













INTRODUCTION

The Phytovirome

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The phytovirome refers to the collection of viruses associated with plants and their related organisms within a specific environment or ecosystem. The study of the phytovirome enhances our understanding of the diversity, interactions, and impact of viruses on plant health and ecosystems. In this introduction to the Phytobiomes Journal focus issue on the phytovirome, we underscore recent discoveries, advancements within the discipline, and the significance of viruses to the phytobiome. This special issue comprises 11 articles on diverse phytovirome topics, capturing the breadth and importance of this field of study. A recurring theme throughout this issue is the transformative role of high-throughput sequencing (HTS) technologies in phytovirome research. The articles delve into viral community ecology, virus transmission between plant hosts, insect-specific viruses, bacteriophages for biocontrol, and the discovery of new viruses and previously known viruses in new hosts. This body of work lays the groundwork for utilizing this knowledge to cultivate healthier plants by controlling plant-pathogenic viruses and harnessing beneficial viruses to improve plant health.

From Viruses to Virome

Over the past decade, HTS technologies have facilitated the discovery of a vast number of new plant-associated viruses, including plant and fungal viruses, bacteriophages, and other viruses within the phytobiome. It is now understood that plant and fungal viruses are ubiquitous in both wild and cultivated plant species, forming virus communities within individual plants (referred to as mixed infections) and across plant communities.

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Research on plant viruses, particularly those found in crops, has demonstrated that they can establish various trophic relationships with their host plants, ranging from pathogenic to beneficial. Plant and fungal viruses are integral members of the phytobiome, and their inter-kingdom roles warrant greater attention. Viruses associated with the plant holobiome, including those linked to bacteria, nematodes, and arthropods, are also of significant importance.

This *Phytobiomes Journal* focus issue on the phytovirome provides a benchmark for the current status of this research area. The papers in this collection encompass research on viral biodiversity, biogeography, community assembly, and dynamics; they describe virus evolution and emergence, multitrophic interactions, and the multipartite relationships that viruses have with each other and their hosts. Additionally, the papers address scaling across space and time, as well as issues of global concern such as food security and climate change.

Plant Virus Ecology

Plant virus ecology emerged from a history of agriculturally focused research. While there is a pressing need to understand plant viromes outside of crops, this does not mean we should leave the farm behind. Schönegger et al. (2025) recognized the value in comparing viromes across crops and their nondomesticated congeners in their study of crop and wild carrot viromes. Cultivated carrots grow alongside wild carrots in the area of study (France), which creates opportunities for viral exchanges. Using HTS, Schönegger et al. (2025) found that wild carrots harbored a greater diversity of virus taxa relative to farmed carrots and documented key differences in the virus communities of each host type. Seedborne virus communities differed considerably by host type, whereas vectorborne virus communities had more overlap, with some notable differences. First, aphid-transmitted viruses, in general, were more prevalent in cultivated carrots. Second, several aphid-transmitted virus species were uniquely found in wild carrots, despite potential vector movement between host types. Additionally, wild carrots

harbored distinct variants of specific virus species relative to those present in farmed carrots. Analyses by landscape type shed further light on mechanisms of community assembly by demonstrating that "off-type" carrots, which grow in cultivated carrot fields but resemble wild carrots phenotypically, have viromes similar to cultivated carrots when they co-occur on farms. This work demonstrates that there are distinct barriers to virus transmission between crop and wild congeners and suggests that the farm landscape itself could shape viromes independent of plant host traits.

Connectivity between cultivated and wild plant communities was also explored in a paper by Hoyle et al. (2025). They used molecular gut content analysis of Spissistilus festinus, the treehopper vector of grapevine red blotch virus, to identify host use patterns between vineyards and nonmanaged habitats. Hoyle et al. (2025) collected 205 treehoppers from 71 vineyard sites and identified DNA from 171 plant genera across 60 families in treehopper guts. These results revealed that S. festinus primarily feeds on natural vegetation, particularly Vitis species, rather than cultivated grapevines themselves. S. festinus frequently interacts with free-living Vitis californica and V. californica hybrids in riparian corridors, establishing an ecological link for virus transmission from wild plants to cultivated grapevines. S. festinus also exhibits seasonality in host preferences, with greater use of free-living *Vitis* species early in the season. This study demonstrates the power of using gut content analysis to understand the dietary history and host use patterns of herbivorous vectors and is the first study to apply these techniques to S. festinus and the recently emerged grapevine red blotch virus pathosystem. In this case, understanding host use patterns lends support to the idea that management of red blotch disease and S. festinus cannot be limited to the vineyard and must include measures to mitigate virus and vector reservoirs in natural habitats.

