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Sweetpotato virus diversity and agroecological factors associated with virus transmission risk in the Papua New Guinea Highlands: Implications for a pathogen-tested (PT) seed system

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Abstract

In 2020, a survey was conducted across Eastern Highlands (EHP), Jiwaka, and Western Highlands (WHP) Provinces of Papua New Guinea (PNG) to assess sweetpotato (*Ipomoea batatas*) virus prevalence, virus vectors (aphids and whiteflies), beneficial insects, and wild *Ipomoea* species as potential virus reservoirs. The study aimed to support the pathogen-tested (PT) sweetpotato seed system by identifying factors associated with virus persistence, transmission, and re-infection. A total of 1,080 sweetpotato samples from 96 gardens were assessed using indicator plants and Nitrocellulose Membrane ELISA (NCM-ELISA). Sweetpotato plants exhibited virus-like symptoms, including leaf curling, vein clearing, distortion, purpling, and chlorosis. EHP recorded the highest disease severity (2.44%), followed by Jiwaka (2.11%), while no visible symptoms were observed in WHP. However, virus indexing detected latent infections, demonstrating the limitations of symptom-based diagnosis. *Sweetpotato feathery mottle virus* (SPFMV) was the most prevalent virus, followed by *Sweetpotato virus G* (SPVG), *Sweetpotato collusive virus* (SPCV), *Sweetpotato leaf curl virus* (SPLCV), and *Sweetpotato chlorotic fleck virus* (SPCFV), occurring as single and mixed infections. SPLV was detected as a first record for PNG, increasing the number of documented sweetpotato viruses; however, molecular confirmation is required. Seven wild *Ipomoea* species were identified as potential alternative hosts, with *I. purpurea* identified as a potential SPFMV reservoir. Aphid populations remained low ($p > 0.05$), suggesting limited vector pressure for potyvirus spread, whereas higher whitefly abundance ($p < 0.05$) indicated potential risk from whitefly-transmitted viruses, particularly *Sweetpotato chlorotic stunt virus* (SPCSV), the causal agent of *sweetpotato virus disease* (SPVD) Beneficial insects showed high diversity ($H' = 5.497$), richness ($S = 274$), and evenness ($E_H = 0.981$), indicating balanced communities supporting natural pest regulation. These findings highlight the importance of virus surveillance, pathogen-tested planting material, and integrated pest and disease management to reduce virus degeneration and sustain sweetpotato production in PNG.

Keywords: sweetpotato virus, PNG highlands, aphids, whiteflies, *Ipomoea spp.*, virus epidemiology, integrated pest and disease management.

Introduction

Sweetpotato (*Ipomoea batatas*) is one of the world's most important root and tuber crops, contributing substantially to food security, nutrition, cash income, climate resilience, particularly in low-input farming systems (Sapakhova *et al.*, 2023). In Papua New Guinea (PNG), it is the principle staple food and cash income for smallholder farmers in the highlands region, where it plays a vital role in household food security and rural livelihoods. However, sweetpotato production is constrained by several biotic factors, particularly viral diseases and sweetpotato weevils, which reduce yield, planting material quality, and overall productivity (Bugajim *et al.*, 2024; Gurr *et al.*, 2016). Viral diseases are of particular concern because they threaten the long-term sustainability of the Pathogen-Tested (PT) or virus-free, sweetpotato seed supply system.

Globally, more than 30 viruses have been reported to infect sweetpotato, occurring either as single or mixed infections that can cause severe disease complexes such as sweetpotato virus disease (SPVD) (Chabi *et al.*, 2024). In PNG, several economically important viruses have been identified, including *Sweetpotato feathery mottle virus* (SPFMV), *Sweetpotato virus G* (SPVG), *Sweetpotato mild mottle virus* (SPMMV), *Sweetpotato speckling virus* (SPMSV), *Sweetpotato ringspot virus* (SPRSV), *Sweetpotato chlorotic fleck virus* (SPCFV), *Sweetpotato collusive virus* (SPCV), and *Sweetpotato leaf curl virus* (SPLCV) (Coleman *et al.*, 2009; Wau and Komolong, 2018). These viruses occur individually or in mixed infections and pose a significant threat to the production and distribution of healthy planting material.

Accurate virus detection is fundamental to understanding virus epidemiology and implementing effective disease management. In PNG, sweetpotato viruses have traditionally been identified using graft transmission to the universal indicator plant *Ipomoea setosa*, followed by Nitrocellulose Membrane Enzyme-Linked Immunosorbent Assay (NCM-ELISA) (Dennien *et al.*, 2013; Wau & Komolong, 2018). Although these technique remain reliable for routine virus indexing, advances in molecular diagnostics, reverse transcript polymerase chain reaction (RT-PCR), quantitative PCR (qPCR), and loop-mediated isothermal amplification (LAMP), provided more more rapid and sensitive detection and improve the characterisation, of virus diversity and mixed infections (Clark *et al.*, 2012; Wanjala *et al.*, 2021). These diagnostic approaches provide an essential foundation for understanding virus epidemiology and strengthening sustainable sweetpotato seed systems in PNG.

Although laboratory-based diagnostic methods provide reliable virus identification, field assessment remains an important component of sweetpotato virus surveillance. Visual assessment of virus symptoms, together with estimate of disease incidence and severity, provides a practical approach for identifying infected plants for subsequent laboratory confirmation (Wau and Komolong, 2018). In PNG, virus-infected plants commonly exhibit symptoms such as leaf curling, vein clearing, chlorosis, purpling, leaf distortion, stunting (Wau and Komolong, 2018). However, symptoms expression varies with the virus species or strain, sweetpotato genotype, environmental conditions, and the presence of mixed infection (Adero *et al.*, 2025; Wau and Komolong, 2018).

