotic stunt virus RNA1 reveals the presence of the p22 RNA silencing suppressor in only a few isolates: Implications for viral evolution and synergism. J. Gen. Virol. 89:573-582.
44. Dangler, J. M., Scheuerman, R. W., Campbell, R. N., and Clark, C. A. 1994. Meristem-tip culture and California's clean foundation sweetpotato program. HortTechnology 4:227-228.
45. De Souza, J., and Cuellar, W. J. 2011. Sequence analysis of the replicase gene of 'sweet potato caulimo-like virus' suggests that this virus is a distinct member of the genus *Cavemovirus*. Arch. Virol. 153:535-537.
46. Di Feo, L., Nome, S. F., Biderbost, E., Fuentes, S., and Salazar, L. 2000. Etiology of sweet potato chlorotic dwarf disease in Argentina. Plant Dis. 84:35-39.
47. Dolja, V. V., Kreuze, J. F., and Valkonen, J. P. T. 2006. Comparative and functional genomics of closteroviruses. Virus Res. 117:38-51.
48. Fauquet, C. M., and Stanley, J. 2003. Geminivirus classification and nomenclature: Progress and problems. Ann. Appl. Biol. 142:165-189.
49. Fenby, N., Seal, S., Gibson, R., and Foster, G. 1998. RNA extraction from virus-diseased sweet potato for reverse transcriptase polymerase chain reaction analysis. Mol. Biotechnol. 10:187-190.
50. Fuentes, S., Mayo, M. A., Jolly, C. A., Nakano, M., Querci, M., and Salazar, L. F. 1996. A novel luteovirus from sweet potato, sweet potato leaf speckling virus. Ann. Appl. Biol. 128:491-504.
51. Fuentes, S., and Salazar, L. F. 1989. Identification of sweetpotato [*Ipomoea batatas* (L.) Lam] viruses. Fitopatología 24:43.
52. Fuglie, K. O., Zhang, L., Salazar, L. F., and Walker, T. S. 1999. Economic Impact of Virus-free Sweetpotato Planting Material in Shandong Province, China. International Potato Center, Lima, Peru.
53. Gasura, E., and Mukasa, S. B. 2010. Prevalence and implications of sweetpotato recovery from sweet potato virus disease in Uganda. Afr. Crop Sci. J. 18:195-205.
54. Gibbs, A., and Mackenzie, A. 1997. A primer pair for amplifying part of the genome of all potyvirids by RT-PCR. J. Virol. Methods 63:9-16.
55. Gibson, R. W., Aritua, V., Byamukama, E., Mpembe, I., and Kayongo, J. 2004. Control strategies for sweet potato virus disease in Africa. Virus Res. 100:115-122.
56. Gibson, R. W., Jeremiah, S. C., Aritua, V., Msabaha, R. P., Mpembe, I., and Ndunguru, J. 2000. Sweet potato virus disease in sub-Saharan Africa: Evidence that neglect of seedlings in the traditional farming system hinders the development of superior resistant landraces. J. Phytopathol.-Phytopathol. Z. 148:441-447.
57. Gibson, R. W., Mpembe, I., Alicai, T., Carey, E. E., Mwanga, R. O. M., Seal, S. E., and Vetten, H. J. 1998. Symptoms, aetiology and serological analysis of sweet potato virus disease in Uganda. Plant Pathol. 47:95-102.
58. Gibson, R. W., Mwanga, R. O. M., Kasule, S., Mpembe, I., and Carey, E. E. 1997. Apparent absence of viruses in most symptomless field-grown sweet potato in Uganda. Ann. Appl. Biol. 130:481-490.
59. Gibson, R. W., Mwanga, R. O. M., Namanda, S., Jeremiah, S. C., and Barker, I. 2009. Review of sweetpotato seed systems in East and Southern Africa. Integrated Crop Management Working Paper 2009-1. International Potato Center (CIP), Lima, Peru.
60. Giner, A., Lakatos, L., García-Chapa, M., López-Moya, J. J., and Burgyán, J. 2010. Viral protein inhibits RISC activity by argonaute binding through conserved WG/GW motifs. PLoS Pathogens 6(7): e1000996. doi:10.1371/journal.ppat.1000996.
61. Gutiérrez, D. L., Fuentes, S., and Salazar, L. F. 2003. Sweetpotato virus disease (SPVD): Distribution, incidence, and effect on sweetpotato yield in Peru. Plant Dis. 87:297-302.
