



## **Assessment of the empowerment of sweetpotato farmers for on-farm virus detection and production of clean planting material in Uganda**

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### **Abstract**

Viral diseases significantly constrain sweetpotato production. While laboratory-based virus cleaning techniques can generate clean planting material, they are often costly and inaccessible to smallholder farmers. This study evaluated a low-cost, farmer-led approach for virus detection and production of virus-free vines using *Ipomoea setosa* as an indicator plant. Farmers' groups in Iganga, Bukedea and Mpigi districts were trained on field-based virus detection and macropropagation of virus-free sweetpotato vines in the screenhouses. The training included practical sessions on vine sampling, grafting onto *I. setosa*, symptom observation, and propagation of clean material. Ten farmers from each of the three districts (Iganga, Bukedea and Mpigi) were trained as future trainers of trainees. After training, farmers' results on virus indexing and macropropagation were similar to those of experts, indicating that the empowerment was successful. Symptoms of viral infections were studied on *I. setosa* grafted with

apparently healthy sweetpotato vines. *Sweet potato feathery mottle virus* (SPFMV) was the most prevalent across all sites, followed by *Sweet potato chlorotic stunt virus* (SPCSV). *Sweet potato leaf curl virus* (SPLCV) was less common and detected only in Iganga and Bukedea. Mpigi had the highest virus incidence (54% SPFMV, 13% SPCSV and 4% Sweet Potato Virus Disease [SPVD]), followed by Iganga (35% SPFMV, 2% SPCSV and 4% SPLCV) and Bukedea (5% SPFMV, 2% SPCSV and 1% SPLCV). Within three months, trained farmers produced 103,610 vines in Iganga, 107,798 in Mpigi and 97,237 in Bukedea. These results demonstrate the feasibility of a decentralized clean seed production system. Scaling up this approach nationally can enhance sweetpotato productivity, household incomes, and food security.

Key words: Clean seed systems, *Ipomoea setosa*, macropropagation, smallholder farmers

## Introduction

Sweetpotato is among the most important food crops in East Africa, with Uganda having the largest production. However, virus infections greatly constrain sweetpotato production (Aritua *et al.*, 2007). Although most viruses in single infections are asymptomatic, they significantly impact yield. *Sweet potato feathery mottle virus* (SPFMV), the aphid-transmitted most prevalent virus in Uganda, causes negligible symptoms but garners yield losses of up to 50% (Adikini *et al.*, 2015). *Sweet potato chlorotic stunt virus* (SPCSV), a whitefly-transmitted second most prevalent virus, causes mild symptoms, also with reported yield losses up to 50% (Adikini *et al.*, 2015). Mixed infections of SPFMV and SPCSV are synergistic and they cause a severe disease known as Sweet Potato Virus Disease (SPVD), and yield losses of up to 98% has been reported (Adikini *et al.*, 2015). Unlike single infections, SPVD affected plants become more conspicuous to farmers so that they are not selected as future planting material. *Sweet potato leaf curl virus* (SPLCV) is the third most prevalent virus with mostly asymptomatic infections on many sweetpotato plants (Wasswa *et al.*, 2011), yet it can cause up to 26% yield loss (Clark and Hoy, 2006). *Sweet potato mild mottle virus* (SPMMV) and *Sweet potato chlorotic fleck virus* (SPCFV) are also among the asymptomatic viruses known to occur in Uganda (Aritua *et al.*, 2007).

In spite of these reports, previous surveys and research have shown that a big proportion of sweetpotato plants in farmers' fields is virus free (Wasswa *et al.*, 2011). This is due to sweetpotato resistance mechanism(s) expressed as recovery, dark-green-islands, and reversion from virus infections (Moore *et al.*, 2001; Wasswa *et al.*, 2011; Ssamula *et al.*, 2020). In the recovery effect, plants respond symptomatically following virus infection, although later developing leaves that are symptomless emerge, accompanied by a marked decline of virus titer (Moore *et al.*,

2001). Similarly, in the dark-green-islands response, virus titer is greatly reduced in asymptomatic patches of green tissue surrounded by symptomatic areas of mosaic disease (Moore *et al.*, 2001). Reversion is the extreme of recovery and dark-green-islands and leads to complete healing of previously virus-infected plants (Ssamula *et al.*, 2020; Andreason *et al.*, 2024). Recovery, dark-green-islands and reversion are due to gene silencing, a virus surveillance system present in all plants. These natural resistance mechanisms ensure that field plants always have some proportion that is virus free. Recovery and reversion have been observed and exploited in a number of plant-virus pathosystems. In cassava, for example, resistance to *African cassava mosaic virus* (ACMV) through reversion is exploited as a control strategy (Gibson and Otim-Nape, 1997) but, in cassava, symptoms are conspicuous so reversion can easily be observed, and hence appreciated, as it happens.

The population of virus-free field sweetpotato plants can be exploited for the decentralized clean seed production by the farmers instead of relying on the expensive tissue culture-based material from centrally located laboratories. Hitherto, the importance of using such naturally occurring virus-free field plants in sustaining sweetpotato productivity has not been explored. The current sweetpotato seed system in Uganda is of many smallholder farmers cyclically using field planting materials sourced from own seed, friends and neighbors but for which no virus indexing has been undertaken. The current study was intentional on performing field-based testing and propagation of the planting material commonly used by farmers. In this study, selected farmers were trained, in a participatory approach, the aspects of field-based sweetpotato virus indexing by plant infectivity assays using *Ipomoea setosa* and greenhouse macropropagation of the identified virus-free plants. *Ipomoea setosa* is known to be a suitable indicator plant for effectively testing for many sweetpotato viruses simultaneously, in both single and mixed virus infections (Wasswa *et al.*, 2022). Additionally, previous research shows that direct macropropagation of sweetpotato in the greenhouse rapidly multiplies large quantities of high-quality planting material and with root yield comparable to that of tissue-culture derived planting material (Wasswa *et al.*, 2022). Here, we report on community-based sweetpotato virus detection and multiplication of virus-free planting material. This initiative will sustainably contribute to solving the challenge of poor-quality sweetpotato seed in Uganda.

