Towards sustainable protection against insect-borne plant viral diseases: Phytohormones and beyond

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Abstract Due to the immobility of plants, around 75% of 1100 plant virus species are piercing-sucking insect transmitted. Host plant-mediated interactions between viruses and insects play vital roles in the population dynamics of vectors and the epidemiology of plant diseases. A successful viral pathogen has to evolve multiple strategies to manipulate host immune responses and also the ecological environment to facilitate effective transmission by insect vectors. Among these strategies, reprogramming the phytohormone signaling pathways is critical to the establishment of an effective virus transmission among plants and disease pandemic. Here, we review recent studies on the plant-virus inter-relationships with a focus on molecular and biochemical mechanisms that drive vector-borne viral diseases. Defense-related phytohormones such as Jasmonic acid (JA), Salicylic acid (SA) and Ethylene (ET) have been reported to be directly regulated by viral proteins. This knowledge is essential for the further design and/or development of effective and sustainable strategies to protect viral damages so as to increase crop yield and food security. Future efforts in this area should be focused on integrating and meta-analysis of big data generating from dynamics and multiple dimensional pathogen-vector-crop interactions under real agricultural conditions to achieve sustainable protection against plant diseases.

Keywords Phytohormone; Jasmonic acid; Virus-vector-plant tripartite interactions; Geminivirus
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1 Introduction

Viruses are small infectious agents that cause disease by residing and replicating within the living cells of other organisms and therefore they are highly dependent on their hosts. Due to the sessile nature of plants, viral transmission between plants depends largely on the behaviors of the vectors of viruses, which are mainly the sucking insects. In the past decades, there have been global emergences of insect-transmitted plant viruses such as geminiviruses and tospoviruses. Interestingly, the transmissions are driven by two supervectors, namely the whitefly, *Bemisia tabaci*, and the Western flower thrips, *Frankliniella occidentalis* [1]. It is estimated that around 800 virus species (75% of the known species) are insect-borne and among these 362 species are transmitted by these two supervectors [2,3]. High reproduction and expansion rate, extreme polyphagy, rapid development of insecticide-resistance and human activities, have been the main factors contributing to the emergence of these insects as global pests. These supervector-transmitted viruses by different mechanisms and mediate novel viral emergence through local evolution, host shifts,

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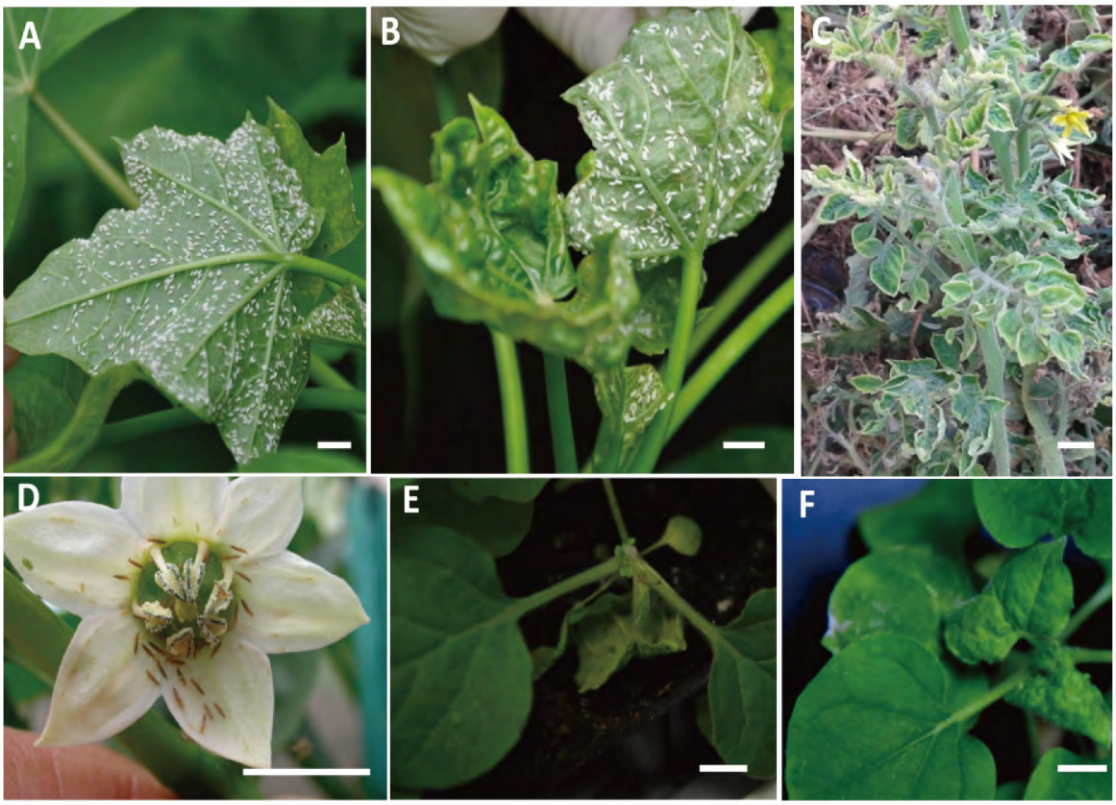