62. Gutierrez-Campos, R., Torres-Acosta, J. A., Saucedo-Arias, L. J., and Gomez-Lim, M. A. 1999. The use of cysteine proteinase inhibitors to engineer resistance against potyviruses in transgenic tobacco plants. Nature Biotechnol. 17:1223-1226.
63. Hahn, S. K. 1979. Effects of viruses (SPVD) on growth and yield of sweet potato. Exp. Agric. 15:252-256.
64. Haible, D., Kober, S., and Jeske, H. 2006. Rolling circle amplification revolutionizes diagnosis and genomics of geminiviruses. J. Virol. Methods 135:9-16.
65. Hansford, C. G. 1944. A probably virus disease of sweet potato. East Afr. Agric. J. 19:126-127.
66. Hoyer, U., Maiss, E., Jelkmann, W., Lesemann, D. E., and Vetten, H. J. 1996. Identification of the coat protein gene of a sweet potato sunken vein closterovirus isolate from Kenya and evidence for a serological relationship among geographically diverse closterovirus isolates from sweetpotato. Phytopathology 86:744-750.
67. Isakov, O., Modai, S., and Shomron, N. 2011. Pathogen detection using short-RNA deep sequencing subtraction and assembly. Bioinformatics: http://bioinformatics.oxfordjournals.org/cgi/doi/10.1093/bioinformatics/btr349.
68. Ishak, J. A. 2002. Pathological studies on some virus diseases of sweet potato. M.Sc. thesis. Ain Shams University, Cairo, Egypt.
69. IsHak, J. A., Kreuze, J. F., Johansson, A., Mukasa, S. B., Tairo, F., Abo El-

Abbas, F. M., and Valkonen, J. P. T. 2003. Some molecular characteristics of three viruses from SPVD-affected sweet potato plants in Egypt. Arch. Virol. 148:2449-2460.
70. James, A. P., Geijskes, R. J., Dale, J. L., and Harding, R. M. 2011. Development of a novel rolling-circle amplification technique to detect *Banana streak virus* that also discriminates between integrated and episomal virus sequences. Plant Dis. 95:57-62.
71. Jones, R. A. C., and Dwyer, G. I. 2007. Detection of Sweet potato chlorotic fleck virus and Sweet potato feathery mottle virus - strain O in Australia. Australas. Plant Pathol. 36:591-594.
72. Kapinga, R., Chirimi, B., Kiflewahid, B., Amour, R., and Carey, T. 2000. Rapid dissemination of improved sweetpotato varieties through informal seed multiplication and distribution channels: Experiences from the Lake Zone of Tanzania. Pages 91-98 in: Proc. 5th Trienn. Conf. Afr. Potato Assoc. E. Adipala, P. Nampala, and M. Osiru, eds. Kampala, Uganda.
73. Kapinga, R., Tumwegamire, S., Lemaga, B., Andrade, M., Mwanga, R., Mtunda, K., Ndolo, P., Nsumba, J., Agili, S., and Serwadda, B. 2005. Development of farmer based seed systems for healthy planting material and increased sweetpotato production in East and Southern Africa. Afr. Crop Sci. Conf. Proc. J. S. Tenywa, E. Adipala, P. Nampala, G. Tusiime, P. Okori, and W. Kyamuhangire, eds. 7:1169-1173.
74. Karuri, H. W., Ateka, E. M., Amata, R., Nyende, A. B., and Muigai, A. W. T. 2009. Characterization of Kenyan sweet potato genotypes for SPVD resistance and high dry matter content using simple sequence repeat markers. Afr. J. Biotechnol. 8:2169-2175.
75. Karyeija, R. F., Gibson, R. W., and Valkonen, J. P. T. 1998. The significance of sweet potato feathery mottle virus in subsistence sweet potato production in Africa. Plant Dis. 82:4-15.
76. Karyeija, R. F., Gibson, R. W., and Valkonen, J. P. T. 1998. Resistance to sweetpotato virus disease (SPVD) in wild East African *Ipomoea*. Ann. Appl. Biol. 133:39-44.
77. Karyeija, R. F., Kreuze, J. F., Gibson, R. W., and Valkonen, J. P. T. 2000. Synergistic interactions of a potyvirus and a phloem-limited crinivirus in sweet potato plants. Virology 269:26-36.