Visual diagnosis alone is often insufficient because several economically important viruses produce mild, transient, or non-specific symptoms. For example, plants infected with SPFMV, may exhibit mild or no visible symptoms when infected singly, allowing infected vines to be unknowingly selected as planting material (Adero *et al.*, 2024; Gurr *et al.*, 2016). Likewise, symptoms associated with *Sweetpotato chlorotic stunt virus* (SPCSV) may resemble nutrient deficiencies or physiological orders, reducing the reliability of field diagnosis (Kreuze and Fuentes, 2008). These limitation highlight the importance of combining field observations with laboratory-based diagnostic techniques for accurate disease surveillance and certification of healthy planting material.

The use of PT or virus-free, planting material is widely recognised as one of the most effective strategies for reducing virus incidence and improving sweetpotato productivity (Namanda *et al.*, 2019; Ogero *et al.*, 2023; Wanjala *et al.*, 2024). In PNG, the PT sweetpotato seed system has contributed to improved crop productivity and strengthened the supply of healthy planting material for smallholder farmers (Brown *et al.*, 2019; Bugajim *et al.*, 2024; Dennien *et al.*, 2013; Gurr *et al.*, 2016; Okpul *et al.*, 2011). However, the long-term effectiveness of PT planting material is constrained by reinfection after planting, as virus-free vines become exposed to locally occurring viruses and their insect vectors under field conditions (Bugajim *et al.*, 2024; Gibson and Kreuze, 2015; Namanda *et al.*, 2019; Ogero *et al.*, 2023). Consequently, understanding the factors influencing virus reinfection is essential for maintaining the effectiveness and sustainability of the PT sweetpotato seed supply system in PNG.

Virus reinfection of PT planting material is primarily driven by insect vectors and the availability of alternative virus reservoirs within agricultural landscapes. Aphids, particularly *Myzus persicae*, *Aphis gossypii*, and *Macrosiphum euphorbiae*, are the principal vectors of potyvirus such as SPFMV and SPVG, whereas whiteflies, especially *Bemisia tabaci*, transmit SPCSV and other economical viruses (Andreason *et al.*, 2024; Mbata *et al.*, 2024). In PNG, *A. gossypii* and *B. tabaci*, are the predominant vector species and have been reported to colonise newly established sweetpotato fields soon after planting, increasing the risk of virus transmission to healthy crops (Gurr *et al.*, 2022; Wau and Komolong, 2022).

The spread of sweetpotato viruses is influenced by complex interactions among the virus, vector, host plant, and environment. Vector abundance, dispersal, host availability, surrounding vegetation, and environment conditions all influence virus transmission and the rate at which PT planting material becomes reinfected (Jangra *et al.*, 2024; Zhang *et al.*, 2024). Consequently, understanding vector abundance and distribution under field conditions is fundamental to developing effective virus surveillance and integrated disease management strategies.

Alternative host plants also contribute to virus persistence by providing reservoirs of inoculum between cropping cycles. Species within the genus *Ipomoea* are of particular importance because they can harbour sweetpotato viruses while occurring

in and around cultivated fields (Chabi *et al.*, 2024; Clark *et al.*, 2012). Although *I. setosa* is widely used as the universal indicator plant for virus indexing, several wild *Ipomoea* species have been reported as natural hosts of economically important sweetpotato viruses. For example, *I. aquatica* is susceptible to Sweetpotato leaf curl virus (SPLCV), whereas *I. hederifolia* and *I. tenuirostris*, and *I. indica* have been reported as hosts of SPFMV or *Ipomoea* yellow vein virus (IYVV) (Galbács *et al.*, 2024; Kondo *et al.*, 2022; Lotrakul *et al.*, 1998; Trenado *et al.*, 2007). These findings suggest that wild *Ipomoea* species may contribute to the persistence and spread of sweetpotato viruses within agricultural landscapes.

Despite the importance of insects vectors and alternative hosts in virus epidemiology, information on their abundance, diversity, and distribution within PNG sweetpotato production systems remains limited. Likewise, the diversity of beneficial insects that may naturally suppress aphid and whitefly populations has received little attention. This lack of epidemiology information limits the development of effective surveillance systems and integrated pest and disease management strategies to minimise virus reinfection and improve the long-term sustainability of the PT sweetpotato seed supply system.

Despite the importance of sweetpotato to food security and cash income in PNG, comprehensive information on the epidemiology of sweetpotato viruses in the highlands region remains limited. In particular, there is a lack of integrated information on virus prevalence, the abundance and distribution of virus vectors, the diversity of beneficial insects, and the occurrence of wild *Ipomoea* species that may serve as alternative virus hosts. This knowledge is essential for understanding virus reinfection pathways and strengthening the long-term susceptibility of the PT sweetpotato see supply system.

This study was conducted in 2020 as part of the Australian Centre International Agricultural Research (ACIAR) Transformative Agriculture and Enterprise Development Program (TADEP), project HORT/2014/097, "*Supporting commercial sweetpotato production and marketing in the PNG highlands*" to investigate the epidemiology of sweetpotato viruses across the Technology, Evaluation and Marketing (TEAM) zones in Eastern Highlands Province (EHP), Jiwaka Province, and Western Highlands Province (WHP). Specifically, the study aimed to: (i) assess the prevalence of sweetpotato viruses in new (2–3 months after planting) and old (>3 months after planting or previously harvested) gardens, seedbeds, and insect-proof igloos; (ii) confirm virus infections using *I. setosa* graft indexing and NCM-ELISA; (iii) quantify the abundance of adult and nymphal aphids and whiteflies to characterise virus-vector populations, with adults representing the principal virus-dispersing stage and nymphs indicating vector establishment and population pressure within gardens; (iv) evaluate the diversity of beneficial insects associated with sweetpotato production systems; (v) identify wild *Ipomoea* species occurring within and around production sites as potential alternative virus hosts.