Figure 1 Supervectors-transmitted viruses causing plant diseases.
A, Whiteflies infest *Jatropha curcas* plants; B, whitefly transmitted begomovirus infection on *Jatropha curcas* plants; C, whitefly transmitted *Tomato yellow leaf curl virus* on tomato (*Solanum lycopersicum*); D, Western flower thrips infest flowers of pepper (*Capsicum annuum*); E, severe strain of TSWV infected *Nicotiana benthamiana*; F, weak strain of TSWV infected *Nicotiana benthamiana*.

mixed infections and global spread (Fig. 1 B, C, E, F) [4—6].

A growing number of studies have demonstrated the ability of these two types of vector-borne plant viruses to manipulate phenotypic traits of their insect hosts. Such manipulations include an increase in chemical communication between insects and viruses, which favor the pathogen transmission in a mutualistic manner (Fig. 2) [4, 7]. To understand the mechanisms of these processes is not only an exciting scientific exploration, but also an important strategy to crop losses due to viral diseases, such as in slowing down the epidemiology of plant diseases and in developing novel pesticides to control insect pests [3]. The most commonly reported changes induced by the vector-borne viruses in their hosts are alterations in host volatile compounds, which are primary chemical cues that attract the insect vectors. In this review, we will elaborate on some potentially interesting and important avenues for future research in this area.

Ideally, a perfect crop plant should possess the ability to resist pathogen challenge and insect infestation. In the long history of evolution, plants have being developed the ability of small RNA-based immunity against virus-infection, and then consequently viruses counter the host-RNA based immunity by encoding silencing suppressor [8—10]. Strategies that engineer small-RNA mediated resistance to plant viral diseases have been successfully developed [5, 11]. However, even for a simplified working model of a vector-borne viral disease development, plant immune responses to viruses or vectors are different, sometime even antagonistic [12]. Besides small RNA-based immunity against pathogen attack, defense-related phytohormones such as Jasmonic acid (JA), Salicylic acid (SA) and Ethylene (ET) play critical roles in nearly every aspect of the complicated virus-plant-insect tripartite interaction [12]. These factors all together may play complicated roles in determining the plant phenotype. The JA pathway activates a plant's defense response against an insect. While activation of SA signaling confers resistance against pathogens including

viruses, it also compromises the resistance to insects because of the antagonistic effects of JA and SA [13]. Additionally, the phenotypes of all three interacting organisms, virus, plant and insect, are highly dependent on specific combinations of their genotypes [14—16]. For example, the well-documented begomovirus-mediated positive effects on whiteflies are highly dependent on genetic background of the same plant species-tomato (*Solanum lycopersicum*). Even under a small variation among cultivars or genotypes of relative viruses, the positive effects may disappear [17, 18]. Despite our improved understanding of the molecular biology of plant viruses, the preventive measures to control viral diseases remain conventional, i. e. pesticide sprays for vector control [1, 3]. However, the overuse of pesticides in the field has been shown to wipe out beneficial non-target insects such as pollinators and predators of supervectors [19—22]. Together with the invasive characteristics of supervector species such as *B. tabaci* and *F. occidentalis*, the spread of vector-borne diseases has posed a serious threat to agriculture. Therefore, it is essential to unravel the complicated virus-plant-insect tripartite interactions (Fig. 2). This knowledge, particularly the roles of different phytohormones in plant disease development and vector transmission, will facilitate ways to effectively design plant control strategies that may reduce insect-borne plant diseases without compromising normal plant growth, since phytohormone accumulation and signaling have often been associated with alterations in plant development. Furthermore, the plant-virus-supervector interactions offer exciting opportunities for basic research and global implementation of generalized disease management strategies to reduce economic and environmental impacts.