The study of Fouad et al. (2025) aimed to broaden knowledge about viral communities in rice landscapes in Burkina Faso and explore the diversity and epidemiological status of viruses in rice fields. Conducted between 2016 and 2019, the survey included 57 small farmers' rice fields under rainfed lowlands and irrigated areas, collecting over 2,700 rice samples along with samples from nearby wild and cultivated Poaceae. Using viral metagenomics, they unexpectedly detected maize streak virus (MSV) in rice samples. Further analyses revealed that MSV was widely distributed and highly prevalent, with MSV-G, the strain found in wild grasses prior to this study, being the most common strain and MSV-A, the maizeinfecting viral strain, being less prevalent. The pathogenicity of both MSV strains in rice was confirmed using infectious clones of the viruses. This paper highlights new epidemiological and pathogenic aspects of MSV for rice production in Burkina Faso.

Virus Discovery and Control

The work of Hackenberg et al. (2025) uses HTS, a highly sensitive methodology for the detection of known and novel viruses, for the comprehensive analysis of viral sequences associated with hemp in Colorado. Their analysis revealed seven different viruses associated with hemp. Sampling plant tissue from multiple sites and years led the authors to conclude that beet curly top virus (BCTV) is a threat to crop production. This knowledge enabled them to assay existing hemp germplasm for resistance to BCTV, which identified two hemp genotypes with lower virus titers and reduced virus symptoms. This work illustrates the power of HTS for identification of primary viral threats to specialty crops and the application of these findings for achieving outcomes for disease control.

The manuscript by West-Ortiz, Olmedo-Velarde, and colleagues (West-Ortiz et al. 2025) describes a novel virus, cotton virus A (CotVA), in Gossypium plants that groups within the family Caulimoviridae. The virus discovery was made using HTS while studying another emerging virus, cotton leafroll dwarf virus (CLRDV), in Mississippi. The presence of CotVA complicates the interpretation of CLRDV symptoms, and additional studies are needed to determine if CotVA is associated with symptomatology. Phylogenetic analysis indicates that CotVA is a member of the genus Ruflodivirus. The researchers also detected endogenous CotVA elements (referred to as endogenous caulimovirids) in the genomes of seven Gossypium species, including widely planted commercial species such as G. hirsutum (Upland cotton) and G. barbadense (Pima cotton). Future studies on CotVA synergies with other cotton viruses and ecology (including vector transmission) are warranted to further assess the impact of this pathogen on cotton production systems.

Fontdevila Pareta et al. (2025) report virome analyses of a pear germplasm collection from Belgium, from which a novel velarivirus (family *Closteroviridae*), putatively named Pyrus virus A, was detected in multiple asymptomatic trees. This discovery was enabled by double-stranded RNA extraction, HTS (Illumina NextSeq, 150-bp paired-end reads), and reverse transcription PCR, and the authors were careful to account for uneven virus distribution in each tree, collecting four leaves (one from each cardinal direction) at two canopy heights, for eight analyzed sample locations per tree via a pooling strategy. Pear germplasm from Slovenia and pear trees from Switzerland were subsequently tested for this new velarivirus, and those two sample sets together with the Belgium germplasm collection were also tested for three known viruses: citrus virus A (CiVA) and apple rubbery wood viruses 1 and 2 (ARWV-1 and -2). These are the first reports of CiVA in Belgium, Slovenia, and Switzerland and the first report of ARWV-1 and -2 in Switzerland.

Schnabel et al. (2025) explored the viromes of cultivated blackberry (genus Rubus) and wild Rubus spp. in symptomatic and asymptomatic plants at eight sites in South Carolina. They used metatranscriptomics to recover 23 known and novel plant viruses, as well as putative viruses of fungi, from leaves. The authors report genomic sequences, features, and phylogenies for many of these viruses, expanding the known sequence diversity of Rubusassociated viruses. Although the novel viruses were not shown to cause disease, their role in the Rubus virome is unknown and worthy of further exploration. This work shows that cultivated and wild Rubus plants share many of the same viruses, suggesting the potential for viral reservoirs in wild plants as an underappreciated source of those in cultivated blackberry varieties.