## Materials and methods

### *Communal indexing for sweetpotato virus infections using I. setosa*

Virus indexing was done in three greenhouses on farms of purposely selected prospective farmers from eastern Uganda (Iganga and Bukedea districts) and central Uganda (Mpigi district). The farm in Iganga district was at Kasolo village, Bulamagi

subcounty, 120 kilometers from Kampala capital city, at latitude of 0°36'25.43" N and longitude of 33°29'20.63" E, and an altitude of ~1095 m above sea level (asl). In Bukedea district, the farm was at Kachul village, Koena sub county, 230 kilometers from Kampala capital city, at latitude of 1°20'26.03" N and longitude of 34°22'60.3" E, and an altitude of ~1120 m asl. The farm in Mpigi district was at Kikoota-Ndiye village, Nkozi sub county, 200 kilometers from Kampala capital city, at latitude of 0°11'23.93" N and longitude 30°58'22.33" E, and an altitude of ~1173 m asl. These districts are among the major sweetpotato growing areas in Uganda (Ddumba *et al.*, 2014). Participatory farmers' training on virus indexing was led by a virologist on the project team through lectures, videos and hands on skilling. Ten farmers (screenhouse host inclusive as their leader), from each of the farmers' groups (Mukama Afaayo in Iganga, Kakibu in Bukedea and Nindye in Mpigi) were considered. Therefore, 10 sweetpotato fields/gardens were assessed per district/farmers' group, with one garden from each farmer.

While working with these farmers, preferred cultivars/landraces (reported by farmers to have good yield and/or taste or resistance to viruses) were noted. For farmers preferred sweetpotato cultivars, a field was assessed and all cultivars noted using breeders' names. For landraces, names were adopted according to farmers' names. A total of seven cultivars were identified, six of which were improved cultivars. Five of the improved cultivars (NASPOT 1, NASPOT 7, NASPOT 8, NASPOT 13 and Ejumula) were grown in all the three districts and these were used for communal virus indexing. In addition, SPK 004 (an improved cultivar also known as Kakamega) was grown in Iganga while landrace 'Export' was grown in Mpigi districts. Farmers often grew more than one sweetpotato cultivar per field.

Shoot scions (~5 cm) were cut from 100 asymptomatic sweetpotato field plants of farmer preferred landraces/cultivars from each of the 10 individual farmer gardens. The 100 scions from each garden were singly side grafted to one week-old virus-free indicator plant (*I. setosa*) for virus identification, where the farmer (garden owner) and the expert grafted 50 scions each. The grafted *I. setosa* seedlings were monitored for symptom development for 4 weeks. To ensure that the *I. setosa* used in this study was virus free and to eliminate any possibility of seed virus transmission, seed production from *I. setosa* used in these experiments was closely monitored in an insect-proof screenhouse. In addition, mock inoculated *I. setosa* plants were included as negative control plants.

#### *Data collection*

Observations were taken on the ability of farmers to effectively graft the sweetpotato scions to *I. setosa* by taking note of the percentage survival of grafted scions by each farmer in comparison to the survival of grafts done by the expert. Symptom

development on *I. setosa* such as chlorosis, mottling, vein clearing, rugosity, leaf curling, severe leaf distortion and stuntedness typical of virus infections was monitored for 4 weeks. *I. setosa* is very sensitive to sweetpotato virus infections and starts developing symptoms within one week after grafting, and by fourth week all infections are fully expressed (Clark and Moyer, 1988). Therefore, entries were a ‘yes’ (positive/infection) or ‘no’ (negative/no infection) on *I. setosa*, accordingly. Observations were also made on the ability of farmers to generally identify the virus infection symptoms on *I. setosa* by comparing their results with those of experts. For confirmation of virus infections, virus indexing was undertaken in the tissue culture and virology laboratory at Makerere University Agricultural Research Institute, Kabanyolo. Virus infection confirmation was by immunological diagnostic technique involving the use of Nitrocellulose Membrane Enzyme Linked Immunosorbent Assay (NCM-ELISA) (Fuentes *et al.*, 2019) by random sampling of 270 sweetpotato scions from each screenhouse 4 weeks after grafting. A primary virus-specific antibody (IgG) was used to capture the antigen on the membrane, and this specific antibody-antigen combination was detected by a secondary enzyme-labeled antibody by means of BCIP (5-Bromo-4-Chloro-3-Indoly-Phosphate) and NBT (NitroBlue Tetrazolium Chloride) substrate solution. Viruses targeted for NCM-ELISA included SPFMV, SPCSV, *Sweet potato mild mottle virus* (SPMMV) and *Sweet potato chlorotic fleck virus* (SPCFV). These viruses are either important in Uganda or have been previously identified there. *Sweet potato leaf curl virus* was not targeted for NCM-ELISA test since there are no antibodies already designed for the virus.

The number of plants that induced typical symptoms of a particular virus infection on *I. setosa* and/or confirmed with NCM-ELISA at each farm were recorded. Disease incidence per district was calculated as a percentage using the formula below (Clive, 1974).

$$\text{Disease incidence \% per district} = \frac{n(1) + n(2) + n(3) + \dots + n(10)}{N} \times 100$$

Where: n(1), n(2), n(3) .....: refers to the number of disease infected plants at sampling points/farms number 1, 2, 3 up to 10 in a district.

N ..... refers to total number of plants surveyed in a district.