78. Karyeija, R. F., Kreuze, J. F., Gibson, R. W., and Valkonen, J. P. T. 2000. Two serotypes of *Sweet potato feathery mottle virus* in Uganda and their interactions with resistant sweet potato cultivars. Phytopathology 90:1250-1255.
79. Kokkinos, C. D., and Clark, C. A. 2006. Real-time PCR assays for detection and quantification of sweetpotato viruses. Plant Dis. 90:783-788.
80. Kokkinos, C. D., and Clark, C. A. 2006. Interactions among S*weet potato chlorotic stunt virus* and different potyviruses and potyvirus strains infecting sweetpotato in the United States. Plant Dis. 90:1347-1352.
81. Kokkinos, C. D., Clark, C. A., McGregor, C. E., and LaBonte, D. R. 2006. The effect of sweet potato virus disease and its viral components on gene expression levels in sweetpotato. J. Am. Soc. Hortic. Sci. 131:657-666.
82. Kreuze, J. F., Karyeija, R. F., Gibson, R. W., and Valkonen, J. P. T. 2000. Comparisons of coat protein gene sequences show that East African isolates of *Sweet potato feathery mottle virus* forms a genetically distinct group. Arch. Virol. 145:567-574.
83. Kreuze, J. F., Klein, I. S., Lazaro, M. U., Chuquiyuri, W. J. C., Morgan, G. L., Mejía, P. G. C., Ghislain, M., and Valkonen, J. P. T. 2008. RNA silencing-mediated resistance to a crinivirus (Closteroviridae) in cultivated sweetpotato (*Ipomoea batatas* L.) and development of sweetpotato virus disease following co-infection with a potyvirus. Mol. Plant Pathol. 9:589-598.
84. Kreuze, J. F., Perez, A., Untiveros, M., Quispe, D., Fuentes, S., Barker, I., and Simon, R. 2009. Complete viral genome sequence and discovery of novel viruses by deep sequencing of small RNAs: A generic method for diagnosis, discovery and sequencing of viruses. Virology 388:1-7.
85. Kreuze, J. F., Savenkov, E. I., Cuellar, W., Li, X., and Valkonen, J. P. T. 2005. Viral Class 1 RNase III involved in suppression of RNA silencing. J. Virol. 79:7227-7238.
86. Kreuze, J. F., Savenkov, E. I., and Valkonen, J. P. T. 2002. Complete genome sequence and analyses of the subgenomic RNAs of *Sweet potato chlorotic stunt virus* reveal several new features for the genus Crinivirus. J. Virol. 76:9260-9270.
87. Kroth, L. L., Daniels, J., and Pierobom, C. R. 2005. Detection of sweetpotato feathery mottle virus by a simplified method of RT-PCR. R. Bras. Agrociência 11:109-111.
88. Kwak, H.-R., Kim, M.-K., Jung, M.-N., Lee, S.-H., Park, J.-W., Kim, K.-H., Ko, S.-J., and Choi, H.-S. 2007. Genetic diversity of *Sweet potato feathery mottle virus* from sweet potatoes in Korea. Plant Pathol. J. 23:13-21.
89. Lee, S. C., and Chang, Y. C. 2006. Multiplex RT-PCR detection of two orchid viruses with an internal control of plant *nad5* mRNA. Plant Pathol. Bull. 15:187-196.
90. Li, R., Mock, R., Huang, Q., Abad, J., Hartung, J., and Kinard, G. 2008. A reliable and inexpensive method of nucleic acid extraction for the PCR-based detection of diverse plant pathogens. J. Virol. Methods 154:48-55.
91. Li, R., Salih, S., and Hurtt, S. 2004. Detection of geminiviruses in sweet potato by polymerase chain reaction. Plant Dis. 88:1347-1351.
92. Liao, C. H., Chien, K., Chung, M. L., Chiu, R. J., and Han, Y. H. 1979. A study on sweetpotato virus disease in Taiwan. I. Sweetpotato yellow spot virus disease. J. Agric. Res. China 28:127-137.
93. Ling, K.-S., Harrison, H. F., Simmons, A. M., Zhang, S. C., and Jackson, D. M. 2011. Experimental host range and natural reservoir of sweet potato leaf curl virus in the United States. Crop Prot. 30:1055-1062.