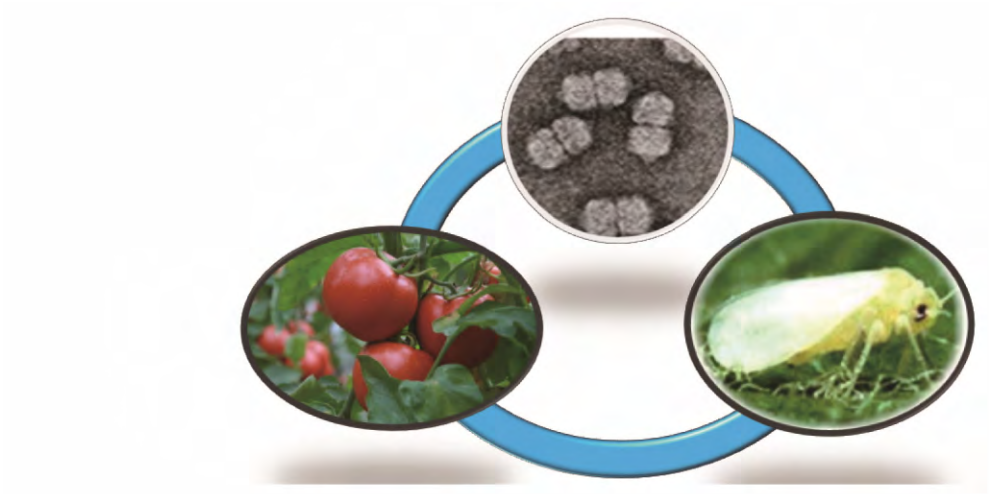

Figure 2 Plant virus transmitted by insects interferes with plant phytohormone pathways.

2 Molecular mechanisms of tripartite interactions demonstrated so far

2.1 Phytohormones are involved in the interplay between insects and pathogens

Phytohormones play a critical role in regulating nearly every aspect of a plant's biological process including development and pathogen defense [23]. Salicylic acid (SA), jasmonic acid (JA) and ethylene (Et) are mainly involved in the defense reaction [24]. Although gibberellins (GA), Auxin (Aux), cytokinins (CK), brassinosteroids (BR) and abscisic acid (ABA) also contribute to plant defense, their primary roles are mainly involved in a plant's physiological process and development [25—27]. During viral infection, a plant's normal physiological process is often disrupted by the alterations in phytohormone signaling and accumulation [12]. Recently, more direct evidence has demonstrated the effects of viral-modulated phytohormone on plant physiology and viral infection [12, 28]. These studies demonstrate how viruses directly manipulate plant hormones to reprogram the cellular environment and defense responses to enhance their own replication and transmission [12]. In this section, we focus on the viral-hijacked plant hormone systems and their effects on the viral transmission by vector insects.

Plant-to-plant transmission is an additional challenge for viruses because plants are immobile. Insect her-

bivores are key link to facilitate such transmission. The majority of these insects are hemipterans including aphids, leafhoppers and whiteflies. The evolution of these insects has become the primary conduit for long-distance movement of viruses as piercing-sucking mouthparts of the insects penetrate through phloem to acquire nutrition and also facilitate the transmission of viruses. Transmission of plant viruses by insect vectors requires a complex interaction between plants, viruses and vectors. Depending on the nature of the interaction, at least four mechanisms of insect transmission have been recognized: non-persistent (most aphid-transmitted viruses), semi-persistent (beetle- and some whitefly-transmitted viruses), persistent circulative (some aphid- and most leafhopper and whitefly-transmitted viruses) and persistent propagative (thrips- and some leafhopper- and aphid-transmitted viruses) [7, 29]. Recent reviews have addressed various aspects of insect-transmitted viruses including the mechanisms of transmission [1, 3, 29—32].

2.2 Geminivirus forms plant-dependent mutualism with whiteflies by inhibition of plant JA response

As stated above, the JA pathway confers resistance against herbivorous insects. A remarkable strategy adopted by such insects to evade this plant response is to disable JA-dependent defenses by employing obligate pathogens that suppress JA-dependent defenses [28]. Plant viruses are obligate pathogens that are often transmitted by insects and often exploit host cells to reproduce. During their interaction with plants, viruses employ effector molecules to manipulate host phytohormone signaling (Fig. 3) [33—36]. For instance, *Tomato yellow leaf curl China virus* (TYLCCNV) and *Cabbage Leaf Curl virus* (CalCuV) are whitefly (*Bemisia tabaci* Middle East-Asia Minor 1, MEAM1, commonly known as bio*type B*) - transmitted geminiviruses that cause diseases in tomato, tobacco and other crops [37, 38]. The TYLCCNV-satellite DNA complex can suppress terpenoid synthesis in plants to promote its mutualism with vectors [39].