#### *Communal macropropagation of virus-free sweetpotato vines*

Here, sweetpotato cultivars NASPOT 1, NASPOT 13 and Ejumula were used. Cultivar Ejumula is susceptible while NASPOT 1 is moderately resistant, and NASPOT 13 is highly resistant to virus infections (Ssamula *et al.*, 2020). These cultivars were all popular in Iganga, Bukedea and Mpigi districts. Sweetpotato

scions surviving on symptomless *I. setosa* and confirmed with NCM-ELISA to be virus free were encouraged to grow rapidly by trimming off the *I. setosa* shoots at around the graft position. When each scion grew to a size of approximately 14 internodes, they were harvested for macropropagation. Still, a participatory approach was used to train 10 prospective farmers from each district (Iganga, Bukedea and Mpigi) a macropropagation technique using the on-farm screenhouses, following a method by Wasswa *et al.* (2022). The training of farmers was by videos, lectures and hands on skilling led by the agronomist on the project team. Well grown-up virus-negative scions (with ~14 internodes) harvested from *I. setosa* root stocks were cut into short two-node vines. The two-node vines were planted in a screenhouse at spacing of 15 cm x 15 cm – with one node buried in soil – in flat-top ridges of 3 m x 1 m x 0.15 m (L x W x H), each ridge accommodating 140 vines. The vines were tendrilled (trained using tendrils) on strings to encourage vertical growth. Each ridge was planted with a single cultivar with a spacing of 0.3 m between ridges. Each farmer and the expert per district separately macropropagated three cultivars on three ridges, giving a total of 33 ridges. The ridges were arranged in a completely randomized design. The ridge soil mixture consisted of 3:1:1 ratio of screenhouse soil: matured cow manure: lake sand. One month after planting, vines in a ridge were harvested leaving two nodes at the base for regrowth. The harvested vines were sliced into two-node cuttings for further multiplication at similar spacing in new same size ridges in the same screenhouse. This process was repeated monthly for two more rounds until the entire screenhouse was occupied with ridges of vines. The accumulated vines per cultivar per farmer (or expert) were harvested and recorded at the end of 3 months. These harvested vines provided planting material for the field demonstration yield trials. The surplus of the material was given to farmers for open field crop production. Insecticide spraying with imidacloprid, at a rate of 30 ml per 20 litres of water, was done weekly to eliminate any whiteflies and aphids that could transmit the viruses. Plants in the screenhouses were also watered daily.

#### *Data collection*

Observations were made on the effectiveness of farmers to macropropagate virus-free sweetpotato vines in comparison to the expert agronomist – by computing the percentage survival of vines planted by each farmer vs. agronomist. Data was also collected for number of vines (planting material) produced by the farmers vs. the agronomist for the different cultivars at the end of three months.

#### *Demonstration of yield advantage from the use of virus-free sweetpotato planting material*

Demonstration gardens using three different sweetpotato cultivars (NASPOT 1, NASPOT 13 and Ejumula) were set up in open fields on-farm in Mpigi and Iganga, following normal agronomic practices. These demonstrations were done to compare

yields of macropropagation-derived virus-free planting material and the visually healthy planting material normally used by farmers. The treatments were laid in a complete randomized block design with three replicates (blocks). The treatments (plots) for each cultivar included four mounds planted with macropropagation-derived material and four mounds planted with visually healthy farmer-sourced material. Each block contained all combinations of planting material categories (farmer-sourced, and macropropagation-derived material) and sweetpotato cultivars (NASPOT 1, NASPOT 13 and Ejumula). The planting material categories for the different cultivars were randomly assigned to plots within each block. Each mound was 1.2 m in diameter and height of ~0.5 m, thus a plot of four mounds was equivalent to 0.000576 ha. Each mound had three vines planted separately on the top. There was a 2 m gap between blocks. Plants were harvested 16 weeks after planting and data recorded separately for each planting material category per cultivar.

#### *Data collection*

Data were collected on total root weight, marketable root weight (between 200–750 g), total root number and marketable root number, and mean root weight and number, were computed accordingly .

#### *Data analysis*

The responses including virus incidence, percentage scion survival in the screen house trial; and four yield advantage parameters (total root weight, marketable root weight, total root number and marketable root number) in the field trial were analyzed using a linear mixed model with model structure varying by response variable. For virus incidence and the yield advantage parameters, virus type and district were specified as fixed effects. For percentage scion survival, expert level and district were included as fixed effects. In all models, replication nested within district was included as a random effect. The analyses were conducted in R version 4.5.1 (2025-06-13 ucrt; R Core Team, 2025). Assumptions of normality and homogeneity of variance were assessed using quantile–quantile (Q–Q) plots and residuals versus fitted values plots, respectively. As these assumptions were violated for marketable root number, the variable was log-transformed prior to analysis. Then, type III analysis of variance (ANOVA) tables were generated with Satterthwaite approximation for denominator degrees of freedom. These ANOVAs were used to compare means of fixed effects for all responses. Post-hoc tests for separation of the means were done using Tukey’s multiple comparisons procedure.