Insect-Specific Viruses

To discover insect-specific viruses (ISV) related to agricultural pests that may have potential to be used as biological agents, Britt-Ugartemendia et al. (2025) used RNA sequencing methods to study the virome of natural populations of a diverse array of citrus insect pests in Florida, including the black citrus aphid, green citrus aphid, Asian citrus psyllid (ACP), citrus leafminer (CLM), brown soft scale, Florida wax scale, and sharp-nosed leafhopper. The ACP as the vector of the causal agent of citrus greening disease and citrus aphids as vectors of several citrus viruses are well-known economically important citrus pests, while some like CLM can be a potential threat. Several known and novel ISV-related sequences belonging to various groups of +ssRNA viruses were identified and validated in this research. This study also compared the virome of the laboratory and field populations of ACP, and as expected, a difference was observed in the two populations. Furthermore, some of the putative viral sequences identified in this research demonstrated conserved domains and motifs such as RdRp of plant viruses. This was also reported in previous studies, recalling the hypothesis that ISVs are probably the origin of vector-borne plant viruses (Roossinck 2003; Wolf et al. 2018). This work provides valuable knowledge for future studies' understanding of plant-virus-vector interactions. In a similar study published in *Phytobiomes Journal* in 2017, Feng et al. (2017) explored the virome of the soybean aphid (Aphis glycines), one of the most economically important pest insects of soybean, which is also the vector of several important soybean viruses. By sequencing the total RNAs of the partially purified virus particles from soybean aphid colonies originally collected in Iowa, Michigan, and Ohio and one dead aphid sample from China, Feng et al. (2017) identified several known and new viral sequences. This paper was the first manuscript on viruses published in *Phytobiomes* Journal.

Bacterial and Phage Communities in Agricultural Systems

The study "The Bacterial and Viral Communities Associated with Onion Bacterial Bulb Rot," conducted by Liakos et al. (2025), investigates the diversity and functional potential of bacterial communities associated with asymptomatic and symptomatic onion bulbs exhibiting bacterial rot. They showed that geographic location significantly impacted bacterial communities, although confounding factors such as cultivar, environmental conditions, and storage practices were also at play. Additionally, the research highlights bacterial survival mechanisms, such as resistance to copper bactericides, which could inform preventive strategies. Viral populations detected in symptomatic bulbs may serve as potential biocontrol agents. This work provides a comprehensive framework for future research into bacterial bulb rot progression and prevention.

The manuscript "Genetically Similar Xanthomonas arboricola pv. pruni Strains and Associated Phages Display Phenotypic and Genotypic Variation Across 35 Years" explores the genetic and phenotypic diversity of X. arboricola pv. pruni (Xap) strains and their associated bacteriophages over 35 years, providing insights into phage-bacteria coevolution in agricultural systems (D'Amico-Willman et al. 2025). Despite high genetic similarity among Xap strains isolated from different locations, they exhibit distinct lytic responses to six phages collected in the same region over four decades. The study reveals that bacterial and phage genomes show minor but significant variations structured by geography and time. The characterized phages belong to the *Kantovirinae* subfamily and may represent a new genus. These findings highlight the role of phages in shaping bacterial genetic diversity and virulence, with potential implications for phage-based disease management strategies. By linking genomic and phenotypic data, this work enhances our understanding of microbial interactions in the phytobiome and their impact on plant disease dynamics.

Impact of Other Trophic Levels on Plant Viruses

The phytovirome can also be impacted by the presence of other microorganisms interacting with the plant. The study from Gaši et al. (2025) investigated the impact of arbuscular mycorrhizal fungi (AMF) on three grapevine viruses: grapevine leafroll-associated virus 3 (GLRaV-3), grapevine rupestris stem-pitting associated virus, and grapevine Pinot Gris virus (GPGV). Through real-time PCR, a diverse range of effects was observed when quantifying the virus concentration. For example, the presence of AMF can differentially influence GLRaV-3 and GPGV accumulation in grapevines, although the effects depended on AMF inoculum type, plant tissue, and infection duration. Overall, this study emphasizes the interest in developing a holistic approach, integrating cellular microbes, for better understanding plant-virus interactions in crops.

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