94. Ling, K. S., Jackson, D. M., Harrison, H., Simmons, A. M., and Pesic-VanEsbroeck, Z. 2010. Field evaluation of yield effects on the USA heirloom sweetpotato cultivars infected by Sweet potato leaf curl virus. Crop Prot. 29:757-765.
95. Loebenstein, G., and Thottappilly, G., eds. 2009. The Sweetpotato. Springer Sciences Business Media BV, Dordrecht, The Netherlands.
96. Loebenstein, G., Thottappilly, G., Fuentes, S., and Cohen, J. 2009. Virus and phytoplasma diseases. Pages 105-134 in: The Sweetpotato. G. Loebenstein and G. Totthappilly, eds. Springer Sciences Business Media BV, Dordrecht, The Netherlands.
97. Lotrakul, P., and Valverde, R. A. 1999. Cloning of a DNA-A-like genomic component of sweet potato leaf curl virus: Nucleotide sequence and phylogenetic relationships. Mol. Plant Pathol. Online. http://www.bspp.org.uk/mppol/1999/0422lotrakul/.
98. Lotrakul, P., Valverde, R. A., and Clark, C. A. 2003. Properties of a begomovirus isolated from sweetpotato [*Ipomoea batatas* (L.) Lam.] infected with *Sweet potato leaf curl virus*. Rev. Mexicana Fitopatol. 21:128-136.
99. Lotrakul, P., Valverde, R. A., Clark, C. A., Hurtt, S., and Hoy, M. W. 2002. *Sweetpotato leaf curl virus* and related geminiviruses in sweetpotato. Acta Hortic. 583:135-141.
100. Lotrakul, P., Valverde, R. A., Clark, C. A., Sim, J., and de La Torre, R. 1998. Detection of a geminivirus infecting sweet potato in the United States. Plant Dis. 82:1253-1257.
101. Lozano, G., Trenado, H. P., Valverde, R. A., and Navas-Castillo, J. 2009. Novel begomovirus species of recombinant nature in sweet potato (*Ipomoea batatas*) and *Ipomoea indica*: Taxonomic and phylogenetic implications. J. Gen. Virol. 90:2550-2562.
102. Luan, Y. S., Zhang, J., Liu, D. M., and Li, W. L. 2007. Molecular characterization of sweet potato leaf curl virus isolate from China (SPLCV-CN) and its phylogenetic relationship with other members of the Geminiviridae. Virus Genes 35:379-385.
103. Mardis, E. R. 2011. A decade's perspective on DNA sequencing technology. Nature 470:198-203.
104. Mbanzibwa, D. R., Tairo, F., Gwandu, C., Kullaya, A., and Valkonen, J. P. T. 2011. First report of Sweetpotato symptomless virus 1 and Sweetpotato virus A in sweetpotatoes in Tanzania. Plant Dis. 95:224.
105. McGregor, C., Miano, D., LaBonte, D., Hoy, M., and Clark, C. 2009. The effect of the sequence of infection of the causal agents of sweet potato virus disease on symptom severity and individual virus titres in sweet potato cv. Beauregard. J. Phytopathol. 157:514-517.
106. McGregor, C. E., Miano, D. W., LaBonte, D. R., Hoy, M., Clark, C. A., and Rosa, G. J. M. 2009. Differential gene expression of resistant and susceptible sweetpotato plants after infection with the causal agents of sweet potato virus disease. J. Am. Soc. Hortic. Sci. 134:658-666.
107. Mcharo, M., LaBonte, D., Mwanga, R. O. M., and Kriegner, A. 2005. Associating molecular markers with virus resistance to classify sweetpotato genotypes. J. Am. Soc. Hortic. Sci. 130:355-359.
108. Miano, D. W., LaBonte, D. R., and Clark, C. A. 2008. Identification of molecular markers associated with sweet potato resistance to sweet potato virus disease in Kenya. Euphytica 160:15-24.
109. Milgram, M., Cohen, J., and Loebenstein, G. 1996. Effects of sweet potato feathery mottle virus and sweet potato sunken vein virus on sweet potato yields and rates of reinfection of virus-free planting material in Israel. Phytoparasitica 24:189-193.