Binary virus prevalence data (yes (presence) = 1, no (absence) = 0) were analyzed using generalized linear models with a binomial error distribution and logit link. Because samples originated from multiple farms, evaluation was done first on whether baseline infection levels differed among farms sufficiently to justify a random effect. This was

done by comparing a generalized linear mixed model with a random intercept for farm against a corresponding GLM without the random effect. The farm term nested within a district was treated as a grouping factor defining random intercepts, allowing baseline infection rates to vary among farms. This step ensured that potential clustering of observations within farms would not bias fixed-effect estimates. The two models were compared using a likelihood-ratio test (LRT) based on model deviances. As variance components in mixed models are constrained to be non-negative, the null hypothesis that the farm-level variance equals zero lies on the boundary of the parameter space. To address this, the p-value for the LRT was computed using a boundary-corrected chi-square distribution ( $(\frac{1}{2} \cdot \chi^2)$ ) following Self and Liang (1987) and Bolker *et al.* (2009), providing a conservative test of the random effect. The LRT indicated that adding the random term did not improve model fit ( $\Delta$ Deviance  $\approx 0$ ,  $\Delta$ df = 1, p = 0.499853) (data not shown), suggesting negligible variation among farms. Therefore, subsequent analyses were conducted using the simpler fixed-effects logistic model. Viruses with zero detection frequency (SPMMV and SPCFV) were excluded from the final model to avoid numerical instability due to complete separation. The refined model, fitted on the remaining virus-district combinations, showed adequate dispersion (0.76), indicating a good model fit. Logistic regression was used to evaluate the effects of virus type and district on prevalence, using SPFMV (the most widespread virus in Uganda (Aritua *et al.*, 2007) and Bukedea as reference categories.

Because several viruses-district combinations contained sparse data, GLM results were corroborated with exact tests. Cross-tabulation by district was performed to form categorical contingency tables. Expected cell counts were computed to evaluate whether the assumptions of the test were met. When all expected counts were  $\leq 5$ , Pearson's chi-square test of independence was considered inappropriate and hence a Fisher's exact test was preferred because it does not rely on large sample approximations.

## Results

### *Indexing for virus infections in preferred sweetpotato cultivars using I. setosa infectivity assay*

All farmers from the three districts effectively grafted scions of sweetpotato on *I. setosa* with average scion survivals of 91.2%, 89.5% and 91.8% for Iganga, Bukedea and Mpigi districts, respectively, with these survivals statistically similar ( $P < 0.05$ ) to those from grafts done by the expert (Table 1). Generally, farmers were able to tell between healthy and diseased *I. setosa* plants by looking at disease symptoms, although they could not use symptoms to differentiate between virus diseases. Specific virus infection symptoms including leaf mottling, chlorosis, curling, severe leaf distortion

Table 1. Percentage survival of the sweetpotato scions grafted by farmers and the expert scientist on to *I. setosa* for the detection of sweetpotato viruses

District	Iganga	Bukedea	Mpigi
Mean scion survival (%) by farmer	91.2 ± 1.12 <sup>a</sup>	89.5 ± 1.45 <sup>a</sup>	91.8 ± 1.59 <sup>a</sup>
Mean scion survival (%) by expert	89.2 ± 1.85 <sup>a</sup>	91.6 ± 1.82 <sup>a</sup>	92.0 ± 1.80 <sup>a</sup>

Means in the column followed by the same superscript are not significantly different (P<0.05)

and stuntedness were observed on *I. setosa* grafted with seemingly healthy sweetpotato vines (Fig. 1). These symptoms signified presence of four sweetpotato viruses including SPFMV, SPCSV, SPLCV and the Sweet Potato Virus Disease (SPVD). These infections were confirmed by NCM-ELISA (Fig. 2).

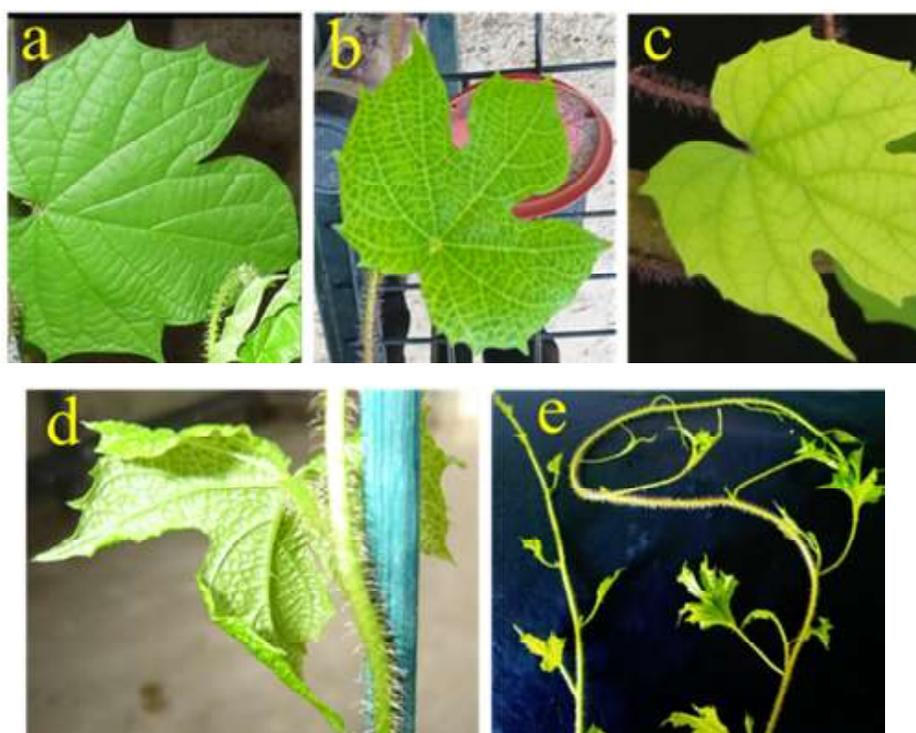


Figure 1. *Ipomoea setosa* leaves showing sweetpotato virus infection symptoms. (a) healthy leaf; (b) feathery mottling symptoms due to *Sweet potato feathery mottle virus* infection; (c) leaf chlorosis symptoms due to *Sweet potato chlorotic stunt virus* infection, (d) leaf-curling symptoms due to *Sweet potato leaf curl virus* infection; (e) severe leaf distortion symptoms due to dual infection of *Sweet potato feathery mottle virus* and *Sweet potato chlorotic stunt virus*.