When the invasive *B. tabaci* species is placed on TYLCCNV- or CalCuV-infected tobacco or *Arabidopsis* plants, growth rates of the insects increase when compared with those placed on un-infected plants [36, 40]. This is due to the TYLCCNV satellite βC1 protein and CalCuV DNA-B encoded BV1 protein which are required for viral pathogenicity. Zhang et al. found that this TYLCCNV-whitefly mutualism is achieved by repressing plant defenses through a viral pathogenencity factor, βC1 protein [35]. βC1-expressing *Arabidopsis* plants show attenuated expression of JA-responsive defense genes including *PDF1. 2*, *PR4* and reduced production of JA in response to wounding [39, 41]. JA inhibits *B. tabaci* MEAM1 survival [27] supporting the view that a whitefly employs TYLCCNV to protect itself from JA-dependent defenses. βC1 also has been identified to interact with AS1, which is known to regulate plant development. The *Arabidopsis* ASYMMETRIC LEAVES complex represses homeobox genes such as *KNAT2* to stably silence stem cell regulators in differentiating leaves [42]. AS1-AS2 complex uses epigenetic-dependent and auxin-dependent mechanisms to directly repress the abaxial determinants, MIR166A, YABBY5 and an auxin regulator AUXIN RESPONSE FACTOR3 (ARF3) [43]. Interestingly, virus-encoded βC1 mimics AS2 to compete with AS1 to disable the repression complex and to up-regulate several stem cell genes including homeobox genes, *KNAT1*, *2*, *6* and *ARF2*, *3*, *4*, whose expression levels correlate well with the severity of the phenotype [36]. Occasionally, βC1 mimics AS2 in the cytosol to inhibit plant RNA interference to allow better viral replication in an AS1-independent model ([10] and Ye et al. unpublished data). Furthermore, in two phenotypically distinct βC1-expressing plant lines, expression levels of the transcription factor, MYC2 that regulates the downstream gene, *Terpene synthase gene* (*TPS*), do not differ, suggesting that the functions of βC1 in plant development and JA defense are distinct (Fig. 3)[36]. Whitefly infestation activates the MYC2 transcription factor in non-virus infected plants. Dimerized MYC2 binds to the G-box/G-box-like elements in the *TPS* gene promoter region resulting in the elevation of *TPS* gene transcript levels. Thus, *TPS* directly and indirectly contributes to the resistance of whiteflies and begomoviral diseases. However, in geminivirus-infected plants, MYC2 protein activity is repressed by βC1 or BV1 due to heterodimerization resulting in reduced *TPS* transcript levels. Decreased biosynthesis and emission of terpenoid volatiles renders geminivirus-infected plants more susceptible to whiteflies [36]. Therefore, geminiviruses benefit from the virus-vector mutualism indirectly through increased probability of transmission to new hosts via whiteflies, especially the invasive types such as Middle East-Asia Minor 1

(MEAM1) and Mediterranean (MED) that may result in a global pandemic of whitefly-transmitted diseases.

Plant virus encoded pathogenicity factors are often multifunctional. For example, βC1 targets at least two transcription factors in the JA pathway. Similarly, majority of the genes regulated by AS1 and MYC2 are different. In this regard, the identification of additional host proteins that interact with βC1 in the phytohormone pathways can help to understand a plant's defense network against whiteflies at the organismal level. In conclusion, the mechanistic analysis of begomovirus-vector mutualism will help to advance virus epidemiology and may provide novel strategies for pest/viral disease management.

It is worthy to note that the mutualism between begomoviruses and *B. tabaci* through common plants is based on specific virus-vector-plant combination. The resulting interactions among these tripartite can be mutualistic, neutral or negative, and the number of cases exhibiting each of these three effects in these indirect interactions between begomoviruses and whiteflies appears evenly distributed as reported previously [17].

2.3 Other viral effectors manipulate host phytohormones to attract insects and promote disease transmission

As indicated above, behavioral alterations during virus-insect interactions are diverse signifying the need to understand the mechanism at work in other virus-insect systems.