110. Moyer, J. W., Jackson, G. V. H., and Frison, E. A., eds. 1989. FAO/IBPGR Technical Guidelines for the Safe Movement of Sweet Potato Germplasm. Food and Agriculture Organization of the United Nations, Rome/International Board for Plant Genetic Resources, Rome.
111. Moyer, J. W., and Salazar, L. F. 1989. Virus and viruslike diseases of sweet potato. Plant Dis. 73:451-455.
112. Mukasa, S. B., Rubaihayo, P. R., and Valkonen, J. P. T. 2003. Incidence of viruses and viruslike diseases of sweetpotato in Uganda. Plant Dis. 87:329-335.
113. Mukasa, S. B., Rubaihayo, P. R., and Valkonen J. P. T. 2006. Interactions between a crinivirus, an ipomovirus and a potyvirus in coinfected sweetpoato plants. Plant Pathol. 55:458-467.
114. Mukasa, S. B., Tairo, F., Kreuze, J. F., Kullaya, A., Rubaihayo, P. R., and Valkonen, J. P. T. 2003. Coat protein sequence analysis reveals occurrence of distinct strains of SPFMV in Uganda and Tanzania. Virus Genes 27:49-56.
115. Mukiibi, J. 1977. Effects of mosaic on the yield of sweet potatoes. Pages 169-170 in: Proc. 4th Int. Soc. Trop. Root Crops. CIAT, Cali, Columbia.
116. Mwanga, R. O. M., Carey, E. E., Moyer, J. W., Zhang, D. P., and Yencho, G. C. 2002. Nature of resistance of sweetpotato to sweet potato virus disease. T. Ames, ed. Acta Hortic. 583:113-119.
117. Mwanga, R. O. M., Kriegner, A., Cervantes-Flores, J. C., Zhang, D. P., Moyer, J. W., and Yencho, G. C. 2002. Resistance to *Sweetpotato chlorotic stunt virus* and *Sweetpotato feathery mottle virus* is mediated by two separate recessive genes in sweetpotato. J. Am. Soc. Hortic. Sci. 127:798-806.
118. Mwanga, R. O. M., Niringiye, C., Alajo, A., Kigozi, B., Namukula, J.,

Mpembe, I., Tumwegamire, S., Gibson, R. W., and Yencho, G. C. 2011. 'NASPOT 11', a sweetpotato cultivar bred by a participatory plant-breeding approach in Uganda. HortScience 46:317-321.
119. Mwanga, R. O. M., Odongo, B., Niringiye, C., Alajo, A., Kigozi, B., Makumbi, R., Lugwana, E., Namukula, J., Mpembe, I., Kapinga, R., Lemaga, B., Nsumba, J., Tumwegamire, S., and Yencho, C. G. 2009. 'NASPOT 7', 'NASPOT 8', 'NASPOT 9 O', 'NASPOT 10 O', and 'Dimbuka-Bukulula' sweetpotato. HortScience 44:828-832.
120. Mwanga, R. O. M., Odongo, B., Ocitti-p'Obwoya, C. N., Smit, N., and Gibson, R. W. 2001. Release of five sweet potato cultivars in Uganda. HortScience 36:385-386.
121. Mwanga, R. O. M., Odongo, B., Turyamureeba, G., Alajo, A., Yencho, G. C., Gibson, R. W., Smit, N. E. J. M., and Carey, E. E. 2003. Release of six sweetpotato cultivars ('NASPOT 1' to 'NASPOT 6') in Uganda. HortScience 38:475-476.
122. Mwanga, R. O. M., Yencho, G. C., and Moyer, J. W. 2002. Diallel analysis of sweetpotatoes for resistance to sweetpotato virus disease. Euphytica 128:237-248.
123. Ngeve, J. M., and Bouwkamp, J. C. 1991. Effects of sweet potato virus disease on the yield of sweet potato genotypes in Cameroon. Exp. Agric. 27:221-225.
124. Nishiguchi, M., Mori, M., Suzuki, F., Nagata, R., Morishita, T., Sakai, J., Hanada, K., and Usugi, T. 1995. Specific detection of a severe strain of sweet potato feathery mottle virus (SPFMV-S) by reverse transcription and polymerase chain reaction (RT-PCR). Ann. Phytopathol. Soc. Jpn. 61:119-122.