The findings provide baseline epidemiological information information to improve understanding of virus distribution, vector ecology, beneficial insect communities, and alternative host reservoirs in the PNG Highlands. This information will support the refinement of integrate pest and disease management strategies and strengthen the long-term sustainability of the PT sweetpotato seed system.

Materials and Methods

Survey TEAM zones

Field surveys were conducted across the ACIAR TADEP TEAM zones in the Highlands of PNG (Figure 1): TEAM zone 1: Asaro, EHP (6°0'31"S 145°18'36"E); TEAM zone 2: North Whagi (5°47'S, 144°41'E) and South Whagi (5°54'34"S 144°41'00"E), Jiwaka Province and TEAM zone 3: Nebilyer (5°56'39"S 144°9'18"E), Mul Baiyer (5°32'0"S 144°9'0"E) and Hagen Central (5°51'36"S 144°14'24"E), WHP.

Surveys were conducted sequentially during the 2020 growing season: EHP (22– 26 June), Jiwaka (19 – 24 July) and WHP (9 – 14 August). Survey locations were selected based on the intensity of sweetpotato production and recommendations from the Fresh Produce Development Agency (FPDA), ensuring the representation of the major sweetpotato-producing areas within the provinces.

Under the ACIAR TADEP, 14 semi-commercial farmers were engaged as project partners in the establishment of PT sweetpotato multiplication systems. Insect-proof igloos were established at each partner farm to facilitate the multiplication and distribution of planting material, comprising four igloos in EHP, three in Jiwaka Province, and seven in WHP. These facilities supplied healthy planting material to neighbouring farming communities through the local seed multiplication networks.

Surveys were undertaken within insect-proof igloos, associated seedbeds established from sprouted PT storage roots, and surrounding sweetpotato gardens planted with either PT or farmer-sourced planting material. Both new and old gardens were surveyed to assess the occurrence of sweetpotato viruses across different production stages and planting material sources. This sampling strategy provide representative coverage of the principal sweetpotato production systems within the highlands region of PNG.

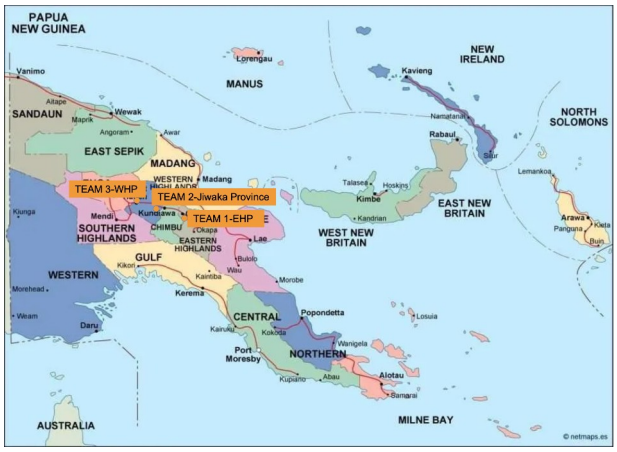

Figure 1. PNG map indicating the survey TEAM zones in the highlands.

Virus, vectors and beneficial insects survey and sampling

Virus diseases, virus vectors, and beneficial insects were surveyed in new and old sweetpotato gardens across the TEAM zones using visual assessment, quadratic sampling, yellow sticky traps, and active insect collection using sweep-net and aspirator.

Within each garden, a 50 cm × 50 cm quadrat was placed sequentially in 10 sampling points along a W- shaped transect. All sweetpotato plants within each quadrat were examined for virus symptoms, the presence of aphids and whiteflies including their nymphs, and associated beneficial insects. Adult and nymphal aphids and whiteflies were recorded separately during field assessments. Adults were assessed as the principal virus-transmitting stage, whereas nymphs were recorded to indicate vector establishment and population pressure within gardens.

Both symptomatic and asymptomatic plants were sampled following standard sweetpotato virus survey protocols (Bisimwa *et al.*, 2019; Musa *et al.*, 2021). Virus symptoms were visually assessed using a 0 – 5 disease severity scale, where 0 = no symptoms, 1–2 = mild, 3 = moderate, 4 = severe, and 5 = very severe symptoms, following established assessment methods (David *et al.*, 2023; Mbewe *et al.*, 2021). Disease intensity was quantified using disease incidence and disease severity:

- Disease Incidence (%) = (Number of infected plants / Total plants assessed) × 100
- Disease Severity (%) = [∑ (Rating × Number of plants at that rating) / (Total plants × Maximum rating)] × 100

These epidemiological parameters are widely used to quantify virus incidence and symptom severity in sweetpotato field studies (Dambolachepa *et al.*, 2023; Musa *et al.*, 2021).

Adult aphids and whiteflies were monitored using Sticky Aphid-Whitefly Traps (4 × 6 in; SeaBright Laboratories, USA). Sticky traps (yellow color) are widely used for monitoring flying aphid and whitefly populations due to their attraction to yellow wavelengths and effectiveness in estimating vector abundance and population dynamics (Abubakar *et al.*, 2022). Prior to field assessment, three traps were randomly installed per garden approximately 30 cm above the crop canopy using wooden stakes. After approximately 2 hours of exposure, traps were collected, labelled, and transported to the National Agricultural Research Institute (NARI) Highlands Regional Centre (HRC), Aiyura, EHP, where trapped aphids, whiteflies, and other arthropods were identified and counted.

Flying and foliage-associated arthropods were additionally collected along the W-shaped transect using sweep nets and aspirators. These methods are widely used for sampling arthropod diversity and pest–natural enemy communities in agricultural systems (Southwood and Henderson, 2000). Collected specimens were preserved in 70% ethanol or ethyl acetate and transported to the laboratory for identification using taxonomic keys and a compound microscope. Specimens were curated following standard entomological procedures (Villet, 2005). Beneficial insects were identified to species level and classified as predators or parasitoids following established entomological guidelines (Van Lenteren, 2012).