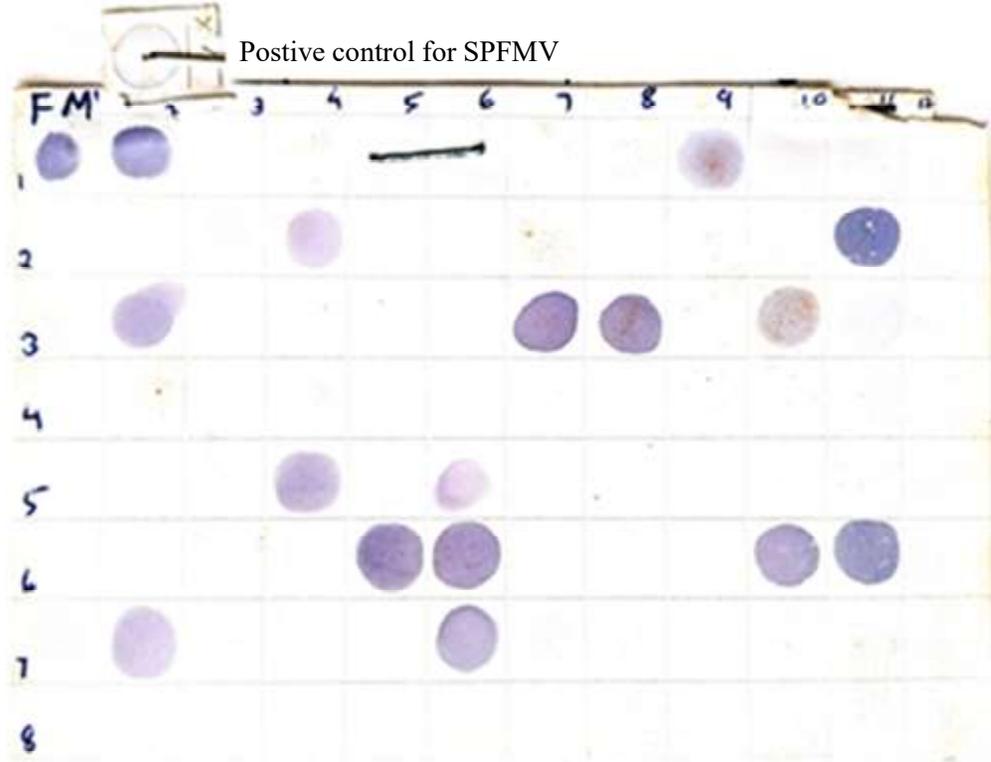


Figure 2. A representative Nitrocellulose Membrane Enzyme Linked Immunosorbent Assay (NCM-ELISA) picture for 96 (12 x 8) plant samples of sweetpotato cultivar Ejumula at a farm in Mpigi district. The purple spots indicate SPFMV-positive plants. A positive control that came with the NCM-ELISA kit is indicated at the top.

Sweetpotato viruses' incidence significantly ( $P < 0.001$ ) varied between districts as well as virus type; and there was a significant district by virus type interaction ( $P < 0.001$ ). The highest virus incidence (54% SPFMV, 13% SPCSV and 4% SPVD) was observed in Mpigi, followed by Iganga (35% SPFMV, 2% SPCSV and 1% SPLCV), and lowest in Bukedea (5% SPFMV, 2% SPCSV and 1% SPLCV) (Table 2). For all districts, the most prevalent virus from the asymptomatic vines was SPFMV at 73.3% (22 farms with the virus out of 30 farms surveyed), followed by SPCSV at 33.3% (10 farms with the virus out of 30 farms surveyed). The SPLCV was the least prevalent at 6.7% (2 farms with the virus out of 30 farms surveyed) and was only detected in Iganga and Bukedea districts (Table 2). At district level, SPFMV was most prevalent in Mpigi (100%), followed by Iganga (80%) and least prevalent in Bukedea district (40%). The SPCSV was also most prevalent in Mpigi district (60%). Sweet Potato Virus Disease (SPVD) in asymptomatic sweetpotato plants was only prevalent (40%) in Mpigi district (Table 2).

Table 2. Incidence and prevalence of sweetpotato virus diseases in three selected major sweetpotato growing districts in Uganda

Virus	Mean virus incidence			Virus prevalence				
	Bukedea	Iganga	Mpigi	Bukedea	Iganga	Mpigi	p.value	Adjusted_p.value
SPCFV	0±1.98 <sup>a</sup>	0±1.98 <sup>a</sup>	0±1.98 <sup>a</sup>	0±0 <sup>a</sup>	0±0 <sup>a</sup>	0±0 <sup>a</sup>	N/A	N/A
SPCSV	2±1.98 <sup>a</sup>	2±1.98 <sup>a</sup>	13±1.98 <sup>b</sup>	20±2.6 <sup>a</sup>	20±12.6 <sup>a</sup>	60±15.5 <sup>a</sup>	0.122	0.735
SPFMV	5±1.98 <sup>a</sup>	35±1.98 <sup>b</sup>	54±1.98 <sup>c</sup>	40±15.5 <sup>a</sup>	80±12.6 <sup>a</sup>	100±0 <sup>a</sup>	0.0109	0.0654
SPLCV	1±1.98 <sup>a</sup>	1±1.98 <sup>a</sup>	0±1.98 <sup>a</sup>	10±9.5 <sup>a</sup>	10±9.5 <sup>a</sup>	0±0 <sup>a</sup>	1.0000	1.0000
SPMMV	0±1.98 <sup>a</sup>	0±1.98 <sup>a</sup>	0±1.98 <sup>a</sup>	0±0 <sup>a</sup>	0±0 <sup>a</sup>	0±0 <sup>a</sup>	N/A	N/A
SPVD	0±1.98 <sup>a</sup>	0±1.98 <sup>a</sup>	4±1.98 <sup>a</sup>	0±0 <sup>a</sup>	0±0 <sup>a</sup>	30±0 <sup>a</sup>	0.0887	0.532