125. Njeru, R. W., Mburu, M. W. K., Cheramgoi, E., Gibson, R. W., Kiburi, Z. M., Obudho, E., and Yobera, D. 2004. Studies on the physiological effects of viruses on sweet potato yield in Kenya. Ann. Appl. Biol. 145:71-76.
126. Nome, S. F. 1973. Sweet potato vein mosaic in Argentina. Phytopathol. Z. 77:44-45.
127. Okada, Y., Nishiguchi, M., Saito, A., Kimura, T., Mori, M., Hanada, K., Sakai, J., Matsuda, Y., and Murata, T. 2002. Inheritance and stability of the virus-resistant gene in the progeny of transgenic sweet potato. Plant Breed. 121**:**249-253.
128. Okada, Y., and Saito, A. 2008. Evaluation of resistance to complex infection of SPFMV in transgenic sweet potato. Breed. Sci. 58:243-250.
129. Okada, Y., Saito, A., Nishiguchi, M., Kimura, T., Mori, M., Hanada, K., Sakai, J., Miyazaki, C., Matsuda, Y., and Murata, T. 2001. Virus resistance in transgenic sweetpotato [*Ipomoea batatas* L. (Lam)] expressing the coat protein gene of sweet potato feathery mottle virus. Theor. Appl. Genet. 103:743-751.
130. Okpul, T., Maltby, J., Guaf, E., Coleman, E. A., Dennien, S., Harding, R. M., Dieters, M. J., and Godwin, I. D. 2011. Cultivar-specific effects of pathogen testing on storage root yield of sweetpotato, *Ipomoea batatas*. Ann. Appl. Biol. 158:288-296.
131. Opiyo, S. A., Ateka, E. M., Owuor, O., Manguro, L. O. A., and Miano, D. W. 2010. Development of a multiplex PCR technique for simultaneous detection of *Sweet potato feathery mottle virus* and *Sweet potato chlorotic stunt virus*. J. Plant Pathol. 92:363-366.
132. Paprotka, T., Boiteux, L. S., Fonseca, M. E. N., Resende, R. O., Jeske, H., Faria, J. C., et al. 2010. Genomic diversity of sweet potato geminiviruses in a Brazilian germplasm bank. Virus Res. 149:224-233.
133. Parrella, G., De Stradis, A., and Giorgini, M. 2006. *Sweet potato feathery mottle virus* is the causal agent of Sweet Potato Virus Disease (SPVD) in Italy. Plant Pathol. 55:818.
134. Perez-Egusquiza, Z., Ward, L. I., Clover, G. R. G., and Fletcher, J. D. 2009. Detection of *Sweet potato virus 2* in sweet potato in New Zealand. Plant Dis. 93:427.
135. Pozzer, L., Dusi, A. N., Silva, J. B. C., and Kitajima, E. W. 1994. The rate of reinfection of virus-free sweet potato plants by sweet potato feathery mottle virus under field conditions. Fitopatol. Bras. 19:231-234.
136. Rännäli, M., Czekaj, V., Jones, R. A. C., Fletcher, J. D., Mu, L., Davis, R., Gweyer, D. I., Coutts, B., and Valkonen, J. P. T. 2008. Molecular genetic characterization of *Sweet potato virus G* (SPVG) isolates from areas of the Pacific Ocean and southern Africa. Plant Dis. 92:1313-1320.
137. Rännäli, M., Czekaj, V., Jones, R. A. C., Fletcher, J. D., Mu, L., Davis, R., and Valkonen, J. P. T. 2009. Molecular characterization of *Sweet potato feathery mottle virus* (SPFMV) isolates from Easter Island, French Polynesia, New Zealand and southern Africa. Plant Dis. 93:933-939.
138. Roy, Y., and Nassuth, A. 2005. Detection of plant genes, gene expression and viral RNA from tissue prints on FTA cards. Plant Mol. Biol. Rep. 23:383-395.
139. Roy-Macauley, H. 2002. Improving the livelihood of the poor in Africa using crop biotechnology. Presented at the first IFS-CODESRIA workshop on Sustainable Agriculture Initiative. Kampala, Uganda. http://www.codesria.org/IMG/pdf/Macauley.pdf.
140. Rukarwa, R. J., Mashingaidze, A. B., Kyamanywa, S., and Mukasa, S. B. 2010. Detection and elimination of sweetpotato viruses. Afr. Crop Sci. J. 18:223-233.