Symptomatic and asymptomatic sweetpotato vines were trimmed to approximately 25 cm, sealed in labelled zip-lock bags, and transported in insulated coolers to the NARI HRC, Aiyura, EHP. In the screenhouse, cuttings were established in plastic pots containing sterilised topsoil, sand, and chicken manure mixed in a 3:2:1 ratio. Plants were maintained under insect-proof conditions with regular manual watering and insecticide (Bifenthrin 10% EC) application during establishment prior to virus diagnosis.

Survey and sampling of wild *Ipomoea* species

Wild *Ipomoea* species occurring within and around sweetpotato gardens at the TEAM zones were surveyed to identify potential alternative hosts of sweetpotato viruses. Plants were identified in the field using standard morphological descriptor (Wood *et al.*, 2020), and their occurrence was documented photographically. Representative samples of each *Ipomoea* species were collected, labelled, and transported with the sweetpotato vine samples to the NARI HRC, Aiyura, EHP.

In the screenhouse, stem cuttings of each *Ipomoea* species were established in pots containing sterilised growing medium prepared using the same ratio of topsoil, sand, and chicken manures described for sweetpotato plant. The cuttings were maintained under insect-proof conditions for subsequent virus indexing. Once established, plants were screen for common sweetpotato viruses using NCM-ELISA following established protocols (Dennien *et al.*, 2013; Wolfenden *et al.*, 2019). The results were used to determine the potential role of wild *Ipomoea* species as alternative virus hosts.

Virus indexing using *Ipomoea setosa* and NCM-ELISA

Virus screening was conducted using *I. setosa*, a universal virus indicator, and NCM-ELISA. Viable *I. setosa* seeds were grown in pots containing sterilised topsoil inside an igloo structure and grafted with scions from both symptomatic and asymptomatic sweetpotato plants collected from farmers' fields. The serological detection of viruses was performed following the protocol described by (Dennien *et al.*, 2013).

Statistical analysis

Species diversity was quantified using the Shannon Diversity Index, denoted by H', and species evenness using the Shannon Equitability Index, denoted by E_H, and species richness (S), based on information entropy described by Shannon (1948). The respective formulae are; $H = -\sum pi \times \ln(pi)$ where: $\sum$ = A Greek symbol that means "sum", In = Natural log, and pi = The proportion of the entire community made up of species. The higher the value of H, the higher the diversity of species in a particular community. The lower the value of H, the lower the diversity. The general range of values: Low Diversity: $H' < 1.5$, Moderate Diversity: $1.5 \leq H' < 3.0$, and High Diversity: $H' \geq 3.0$.

The $E_H = H/In(S)$, where: H = The Shannon Diversity Index, S = The total number of unique species. This value ranges from 0 to 1 where 1 indicates complete evenness. The general range of values: Low Diversity: $E_H < 0.5$, Moderate Diversity: E_H 0.5-0.8, and High Diversity: $E_H \geq 0.8$.

Species richness was calculated as the total number of unique species recorded within each sampling site: S = number of observed species, where S represents the total count of distinct species present in the community, regardless of their abundance.

Mean abundance (± SE) of major insect groups across TEAM zones were analysed using GenStat (20th Edition). Differences among site means were compared using Tukey's HSD test at $p < 0.05$, with means sharing the same letter considered not significantly different.

Results

Virus survey across the TEAM zones

A total of 48 farmers (16 per site) were surveyed across the TEAM zone, covering 96 gardens comprising equal numbers of newly established and old gardens. TA total of 960 quadrat observations (10 quadrats per garden × 96 gardens) were conducted, and 1,080 sweetpotato plant samples were collected and assessed for virus-like symptoms.

The most commonly observed symptoms included leaf curling, vein clearing, leaf distortion, purpling spots, and chlorosis (Figure 2). Chlorotic spots and necrosis were the most prevalent symptoms, followed by vein clearing; leaf curling or cupping associated with chlorosis and stunting. Netted vein clearing was also observed at a low frequency.

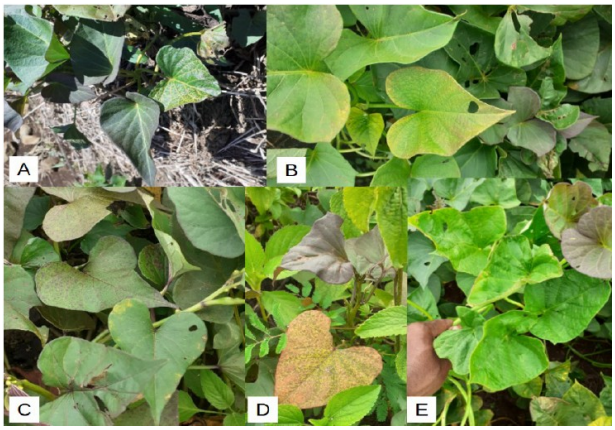

Figure 2. Commonly observed virus-like symptoms in field-grown sweetpotato: (A) vine clearing accompanied by leaf distortion, (B) vine clearing with leaf chlorosis, (C) purplish spot on leaf surfaces, (D) mottling of leaf, and (E) leaf curling/cupping and distortion.

Symptoms expression was more pronounced in old gardens compared to newly established ones. However, several fields appeared asymptomatic, but subsequent sampling confirmed the presence of latent infections.

Disease incidence and severity varied across sites and were quantified based on symptoms expression (Table 1).

Table 1. Disease incidence (DI) and severity (DS) (%) across the TEAM zones.