The western flower thrips (*F. occidentalis*) are an important type of polyphagous pest insect that causes serious agricultural and economic losses. Apart from the mechanical damage caused by feeding, the western flower thrips are also vectors that transmit *Tomato spotted wilt virus* (TSWV) [44]. Recent reports have shown that thrips feeding induces JA-regulated plant defense, which negatively affects the performance and preference (i. e. host plant attractiveness) of thrips. The antagonistic interaction of JA-regulated plant defense and salicylic acid (SA)-regulated plant defense has been well studied [44]. It has been reported that TSWV infection allows thrips to feed heavily and multiply on *Arabidopsis* plants. TSWV infection elevates SA contents and induces SA-regulated gene expression in plants. On the other hand, TSWV infection decreases the expression level of JA-regulated genes induced by thrips feeding. Thrips prefer TSWV-infected plants to un-infected plants, which is similar to how the whitefly prefers geminivirus-infected plants. In JA receptor COI1-deficient mutants, however, thrips do not show a preference for TSWV-infected plants. In addition, SA application to wild-type plants increased their attractiveness to thrips, which may be explained by antagonistic effects of SA-JA plant defense systems [45]. However, no mechanistic research has been conducted to understand the genetic basis of TSWV-thrips mutualism.

Potyvirus is a genus consisting of 158 species including Turnip mosaic virus (TuMV) [46]. Nearly 30% of the currently known potyviruses are plant viruses. For instance, geminivirus, a member of this genus causes significant losses to agricultural, pastoral, horticultural, and ornamental crops. More than 200 species of aphids are known to spread potyviruses mainly in a non-persistent manner. Among these, most belong to the subfamily, *Aphidinae*. TuMV infection has been found to suppress callose deposition, an important plant defense response to feeding by its aphid vector, the green peach aphid (*Myzus persicae*). It was also found to increase aphid fecundity compared with the un-infected control plants. Casteel et al. identified that production of Nuclear Inclusion a-Protease (NIa-Pro) by TuMV largely regulates the changes in host plant physiology and increases green peach aphid reproduction by manipulating the ET signaling pathway.

Cucumber mosaic virus (CMV) is another important plant pathogenic virus in the family, *Bromoviridae* of the genus, *Cucumovirus* [47]. This virus has a worldwide distribution and a wide host range. In fact, it is known to have the widest host range of any documented plant virus. It can be transmitted from plant to plant both mechanically through sap transfer and by aphids in a stylet-borne fashion. The 2b RNA silencing suppressor in CMV is another viral effector that interferes with the JA signaling pathway, presumably by acting as a decoy to promote its own transmission by the aphid [48]. *Arabidopsis* plants expressing 2b showed reduced expression of JA responsive genes after CMV infection, leading to a hypothesis that 2b-mediated suppression of JA dependent defenses which is beneficial to the aphid. Indeed, aphid survival increased on tobacco infected with CMV, whereas it reduced on tobacco plants infected with a CMV strain

lacking the 2b protein [13, 34]. Another type of plant pathogen-phytoplasma, which is an obligate bacterial pathogen, also interferes with JA-dependent defenses [49, 50]. Secreted AY-WB protein 11 (SAP11) is an effector produced by the Aster Yellows Witches' Broom (AY-WB) phytoplasma, and interferes with JA biosynthesis in *Arabidopsis* by binding and destabilizing class II CIN-TCP (CINCINNATA-RELATED-TEOSINTE BRANCHED1, CYCLOIDEA, PCF) transcription factors, which act as positive regulators of the LOX2 (LIPOXYGENASE2) gene involved in JA biosynthesis [51] (Table 1 and Fig. 3). Transgenic *Arabidopsis* plants expressing SAP11 as well as wild-type plants infected with AY-WB show down-regulated LOX1 expression and reduced JA levels when wounded. Consequently, phytoplasma-transmitting the leafhopper pest, *Macrosteles quadrililineatus*, produces more progeny on AY-WB – infected, SAP11-expressing or lox2-silenced plants (Fig. 3)[52]. These examples suggest the independent evolution of complex tritrophic interactions, in which insects exploit microbes to disarm plant defenses while microbes (e. g. , viruses) gain increased dispersal to host plants, which may be impossible in the absence of an insect [53].

Our recent work also identified new viral proteins that are involved in the tritrophic interactions. We reported the role of ET and JA in the whitefly transmission of geminiviruses, indicating that virus-induced changes in ET and JA responses may mediate vector-plant interactions more broadly and thus represent a conserved mechanism for increased transmission by insect vectors across generations.