141. Ryu, K. H., and Choi, S. H. 2002. Molecular detection and analysis of *Sweet potato feathery mottle virus* from root and leaf tissues of cultivated sweet potato plants. Plant Pathol. J. 18:12-17.
142. Schaefers, G. A., and Terry, E. R. 1976. Insect transmission of sweetpotato disease agents in Nigeria. Phytopathology 66:642-645.
143. Sim, J., Valverde, R. A., and Clark, C. A. 2000. Whitefly transmission of *Sweetpotato chlorotic stunt virus.* Plant Dis. 84:1250.
144. Sim, J., Valverde, R., Clark, C., and Chun, S.-C. 2008. Virus-like particles and cellular changes in plants infected with sweetpotato viruses. Plant Pathol. J. 24:36-45.
145. Simmons, A. M., Ling, K., Harrison, H. F., Jr., and Jackson, D. M. 2009. *Sweet potato leaf curl virus*: Efficiency of acquisition, retention and transmission by *Bemisia tabaci* (Hemiptera: Aleyrodidae). Crop Prot. 28:1007-1011.
146. Smith, M. 2005. Born-again crops give hope to Zimbabwean farmers: Ian Robertson and his colleagues have found a way to free staple crops from viruses, with dramatic results for their growers. The Free Library. http://www.guardian.co.uk/world/2005/feb/18/outlook.development.
147. Souto, E. R., Sim, J., Chen, J., Valverde, R. A., and Clark, C. A. 2003. Properties of strains of sweet potato feathery mottle virus and two newly recognized viruses infecting sweetpotato in the United States. Plant Dis. 87:1226-1232.
148. Tairo, F., Jones, R. A. C., and Valkonen, J. P. T. 2006. Potyvirus complexes in sweetpotato: Occurrence in Australia, serological and molecular resolution, and analysis of the Sweet potato virus 2 (SPV2) component. Plant Dis. 90:1120-1128.
149. Tairo, F., Kullaya, A., and Valkonen, J. P. T. 2004. Incidence of viruses infecting sweet potato in Tanzania. Plant Dis. 88:916-920.
150. Tairo, F., Mukasa, S. B., Jones, R. A. C., Kullaya, A., Rubaihayo, P. R., and Valkonen, J. P. T. 2005. Unravelling the genetic diversity of the three main viruses involved in Sweet Potato Virus Disease (SPVD), and its practical implications. Mol. Plant Pathol. 6:199-211.
151. Takano, Y., Amemiya, A., and Ino, M. 2006. Deterioration in varietal resistance to sweet potato soil rot disease by virus-free treatment. Annu. Rep. Kanto-Tosan Plant Prot. Soc. 53:29-33.
152. Tugume, A. K., Cuellar, W. J., Mukasa, S. B., and Valkonen, J. P. T. 2010. Molecular genetic analysis of virus isolates from wild and cultivated plants demonstrates that East Africa is a hotspot for the evolution and diversification of *Sweet potato feathery mottle virus*. Mol. Ecol. 19:3139-3156.
153. Tugume, A. K., Mukasa, S. B., Kalkkinen, N., and Valkonen, J. P. T. 2010. Recombination and selection pressure in the ipomovirus *Sweet potato mild mottle virus* (Potyviridae) in wild species and cultivated sweetpotato in the centre of evolution in East Africa. J. Gen. Virol. 91:1092-1108.
154. Tugume, A. K., Mukasa, S. B., and Valkonen, J. P. T. 2008. Natural wild hosts of *Sweet potato feathery mottle virus* show spatial differences in virus incidence and virus-like diseases in Uganda. Phytopathology 98:640-652.
155. Untiveros, M., Fuentes, S., and Kreuze, J. 2008. Molecular variability of sweet potato feathery mottle virus and other potyviruses infecting sweetpotato in Peru. Arch. Virol. 153:473-483.
156. Untiveros, M., Fuentes, S., and Salazar, L. F. 2007. Synergistic interaction of *Sweet potato chlorotic stunt virus* (*Crinivirus*) with carla-, cucumo-, ipomo-, and potyviruses infecting sweetpotato. Plant Dis. 91:669-676.
157. Untiveros, M., Quispe, D., and Kreuze, J. 2010. Analysis of complete genomic sequences of isolates of the *Sweet potato feathery mottle* virus strains C and EA: Molecular evidence for two distinct potyvirus species and two P1 protein domains. Arch. Virol. 155:2059-2063.