TEAM zone	Total plants assessed	Total infected	Avg DI (%)	Avg DS (%)	Sites with infection
EHP	320	13	4.06	2.44	Old gardens in Gimisave and Meteyufa
Jiwaka	390	24	2.11	2.11	Kurumul 2 (new and old gardens, and S/Bed)
WHP	370	0	0	0	None

Sweetpotato viruses detected

Sweetpotato viruses were detected across the TEAM zones using herbaceous grafting and NCM-ELISA, with SPFMV being the most prevalent, followed by SPVG, SPCV, SPCFV, and Sweetpotato latent virus (SPLV). SPLV was detected to be a new record in PNG. Majority of these viruses were identified in samples from new gardens (48%), followed by old gardens (43%), seedbeds (7%), and virus host plants (2%). Table 2 summarises the specific viruses identified, along with their infection status, categorised as single, dual, or multiple infections.

Table 2. Summary of viruses detected from samples collected from the TEAM zones.

TEAM zone	Infected samples/Total samples	Single infections	Dual/Co-infections	Multiple infections
EHP	36/60	SPFMV SPVG	SPFMV+SPVG	No detection
Jiwaka	44/240	SPFMV SPVG	SPFMV+SPVG	No detection
WHP	72/196	SPFMV SPVG SPCV	SPFMV+SPVG	SPFMV + SPCFV + SPLV SPVG + SPCV + SPLV

Abbreviations: Sweetpotato feathery mottle virus (SPFMV), *Sweetpotato virus G* (SPVG), *Sweetpotato collusive virus* (SPCV), *Sweetpotato chlorotic fleck virus* (SPCFV), *Sweetpotato latent virus* (SPLV).

Alternative host plants (*Ipomoea spp.*) identified

Seven *Ipomoea* species were identified across the TEAM zones as potential alternative hosts of viruses: *I. coccinea*, *I. purpurea*, *I. indica*, *I. cairica*, *I. heptaphylla*, *I. alba*, and *I. hederacea*.

Of these, only three species were successfully established under insect-proof screenhouse conditions and subsequently tested for viral infection, as the remaining species failed to survive after transportation. Among the tested species, *I. purpurea* was confirmed positive for SPFMV.

A detailed summary of the identified *Ipomoea* species, including their morphological characteristics, is provided in Appendix 1.

TEAM zone 1 – Population abundance of virus vectors

Population abundance of whiteflies and aphids including their nymphs in new and old gardens at Asaro, EHP (Figure 3). Adult whiteflies dominated across sites, peaking in new gardens at Nipuka and old gardens at Gimisave. Nymph aphids were highest at Nipuka and lowest at Kuka (new gardens), while adult aphids recorded highest at Meteyufa (new gardens) and lowest at Kuka (new gardens). Seedbeds showed higher vector abundance at Kuka than Meteyufa; none were present at other sites.

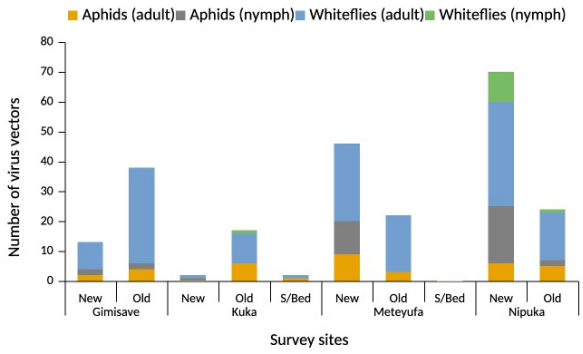

Figure 3. Virus vectors abundance in both new and old gardens at Asaro, EHP.

TEAM zone 1 – Population abundance of beneficial insects

The abundance of beneficial insects in new and old gardens at Asaro, EHP (Figure 4). Overall abundance was highest at Nipuka in both new and old gardens. Meteyufa recorded the highest counts in new gardens, Nipuka dominated in old gardens, Gimisave and Kuka records followed relatively in both new and old gardens. Seedbeds had higher abundance at Meteyufa than Kuka; none occurred at other sites.

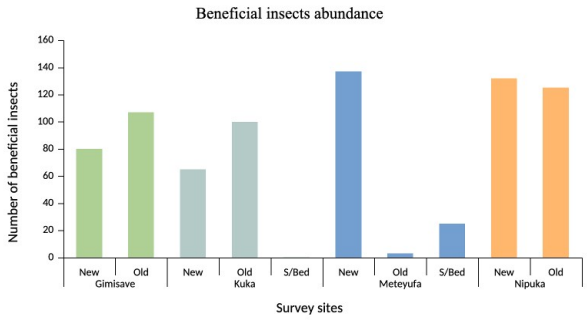

Figure 4. Beneficial insects abundance in both new and old gardens at TEAM zone 1.

TEAM zone 1 – Statistical analysis of virus vectors and beneficial insects

Table 3 summarises the mean abundance (± SE) of aphids (adult and nymphs), whiteflies, and beneficial insects across sites in Asaro, EHP. Aphid population (adult and nymphs) were low and did not differ among sites. In contrast, whitefly abundance differed, with the highest values at Nipuka and the lowest at Kuka, while Meteyufa and Gimisave were intermediate. Beneficial insect also varied, with higher abundance at Nipuka, Kuka, and Gimisave than at Meteyufa, indicating site-specific variation in natural enemy populations.

Table 3. Mean abundance (± S.E) of aphid, whitefly, and beneficial insects across sites in Asaro, EHP.

	Mean±SE			
Survey sites	Aphids (adult)	Aphids (nymph)	Whitefly (adult)	Beneficial insects
Meteyufa	0.15±0.72^{a}	0.14±0.8^{a}	0.56±1.9^{ab}	0.85±1.9^{a}
Kuka	0.08±0.72^{a}	0.01±0.8^{a}	0.14±1.9^{a}	3.00±1.9^{b}
Gimisave	0.13±0.72^{a}	0.10±0.8^{a}	0.60±1.9^{ab}	2.70±1.9^{b}
Nipuka	0.14±0.72^{a}	0.26±0.8^{a}	0.64±1.9^{b}	3.20±1.9^{b}

Means followed by the same letter within a column are not significantly different according to Tukey's HSD test ($p < 0.05$).