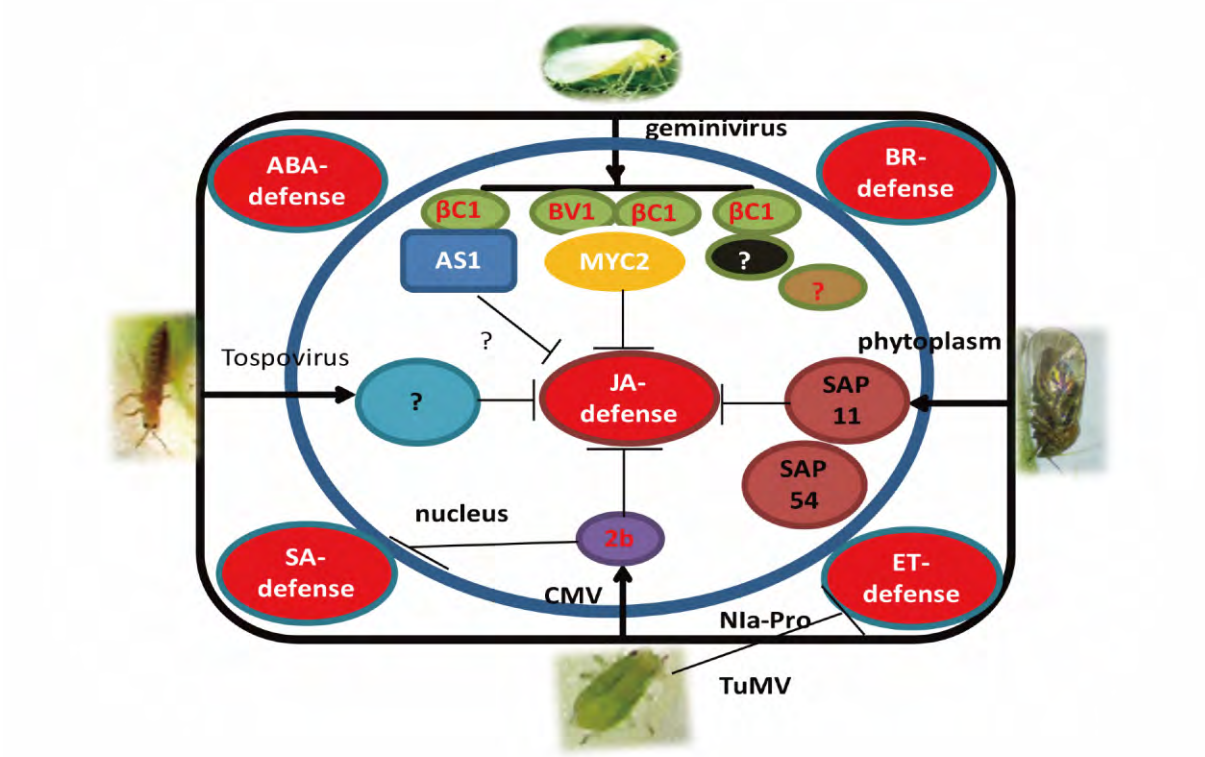

Figure 3 Plant virus transmitted by insects interferes with plant phytohormone pathways. Diverse phytopathogens have evolved common strategies to disable host jasmonate-dependent defenses that confer resistance to insect pests in complex tritrophic (pathogen-insect vector-plant) interactions. See text for additional information.

2.4 Host metabolic changes affect insect behavior

Comparative analysis of 'omic' datasets across different plant pathosystems has revealed that photosynthesis and related pathways, such as glycolysis and respiration, are main targets of pathogen infection. Terpenoids confer effective resistance to insects and some terpene biosynthesis genes have been reported to be involved in this process [54—59]. 7-epizingiberene synthase, a key enzyme in the biosynthesis of sesquiterpene 7-epizingiberene in wild tomato, is a major factor in conferring resistance to wild tomatoes against whiteflies [55, 60]. *TPS*-silenced *Arabidopsis* or *N. tabacum* plants were more susceptible to

whiteflies [36, 56]. Terpenoids may be constitutively emitted by plants such as tomato, and their emission levels are elevated after whitefly infestation [54, 57]. However, emission of terpenes by *Arabidopsis* requires induction by chemicals such as JA or biotic stress [61]. In *Arabidopsis* plants exposed to whitefly feeding, transcription of both *AtTPS10* and *AtMYC2* is up-regulated suggesting that *Arabidopsis* MYC2-TPS10 pathway is part of an induced defense system against these insects [36]. Furthermore, tomato SlMYC1, the homolog of AtMYC2, can transiently trans-activate some tomato *TPS* promoters in *N. benthamiana* leaves [59]. We hypothesize that MYC2-TPS is a conserved element of plant resistance against whitefly infestation. Hong et al. [62] showed that MYC2 positively regulates genes expressing sesquiterpene synthase (*AtTPS11* and *AtTPS21*) in the inflorescence, although they were not implicated in the sesquiterpenes in plant-insect interaction. The discovery that three *TPSs* are directly regulated by MYC2 suggests that both *TPS* and *MYC2* are promising targets to improve plant resistance to whiteflies. CMV affects host volatiles to decoy aphids for effective viral transmission.