Means followed by the same superscript in a column are not significantly different at ( $P < 0.05$ )

Keys: SPCFV – *Sweet potato chlorotic fleck virus*, SPCSV – *Sweet potato chlorotic stunt virus*, SPFMV – *Sweet potato feathery mottle virus*, SPLCV – *Sweet potato leaf curl virus*, SPMMV – *Sweet potato mild mottle virus*, SPVD – *Sweet potato virus disease*; N/A – not applicable

#### *Communal macropropagation of virus-free sweetpotato vines*

The virus-free scions grew to a length of 12 -14 nodes in 5 weeks after grafting. These scions were used for macropropagation. Thirty farmers (10 from each of the districts Iganga, Bukedea and Mpigi) were trained as future trainers of trainees in screenhouse macropropagation of virus free vines. The trained farmers were as good as the expert at macropropagation, with a hundred percent survival of the vines by both the farmers and the expert, and vines showed vigorous growth within the screenhouses. Within three months, trained farmers produced 103,610 virus-free vines in Iganga, 107,798 vines in Mpigi, and 97,237 vines in Bukedea (Table 3). A total of 308,645 virus-free vines were produced in the three districts in a period of three months. Cultivar NASPOT 13 yielded the biggest number of vines in Iganga while in Mpigi and Bukedea districts, cultivar NASPOT 1 was the best yielder of vines. Cultivar Ejumula was the least yielder of vines in all the three districts (Table 3).

#### *Demonstration of yield advantage from virus-free macropropagated planting material*

Generally, cultivar, planting material source and district significantly influenced yield. Sweetpotato yield was higher in Iganga district in eastern than in Mpigi district in central Uganda, across all cultivars (Table 4). In Mpigi district, sweetpotato yield for marketable root weight ( $\text{kg ha}^{-1}$ ) was significantly ( $P < 0.05$ ) different between the different planting material categories for all the cultivars, and yield was significantly ( $P < 0.05$ ) higher for macropropagated planting material than for farmer-sourced

Table 3. Cumulative number of sweetpotato planting material (vines) produced per cultivar through macropropagation technique in the different districts over a period of three months

District	Cultivar	Cumulative number of vines			
		Planting material at start	At the end of month 1	At the end of month 2	At the end of month 3
Iganga	NASPOT 1	140	721	5,047	30,230
	NASPOT 13	140	825	4,125	48,426
	Ejumula	140	628	4,159	24,954
	Sub total				103,610
Mpigi	NASPOT 1	140	765	5,948	40,710
	NASPOT 13	140	674	5,021	35,261
	Ejumula	140	788	5,322	31,827
	Sub total				107,798
Bukedea	NASPOT 1	140	820	5,890	35,802
	NASPOT 13	140	798	5,113	32,876
	Ejumula	140	689	4,256	28,559
	Sub total				97,237
Grand total of vines					308,645

healthy-looking planting material. Marketable root number in Mpigi district varied significantly between different planting material categories for cultivar Ejumula while marketable root number for cultivars NASPOT 1 and NASPOT 13 did not significantly differ ( $P>0.05$ ) with the varying planting material categories (Table 4). In Iganga district, marketable root weight ( $\text{kg ha}^{-1}$ ) did not significantly ( $P>0.05$ ) differ between the macropropagated and farmer-sourced healthy-looking planting material (Table 4). However, marketable root number significantly varied ( $P\leq 0.05$ ) between the macropropagated and farmer-sourced healthy-looking planting material for cultivars NASPOT 1 and NASPOT 13, where the macropropagated material yielded significantly higher than farmer-sourced healthy-looking planting material (Table 4).

In terms of marketable root weight ( $\text{kg ha}^{-1}$ ) for the different planting material categories, NASPOT 13 was the best cultivar in Mpigi district while NASPOT 1

Table 4. Yield of sweetpotato for the different cultivars using different planting material categories in Mpigi and Iganga districts

District	Cultivar	Total root weight (kg ha <sup>-1</sup> )	Marketable root weight (kg ha <sup>-1</sup> )	Total root number (ha <sup>-1</sup> )	Marketable root number (mean log scale)
Mpigi	Ejumula-hl	4745±964 <sup>ab</sup>	2410±960 <sup>a</sup>	34722±2170 <sup>bc</sup>	9.58±0.107 <sup>a</sup>
	NASPOT 1-hl	3704±964 <sup>a</sup>	3057±960 <sup>ab</sup>	20833±2170 <sup>a</sup>	9.61±0.107 <sup>ab</sup>
	NASPOT 13-hl	6829±964 <sup>abc</sup>	6820±960 <sup>abc</sup>	21991±2170 <sup>a</sup>	9.79±0.107 <sup>ab</sup>
	Ejumula-macro	8218±964 <sup>abc</sup>	7466±960 <sup>bc</sup>	34143±2170 <sup>bc</sup>	10.15±0.107 <sup>b</sup>
	NASPOT 1-macro	8565±964 <sup>abc</sup>	5761±960 <sup>abc</sup>	25463±2170 <sup>abc</sup>	9.78±0.107 <sup>ab</sup>
	NASPOT 13-macro	8449±964 <sup>abc</sup>	8113±960 <sup>c</sup>	23727±2170 <sup>ab</sup>	9.89±0.107 <sup>ab</sup>
Iganga	Ejumula-hl	7755±964 <sup>abc</sup>	6192±960 <sup>abc</sup>	26042±2170 <sup>abc</sup>	9.68±0.107 <sup>ab</sup>
	NASPOT 1-hl	10822±964 <sup>c</sup>	8796±960 <sup>c</sup>	35301±2170 <sup>c</sup>	9.95±0.107 <sup>ab</sup>
	NASPOT 13-hl	8912±964 <sup>bc</sup>	8102±960 <sup>c</sup>	26042±2170 <sup>abc</sup>	9.92±0.107 <sup>ab</sup>
	Ejumula-macro	8796±964 <sup>bc</sup>	7234±960 <sup>abc</sup>	29514±2170 <sup>abc</sup>	9.87±0.107 <sup>ab</sup>
	NASPOT 1-macro	10938±964 <sup>c</sup>	9201±960 <sup>c</sup>	35880±2170 <sup>c</sup>	10.14±0.107 <sup>b</sup>
	NASPOT 13-macro	9144±964 <sup>bc</sup>	8102±960 <sup>c</sup>	29514±2170 <sup>abc</sup>	10.01±0.107 <sup>b</sup>