TEAM zone 2 – Population abundance of virus vectors

The abundance of whiteflies and aphids including their nymphs across sites in Jiwaka Province (Figure 5). Adult whiteflies were consistently more abundant, peaking at Kongabil and Kurumul2 (new gardens) and Gunn (old gardens). Nymph aphids were highest at Gunn, while adult peaks at Kongabil (new gardens). Seedbeds showed higher adult whiteflies at Gunn, while nymph aphids remained low.

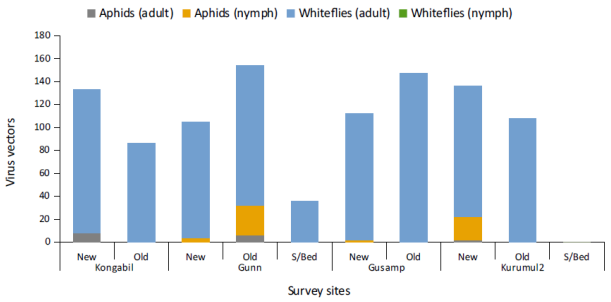

Figure 5. Virus vectors abundance in both new and old gardens at TEAM zone 2.

TEAM zone 2 – Population abundance of beneficial insects

The abundance of beneficial insects in new and old gardens across sites in Jiwaka Province (Figure 6). Abundance was highest at Kurumul2, followed by Kongabil, Gunn, and Gusamp. Kurumul2 recorded the highest counts in both garden types, while other sites showed lower and variable levels. Seedbeds showed similar abundance at Gusamp and Kurumul2.

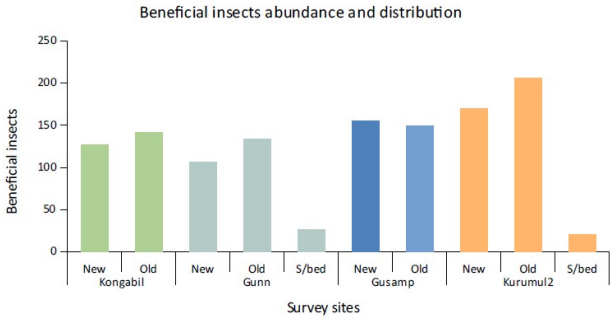

Figure 6. Beneficial insects abundance in both new and old gardens in Jiwaka Province.

TEAM zone 2 – Statistical analysis of virus vectors and beneficial insects

Table 4 summarises the mean abundance (± SE) of aphids (adult and nymphs), whiteflies, and beneficial insects across sites in Jiwaka. Aphids population (adult and nymphs) were low and did not differ among sites. Adult whiteflies abundance varied with higher population at Gunn, Gusamp, and Kongabil compared to Kurumul2. Beneficial insects also differed, with the highest abundance at Gusamp, followed by Gunn (intermediate), and lower counts at Kongabil and Kurumul2.

Table 4. Mean abundance (± S.E) of aphid, whitefly, and beneficial insects across sites in Jiwaka Province.

	Mean±SE			
Survey sites	Aphids (adult)	Aphids (nymph)	Whitefly (adult)	Beneficial insects
Gunn	0.08±0.29^{a}	0.38±1.0^{a}	2.8±1.7^{b}	3.0±1.7^{ab}
Gusamp	0.00±0.29^{a}	0.03±1.0^{a}	2.9±1.7^{b}	3.5±1.7^{b}
Kongabil	0.05±0.29^{a}	0.00±1.0^{a}	2.3±1.7^{b}	2.7±1.7^{a}
Kurumul2	0.01±0.29^{a}	0.13±1.0^{a}	1.4±1.7^{a}	2.4±1.7^{a}

Means followed by the same letter within a column are not significantly different according to Tukey's HSD test ($p < 0.05$).

TEAM zone 3 – Population abundance of virus vectors

The abundance of whiteflies and aphids across sites in WHP (Figure 7). Whiteflies dominated, peaked at Hagen Central (2) (new gardens) and Hagen Central (1) (old gardens). Aphid nymphs were highest at Nebilye (old gardens) and Hagen Central (2) (new gardens). Adult aphids occurred only at Hagen Central (1) and in seedbeds at Hagen Central (2), indicating generally low aphid occurrence across the surveyed sites.

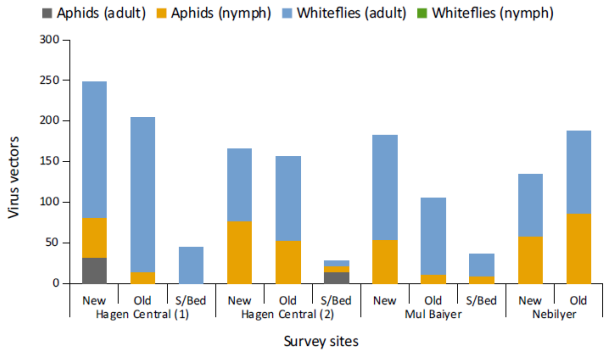

Figure 7. Virus vectors abundance in both new and old gardens in TEAM zone 3.

TEAM zone 3 – Population abundance of beneficial insects

The abundance of beneficial insects in new and old gardens across sites in WHP (Figure 8). Overall abundance was highest at Hagen Central (1), followed by Hagen Central (2), Mul Baiyer, and Nebilye. Mul Baiyer recorded the highest counts in new gardens, while Hagen Central (2) dominated in old gardens. Seedbeds showed the highest abundance at Hagen Central (1), followed by Mul Baiyer and Hagen Central (2).