Table 1 Vector-borne parasites manipulate phenotypic traits of their vectors and hosts in ways that increase contacts between them, and hence favor the parasites' transmission.

Pathogen			Host	Vector	Effector
Virus	*Tomato yellow leaf curl virus* (TYLCV)	Geminiviridae	Tomato	*Bemisia tabaci* (Hemiptera)	?
	Cabbage leaf curl virus (CaLCuV)	Geminiviridae	Arabidopsis	*Bemisia tabaci* (Hemiptera)	BV1
	Tomato yellow leaf curl China virus (TYLCCNV)	Geminiviridae	Tobacco	*Bemisia tabaci* (Hemiptera)	βC1
	Turnip mosaic virus (TuMV)	Potyviridae	Arabidopsis	*Myzus persicae* (*Hemiptera*)	NIa-Pro
	Potato leafroll virus (PLRV)	Luteoviridae	Potato	*Myzus persicae* (*Hemiptera*)	?
	Barley yellow dwarf virus (BYDV)	Luteoviridae	Barley	*Rhopalosiphum padi* (*Hemiptera*)	?
	Cucumber mosaic virus (CMV)	Bromoviridae	Gourd	*Myzus persicae* (*Hemiptera*)	2b
	Tomato spotted wilt virus (TSWV)	Bunyaviridae	Tomato	*Frankliniella occidentalis* (Thysanoptera)	?
	Southern rice black-streaked dwarf virus (SRBSDV)	Reoviridae	Rice	*Sogatella furcifera* (Hemiptera)	?
Other pathogens	*Erwinia tracheiphila*	*Enterobacteriaceae*	Gourd	*Acalymma vittatum* (Coleop-tera Coleptera)	?
	Candidatus Liberibacter	Rhizobiacea	Citrus	*Diaphorina citri* (Hemiptera)	?
	Candidatus (Ca.) Phytoplasma	*Candidatus* Liberibacter	Multiple plants	*Macrosteles quadrilineatus* (Hemiptera)	SAP11, SAP54
	Plasmodium falciparum	Plasmodiidae	Mouse and human	*Anopheles*(Diptera)	?

The cell wall-lacking bacterium, *Candidatus Phytoplasma mali* can alter both the odor of its host plant and the behavior of its vector, the univoltine psyllid, *Cacopsylla picta* (Table 1, Fig. 3) [63, 64]. Apple trees infected by this phytoplasma emit higher amounts of beta-caryophyllene when compared with un-infected trees. Psyllids with no previous contact with *Ca. P. mali*, as well as infected pyllids, are more attracted to volatiles emitted from phytoplasma-infected apple plants than from un-infected plants. Psyllids that developed on infected plants showed the opposite behavior. These results suggest that the pathogen modifies host plant odor to lure its vector to infected plants. This may result in higher numbers of trans-

mitting vector insects within the population.

2.5 Other mechanisms in tripartite interactions

It has been shown that infection by tomato spotted wilt virus (TSWV), the only plant-infecting member of the Bunyaviridae, alters the feeding behavior of its thrips vector, *F. occidentalis* [65]. Male thrips infected with TSWV fed more than un-infected males, resulting the frequency of all feeding behaviors to be increased by up to 3-fold, and thus increasing the probability of virus inoculation [66, 67]. Importantly, infected males made almost three times more non-ingestion probes (probes in which they salivate, but leave cells largely undamaged) compared with the un-infected males. A living cell is required for TSWV to establish an infection and for cell-to-cell movement; thus, this behavior is more likely to benefit establishing a viral infection. Some animal-infecting members of the Bunyaviridae genus (*La Crosse virus and Rift Valley fever virus*) are also reported to increase the biting rates of infected vectors. Concomitantly, these data support the hypothesis that the capacity of a virus in modifying the feeding behavior of its vector is a conserved trait among the members of the Bunyaviridae to enhance viral transmission [68].