Means in a column with same superscript are not significantly different for each yield parameter ( $P \leq 0.05$ ). Macro is virus clean macropropagated while hl is healthy looking farmer sourced planting material

was the best cultivar in Iganga district. For the marketable root number, Ejumula was the best cultivar in Mpigi district while NASPOT 1 was the best in Iganga district (Table 4).

## Discussion

This study aimed at assessing the empowerment of farmers in decentralized sweetpotato virus indexing and macropropagation of virus-free planting material of farmers' preferred sweetpotato cultivars. This capacity building of farmers for virus indexing and production of virus-free sweetpotato planting material is the first of its kind. It helped to create farmers' awareness on prevalence of viruses in apparently healthy sweetpotato plants, virus-infection symptoms, impact of virus infections on yield; and the production of virus-free sweetpotato planting materials through the macropropagation technique. After training, farmers' results on virus indexing and macropropagation were similar to those of experts, indicating that the empowerment was successful. Empowerments like these are vital in combating sweetpotato viral diseases and should thus be extended to cover and train more farmers across the country.

Farmers mostly preferred and grew improved cultivars. This is probably because these cultivars outperform landraces in resistance to virus infections and yield. Indeed, in a study by Ssamula *et al.* (2020), it was observed that cultivar NASPOT 13 was very difficult to graft inoculate with individual viruses. Ejumula was among the popular cultivars although it is known to be susceptible to virus infections (Ssamula *et al.*, 2020). However, cultivar Ejumula is known for its high yields of up to 38 t ha<sup>-1</sup> when virus-free planting material are used (Tumwegamire *et al.*, 2007). The high yields of Ejumula were evidenced in the current study where in Mpigi district it outperformed other cultivars in terms of marketable root number when virus-free macropropagated material was used. Tumwegamire *et al.* (2007) further showed that cultivar Ejumula is preferred by the adults and children because of its appealing deep orange flesh colour.

The effectiveness displayed by farmers during the grafting of sweetpotato scions and detecting virus infection symptoms shows the feasibility of the plant infectivity assay for the decentralized virus detection. Farmers deciphered between healthy and diseased *I. setosa* plants. However, farmers employed the plant infectivity assay only as a screening tool since they could not assign specific symptoms to particular virus infections. Plant infectivity assay using *I. setosa* is a powerful virus detection technique with efficiencies comparable to polymerase chain reaction technique (Wasswa *et al.*, 2022).

The high prevalence of SPFMV and SPCSV in all the three studied districts of Uganda, moreover on asymptomatic plants of different cultivars suggests that these diseases are very important in the country. The asymptomatic virus infections observed in this study confirm the frequent latent infections in sweetpotato (Aritua *et al.*, 2007). The occurrence of virus positive but symptomless plants of all the tested cultivars may suggest that these cultivars are tolerant to virus infection (Ssamula *et al.*, 2020). Secondly, the failure to observe virus-infection symptoms on sweetpotato plants yet actually infected could as well be attributed to the fact that expression of foliar symptoms varies with season, variety, crop age, virus species and whether single or multiple infections (Ssamula *et al.*, 2020). Single infections rarely show any symptoms yet multiple infections – for example SPCSV + SPFMV – lead to severe symptoms (Wasswa *et al.*, 2022). In the absence of symptoms, farmers easily re-use infected plants as planting material and so, over several cycles of propagation through vine cuttings, crop infection can build up. Therefore, cumulatively, probably asymptomatic single virus infections cause the most damage. With planting material largely infected, yields degenerate (Adikini *et al.*, 2015). This study was thus timely for addressing the problem of poor seed quality in sweetpotato production and the awareness should be scaled up to other regions.

Similar to the findings of this study, Aritua *et al.* (2007) also found SPFMV and SPCSV to be the most prevalent sweetpotato viruses in Uganda. The likely reason for high virus prevalence may be the unrestricted movement of planting material between farmers. Viruses could rapidly spread as farmers obtain planting material from their previous crop or obtain them from their neighbours and from distant regions. In fact, this can be the major means for the spread of SPCSV since rates of SPCSV transmission by whiteflies are low (Valverde *et al.*, 2004). In a transmission experiment by Valverde *et al.* (2004), SPCSV was transmitted by whiteflies at a rate of only 15 - 20%. On the other hand, SPFMV is much more efficiently spread by aphids. For instance, in Israel, plants that were near infected plots got 51 - 85% infected just in a single growing cycle (Milgram, *et al.*, 1996) while it took only 5 - 10 weeks for experimental field plants in Brazil and USA to get 80% - 100% SPFMV-infected (Pozzer *et al.*, 1994; Bryan, 2002). This great transmission efficiency of SPFMV by the aphids coupled with spread by farmers could partly explain the generally higher prevalence of SPFMV than any other virus observed in this study.