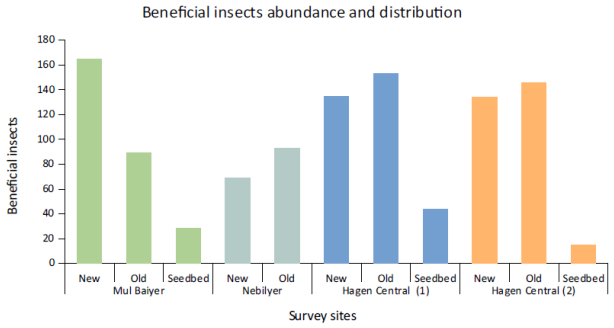

Figure 8. Virus vectors abundance in both new and old gardens in TEAM zone 3.

TEAM zone 3 – Statistical analysis of virus vectors and beneficial insects

Table 5 summarises the mean abundance (± SE) of aphids (adult and nymphs), whiteflies, and beneficial insects across sites in WHP. Adult aphid populations were consistently low and did not differ among sites. Nymph aphids were slightly higher, with no difference between sites. Adult whiteflies abundance differed among sites with highest at Hagen Central (1), followed by Nebilye, Mul Baiyer, and Hagen Central (2). Beneficial insects abundance was relatively uniform with no differences among sites.

Table 5. Mean abundance (± S.E) of aphid, whitefly, and beneficial insects across sites in WHP.

	Mean±SE			
Survey sites	Aphids (adult)	Aphids (nymph)	Whitefly (adult)	Beneficial insects
Hagen Central (1)	0.4±0.7^{a}	0.8±2.7^{a}	4.5±2.2^{c}	3.6±2.2^{a}
Hagen Central (2)	0.0±0.7^{a}	1.6±2.7^{ab}	2.4±2.2^{a}	3.5±2.3^{a}
Nebilyer	0.3±0.7^{ab}	2.4±2.7^{a}	3.5±2.2^{b}	3.5±2.5^{a}
Mul Baiyer	0.0±0.7^{a}	0.8±2.7^{a}	3.0±2.2^{ab}	3.2±2.8^{a}

Means followed by the same letter within a column are not significantly different according to Tukey's HSD test ($p < 0.05$).

Diversity of beneficial insects across TEAM zones

Beneficial insects recorded the highest Shannon diversity index across all TEAM zones, increasing from 5.307 in zone 2 and 5.637 in zone 3. Species richness followed a similar trend, increasing from 225 species in zone 1 to 284 species in zone 2 and 313 species in zone 3. Shannon evenness remained consistently high across the three zones, ranging from 0.980 in zone 1 to 0.982 in zone 2 and 0.981 in zone 3. Overall, beneficial insects exhibited the highest diversity, species

richness, and evenness among the insect groups surveyed, with zone 3 recording the greatest Shannon diversity and species richness.

Discussion

Symptomatic and latent sweetpotato virus prevalence in TEAM zones

Sweetpotato plants across the TEAM zones exhibited a range of virus-like symptoms, including leaf curling, vein clearing, leaf distortion, purpling, and chlorosis, consistent with previous observations from the PNG highlands (Wau and Komolong, 2018). Although symptom expression was slightly higher in older gardens, virus indexing detected a greater proportion of infected plants in newly established gardens (48%) compared with older gardens (43%), indicating that a substantial proportion of infections were latent and not visually detectable.

The presence of latent infections in apparently healthy plants highlights the limitations of visual assessment as the sole method for virus diagnosis. The similarity in infection levels between new and old gardens may reflect traditional farmer practices of sourcing planting vines from older gardens, where symptomless but infected materials can inadvertently introduce viruses into new production areas. Repeated use of infected planting material can contribute to virus accumulation and progressive yield decline, commonly referred to as "virus degeneration" (Dennien *et al.*, 2013; Gibson and Kreuze, 2015; Wau and Komolong, 2018).

Based on visual disease assessment, EHP recorded the highest disease severity (2.44%), followed by Jiwaka (2.11%), while no visible symptoms were observed in WHP. However, virus indexing detected single, dual, and multiple infections in symptomless plants from these areas, further demonstrating that symptom absence does not necessarily indicate virus-free status.

These findings emphasis the need to move beyond visual selection of planting materials and strengthen systematic virus indexing and pathogen-tested seed systems. Early detection of latent infections is essential for reducing virus dissemination, maintaining healthy planting materials, and improving the sustainability of sweetpotato production.

First detection of Sweetpotato latent virus (SPLV) in PNG

The detection of *Sweetpotato latent virus* (SPLV), a member of the genus *Potyvirus*, represents the first published record of this virus in PNG, increasing the number of documented sweetpotato viruses in the country from eight to nine. Although SPLV was previously detected during an ACIAR TADEP project activity, the finding was not formally reported. SPLV is transmitted by aphids in a non-persistent manner and, importantly, through infected vegetative planting material (Wang *et al.*, 2013).

SPLV is generally latent, producing few or no visible symptoms; however, infected plants can act as reservoirs and frequently occur in mixed infections with other sweetpotato viruses, potentially influencing disease expression and crop performance (Wang *et al.*, 2013). As this represents the first published detection of SPLV in PNG, molecular confirmation through RT-PCR and sequencing is recommended to validate virus identity and determine its genetic relationship with isolates from other regions.

This finding also highlights important knowledge gap regarding the distribution, incidence, genetic diversity, epidemiology, and economic impact of SPLV in PNG. Addressing these gaps will strengthen virus surveillance, support the production of virus-tested planting material, and improve integrated disease management strategies for sustainable sweetpotato production.