158. Valli, A., Lopez-Moya, J. J., and Garcia, J. A. 2007. Recombination and gene duplication in the evolutionary diversification of P1 proteins in the family Potyviridae. J. Gen. Virol. 88:1016-1028.
159. Valverde, R. A., Clark, C. A., and Valkonen, J. P. T. 2007. Viruses and virus disease complexes of sweetpotato. Plant Viruses 1:116-126.
160. Valverde, R., and Moreira, M. A. 2004. Identificación de virus en el cultivo de camote (*Ipomoea batatas* L.) en Costa Rica. Agron. Mesoam. 15:1-7.
161. Valverde, R. A., Sim, J. G., and Lotrakul, P. 2004. Whitefly transmission of sweet potato viruses. Virus Res. 100:123-128.
162. Vetten, H. J., Hoyer, U., Maiss, E., and Jelkmann, W. 1996. Serological detection and discrimination of geographically diverse isolates of sweet potato sunken vein closterovirus. (Abstr.) Phytopathology 11(Suppl.):100.
163. Villordon, A. Q., and LaBonte, D. R. 1996. Genetic variation among sweetpotatoes propagated through nodal and adventitious sprouts. J. Am. Soc. Hortic. Sci. 121:170-174.
164. Wambugu, F. M. 1991. In vitro and epidemiological studies of sweet potato (*Ipomoea batatas* (L.) Lam.) virus diseases in Kenya. Ph.D. thesis. Univesity of Bath, Bath, UK.
165. Wang, H., Qi, M., and Cutler, A. J. 1993. A simple method of preparing plant samples for PCR. Nucleic Acids Res. 21:4153-4154.
166. Wang, L.-Y., Chen, K.-C., Chen, T.-C., and Yeh, S.-D. 2007. Nucleotide sequence analysis and detection application of the coat protein gene of *Sweet potato latent virus* isolated from central Taiwan. Plant Pathol. Bull. 16:141-148.
167. Wang, Q. C., Bart, P., Engelmann, F., Lambardi, M., and Valkonen, J. P. T. 2009. Cryotherapy of shoot tips: A technique for pathogen eradication to produce healthy planting material and for cryopreservation of healthy plant genetic resources. Ann. Appl. Biol. 154:351-363.
168. Wang, Q. C., and Valkonen, J. P. T. 2008. Elimination of two viruses which interact synergistically from sweetpotato using shoot tip culture and

cryotherapy. J. Virol. Methods 154:135-145.
169. Wasswa, P., Otto, B., Maruthi, M. N., Mukasa, S. B., Monger, W., and Gibson, R. W. 2011. First identification of a sweet potato begomovirus (sweepovirus) in Uganda: Characterization, detection and distribution. Plant Pathol. DOI: 10.1111/j.1365-3059.2011.02464.x.
170. Wolters, P., Collins, W., and Moyer, J. W. 1990. Probable lack of seed transmission of sweet potato feathery mottle virus in sweet potato. HortScience 25:448-449.
171. Wosula, E., Clark, C. A., and Davis, J. A. 2011. Potyvirus and insect vector movement in Louisiana sweetpotato fields. (Abstr.) Phytopathology 91:S270.
172. Yamasaki, S., Sakai, J., Kamisoyama, S., and Hanada, K. 2009. Characterization of an isolate belonging to the common strain group of *Sweetpotato feathery mottle virus* obtained from sweetpotato in Japan. Jpn. J. Phytopathol. 75:156-163.
173. Yinghong, G., Haoru, T., and Yizheng, Z. 2006. Cloning and sequence analysis of coat protein gene of sweet potato virus G. Chinese Agric. Sci. Bull. 22:50-55.
174. Zhang L.-M., Wang, Q.-M., Ma, D.-F., and Wang, Y. 2005. Major viruses and effect of major virus diseases and virus-eliminating meristem culture on sweetpotato yield and quality in China. Acta Bot. Boreali-Occidentalia Sin. 25:316-320.
175. Zhang, S.-C., and Ling, K.-S. 2011. Genetic diversity of sweet potato begomoviruses in the United States and identification of a natural recombinant between sweet potato leaf curl virus and sweet potato leaf curl Georgia virus. Arch. Virol. 156:955-968.


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