Additionally, in SPVD affected plants, resistance is so much compromised by the SPCSV RNase3 and p22 genes allowing SPFMV to multiply freely to reach titers up to 600-fold greater than when infecting alone (Ssamula *et al.*, 2020). SPVD affected plants thus become severely stunted and easily selected against by farmers and so perhaps less apparent to vectors. Whilst these are disadvantages for both SPFMV and SPCSV, they come with the huge advantage to SPFMV of a high titer, making it readily available for acquisition by its aphid vectors and eventual spread to new plants. SPCSV appears to have no such compensating advantage, titers of some isolates even being slightly reduced in co-infections (Ssamula *et al.*, 2020). This one-sidedness of the relationship could further account for the higher prevalence of SPFMV than SPCSV in sweetpotato fields.

The prevalence of sweetpotato viruses was generally higher in central than eastern Uganda, and this again is in agreement with previous research findings (Aritua *et al.*, 2007). The high virus prevalence in central region could probably be due to the fact that Mpigi district in central region is near to equator and is characterized by an evenly distributed bimodal rainfall pattern (Majaliwa *et al.*, 2015) that allows continuous survival of sweetpotato plants, and thus the virus cycle, throughout the year. This year-round planting leads to the overlap between new and mature gardens thus maintaining some plants as reservoirs for the viruses. Availability of sweetpotato plants throughout the year also ensures abundance of food and therefore multiplication of aphids and whiteflies vectors. On the contrary, the eastern region which is further from the equator largely experiences a unimodal or unevenly distributed rainfall pattern (Majaliwa *et al.*, 2015) that breaks down the crop cycle during the prolonged dry season, and this ends up breaking down the virus cycle. This emphasizes the

importance of using virus-free planting materials for each planting season especially in Mpigi district (central Uganda) to help in breaking the virus infection cycles.

The efficiency with which farmers macropropagated the sweetpotato vines further reveals the possibility of a decentralized farmer-participatory multiplication of virus free material. Vines showed vigorous growth in the screenhouse where, 103,610, 107,798 and 97,237 vines were produced in Iganga, Mpigi, and Bukedea districts, respectively, just in a period of three months. These findings are in agreement with findings by Wasswa *et al.* (2022) who observed a high rate of sweetpotato plantlet multiplication in a screenhouse. This shows the great potential of macropropagation in the rapid production of virus free planting material. The vigorous growth of plants in the screenhouse is not being observed for the first time. In a study by Romero-Gómez *et al.* (2012) where they compared the growth of green beans (*Phaseolus vulgaris* L.) in screenhouse as well as open-field cropping systems, screenhouse treatment gave better yields. The vigorous growth could be attributed to the reduced environmental impact in the screenhouse, and thus further investigation is needed on the impact of screenhouse microenvironment on the growth of macropropagated sweetpotato plants.

Cultivar NASPOT 1 was the best yielder of vines in Mpigi and Bukedea districts yet, in Iganga district NASPOT 13 was the best yielder of vines. Cultivar Ejumula was the least yielder of vines in all the three districts. The variability between cultivars in the production of vines is probably due to the genetic differences between the cultivars and the environment variations between the districts (Hejjejar *et al.*, 2023). This could imply that vine multiplication for particular cultivars be purposely done from particular regions to maximize multiplication, but this may need more repeated studies and in additional regions using more sweetpotato cultivars before a final recommendation is drawn.

Sweetpotato root yield was higher when virus-free macropropagated planting material was used than when farmer-sourced healthy-looking planting material was used, suggesting that probably the farmer-sourced material was latently virus-infected. The latent infections of apparently healthy sweetpotato vines were witnessed when such material was tested on *I. setosa* where a good proportion of it turned out to be virus-infected. Symptomless infections in sweetpotato are mainly due to single infections and these are known to seriously impact yield. For instance, SPFMV and SPCSV single infections can lead to more than 50% yield loss (Adikini *et al.*, 2015). SPLCV was reported to reduce yields by 26% in field trial in the USA (Clark and Hoy, 2006). Sweetpotato yield from virus-free macropropagated planting material was higher in Iganga district than in Mpigi district probably because of the higher prevalence

of virus diseases observed in central than in eastern Uganda. Despite the fact that clean planting material were used, they can be re-infected when planted in open field, and more quickly in areas of high disease pressure (Bryan, 2002; Wasswa *et al.*, 2022). This signifies further the role of using high-quality virus-free planting material when establishing new sweetpotato gardens since picking planting material from previous gardens means picking already re-infected material which greatly compromises yields (Adikini *et al.*, 2015). In Iganga district, total root weight for NASPOT 1 and NASPOT 13, and marketable root weight for all the three cultivars did not differ between the macropropagated and farmer-sourced healthy-looking planting material. Again, this is attributed to the low virus incidence observed in Iganga district. This implies, when farmers in Iganga district are sourcing asymptomatic planting material from previous crops chances are high that they pick virus free material most of the times. Regarding root weight for the different planting material categories, NASPOT 13 was the best cultivar in Mpigi district while NASPOT 1 was the best cultivar in Iganga district. In terms of root number, Ejumula was the best cultivar in Mpigi district while NASPOT 1 was the best in Iganga. This variability in performance may be due to the genotype by environment interaction where different genotypes are affected differently by the environment (Hejjejar *et al.*, 2023).

## Conclusions

Farmers effectively grafted and indexed for sweetpotato viruses using the indicator plant *I. setosa*, showing the feasibility of using this technique for the field-based detection of sweetpotato viruses by farmers. Farmers also effectively macropropagated sweetpotato vines of virus-free plants, demonstrating the possibility of a decentralized clean seed production system. The higher sweetpotato root yield from virus-free macropropagated planting material compared to farmer-sourced healthy-looking planting material signifies the importance of using virus-free planting material for sweetpotato production. The adoption of this approach can enable smallholder farmers to sustainably manage viral diseases, and scaling up the approach nationally can enhance sweetpotato productivity, household incomes, and food security.

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