Limited role of aphids in virus transmission

All virus detected belonged to the Potyvirus genus, which is transmitted by aphids in a non-persistent manner and through vegetative cuttings, but not by whiteflies. However, aphid populations (adult and nymph) were consistently low across the TEAM zones, suggesting that aphid-mediated transmission was limited during the study. Therefore, the widespread occurrence of these viruses is more likely attributable to the use of infected planting material. This interpretation is consistent with traditional farmer practices in the region, where vines are commonly sourced from older, seemingly healthy gardens to establish new plots, inadvertently enabling the continued spread of viruses (Dennien *et al.*, 2013; Wau and Komolong, 2018).

Whitefly abundance and the emerging risk of whitefly-transmitted viruses

Although no whitefly-transmitted viruses were confirmed in this study, our previous virus indexing and serology assays detected faint reactions to SPCSV in PNG highlands sweetpotato fields, suggesting that the virus may be present at low incidence or remain undetected (Wau and Komolong, 2018). As whiteflies are the exclusive vectors of SPCSV, the high abundance and distribution of whitefly populations recorded across the TEAM zones indicate a potential risk for

future establishment and spread of this virus. If introduced or established, SPCSV could co-infect plants with aphid-transmitted potyviruses such as SPFMV, resulting in SPVD, one of the most destructive viral diseases of sweetpotato, with reported yield losses of up to 80 – 90% (Gibson and Kreuze, 2015; Zhang *et al.*, 2020). These findings highlight the importance of routine surveillance, molecular confirmation of suspected SPCSV infections, and IPDM strategies.

Diversity and ecological importance of beneficial insect communities across TEAM zones

Beneficial insect communities were recorded across all TEAM zones, with high diversity (H' = 5.307–5.637), evenness (E_H = 0.980–0.982), and species richness (225–313 taxa), indicating diverse and balanced natural enemy assemblages in sweetpotato production systems. Similar studies in sweetpotato agroecosystems have documented diverse predators and parasitoids, including ladybird beetles, lacewings, predatory bugs, spiders, and parasitoid wasps, which contribute to the regulation of aphids, whiteflies, and other insect pests (Gurr *et al.*, 2022; Johnson and Gurr, 2016).

The increasing diversity and richness from TEAM zone 1 to TEAM zone 3 suggests that local habitat conditions influenced beneficial insect assemblages. Greater vegetation diversity, habitat complexity, and floral resources are known to enhance natural enemy populations by providing shelter, alternative prey, and food resources (Gurr *et al.*, 2017; Landis *et al.*, 2000). These ecological functions are particularly valuable in smallholder systems where conservation biological control provides a practical and sustainable pest management option.

The consistently high evenness across TEAM zones indicates a stable community structure with limited dominance by individual taxa. Diverse natural enemy communities can suppress aphid and whitefly populations, potentially reducing secondary virus spread, although they do not prevent initial virus acquisition and transmission by mobile adult vectors (Heimpel and Mills, 2017; Snyder, 2019).

Overall, the diverse beneficial insect communities observed across the PNG Highlands represent an important ecological resource for strengthening IPDM. Conserving habitat diversity and reducing unnecessary insecticide use will help maintain natural enemy populations and improve the resilience of sweetpotato production systems.

Wild *Ipomoea* species as alternative hosts of sweetpotato viruses

Seven wild *Ipomoea* species (*I. coccinea*, *I. purpurea*, *I. indica*, *I. cairica*, *I. heptaphylla*, *I. alba*, and *I. hederacea*) were recorded across the surveyed TEAM zones, highlighting their potential role as alternative hosts for sweetpotato viruses. While *I. indica* and *I. alba* are well documented in PNG, records for *I. coccinea*, *I. purpurea*, *I. heptaphylla*, and *I. hederacea* are limited or absent from available botanical databases and herbarium collections, suggesting that their occurrence in the PNG highlands is either previously under-documented or represents a new distribution record (An Eco-sustainable World, 2025; Bergmann, 2015; EPPO Global Database, 2025).

Among the species surveyed, only *I. purpurea* tested positive for SPFMV, indicating its potential role as a virus reservoir. Its widespread occurrence across the TEAM zones suggests that it may contribute to the persistence and reintroduction of SPFMV into sweetpotato crops, particularly where infected volunteer plants and weeds occur near production fields. Similar roles of wild *Ipomoea* species as reservoirs of sweetpotato viruses have been reported in other sweetpotato growing regions, where they facilitate virus survival between cropping cycles and provide a source of inoculum for aphids-mediated transmission (Clark *et al.*, 2012; Loebenstein, 2009).

Wild *Ipomoea* species were most abundant in Jiwaka province, where disease severity was also highest (2.11%). Although this association does not demonstrate a causal relationship, it suggests that alternative host may contribute to virus persistence and increase opportunities for virus spread within sweetpotato production systems. Further studies using molecular epidemiology and vector transmission experiments are needed to determine the role of these species in the epidemiology of sweetpotato viruses in PNG. These findings highlight the importance of including wild *Ipomoea* species in virus surveillance and integrate disease management programmes, particularly where PT planting material is promoted.

This study highlights the importance of implementing a PT sweetpotato seed system in the highlands, as asymptomatic gardens demonstrate the limitations of visual virus assessment alone. The detection of SPLV increases the number of documented sweetpotato viruses in PNG from eight to nine and emphasis the need to strengthen virus surveillance. It also identified important knowledge gaps regarding the distribution, incidence, genetic diversity, epidemiology, and economic impact of SPLV in the country. The detection of viruses in wild *Ipomoea* species, particularly *I. purpurea*, suggests that these plants may serve as alternative virus hosts and contribute to virus persistence in the landscape. In addition, the diverse beneficial insect communities recorded across the sites indicate their potential role in maintaining ecological stability and support natural pest regulation. Despite low aphid populations and no current detection of whitefly-transmitted viruses, the high whitefly abundance poses a latent risk for SPVD. Collectively, these findings provide a foundation of

strengthening virus surveillance, conserving healthy planting material, and implementing IPDM strategies to improve the sustainability of sweetpotato production in PNG.

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