Phytoplasmas produce a novel effector protein, SAP54 that interacts with members of the MADS-domain transcription factor family including SEPALLATA3 and APETALA1, which play a major role in the regulation of floral development [69, 70]. SAP54 mediates the degradation of MADS family of proteins by interacting with proteins of the RADIATION SENSITIVE23 (RAD23) family, which are eukaryotic proteins that shuttle substrates to the proteasome. *Arabidopsis rad23* mutants do not convert flowers into leaf-like tissues in the presence of SAP54 and during phytoplasma infection, emphasizing the importance of RAD23 in the activity of SAP54. Remarkably, plants with SAP54-induced leaf-like flowers are more attractive to phytoplasma leafhopper vectors and this colonization preference is dependent on RAD23. A phytoplasma effector promotes insect herbivore colonization and also produces more vegetative tissues for better nutrition to vector insects. Thus, SAP54 generates a short circuit between two key pathways of the host to alter development, resulting in sterile plants, and promotes attractiveness of these plants to leafhopper vectors and helps the obligate phytoplasma to reproduce and propagate [69]. The model that affects plant growth may be also important for other animal or plant vector-borne diseases.

3 Structure mimicry—a possible general strategy for vector-borne pathogens to promote their transmission

It is well known that pathogens and parasites manipulate phenotypic traits of their insect vectors and hosts in ways that increase contacts between them, and hence favor pathogen transmission. However, the detailed molecular mechanisms regulating each phytosystem are less understood. Particular efforts including comparative proteomics, transcriptomics, and metabolomics have been undertaken to tentatively unravel the molecular mechanisms underlying vector hijacking by pathogens. It is suggested that only a few baculoviral genes facilitate viral infection by manipulating insect behaviors. Our own and other recent studies on various virus-insect-plant tripartite interactions indicate that pathogens could deploy structural mimicry in proteins to alter plant physiology and environments to promote their transmission.

MYC2 is a master regulator and has several structural domains [71]. We found that βC1 binds to the HLH domain, which is essential for the dimerization of the MYC2 protein. In general, MYC2 forms homodimers with itself or heterodimers with other bHLH (e.g. MYC3 and MYC4) and MYB transcription factors to bind *cis*-regulating elements of target promoters [71]. *In vitro* competitive pull-down assays showed that βC1 interferes with MYC2 dimerization [36]. The two α-helix structures of βC1 contribute to the heterodimerization of MYC2. Animal viruses use molecular mimicry to reprogram their host signaling pathways, particularly the Notch and Wnt pathways, to enhance viral gene expression and down-regulate host defensive gene expression [72]. Li *et al*. [36] showed that begomoviruses use a structure-mimicking strategy to manipulate the MYC2-mediated defensive JA signaling pathway to benefit their insect vector. It has been reported that the phytoplasma effector, SAP11 protein, interacts with and destabilizes *Arabidopsis* TCP transcription factors to impair plant defense against its vector. The HLH domain of the TCP pro-

tein was predicted to be the putative binding site of SAP11 [52, 53]. The phytoplasma effector, SAP54 protein, may fold into a structure that is similar to the K-domain, a protein-protein interaction domain of MADS-domain proteins, which in part, regulates flowering and flower organ development. It has been suggested that SAP54 was evolved to mimic K-domain by undergoing convergent structural and sequence evolution. Given the highly specific developmental alterations, phytoplasmas could be used to study flower development in genetically intractable plants. A recent research on polerovirus (*Potato leafroll virus*, PLRV) with high resolution mass spectrometry identified several interacting partners in the vector transmission with PLRV-coat protein being the key protein. Structure modeling for these protein partners identified a common structural mimicry as the hallmark of host-pathogen interactions [73].

Therefore, we speculate that the formation of structural mimicry may be one way for a pathogen to enhance vector transmission. However, additional molecular and biochemical evidence based on other phytosystems such as aphid transmission-CMV and potyviral diseases is needed.

4 Conclusion and future perspectives

Research on vector-pathogen interactions has numerous avenues because of the emergence and recurrence of infectious diseases that not only infect crops but also humans and animals. Because traditional plant protection approaches do not always provide suitable solutions for an effective control (e. g. , too expensive for the countries concerned), fundamental investigations of the ecology and the evolution of vector-pathogen interactions remain a key aspect of the research in the virus-plant-insect tripartite interaction area. Additionally, attention should be directed to investigations on real agricultural field settings, as behavior in the laboratory may not precisely reflect field conditions. Behavioral manipulation of vectors is very complex so that a single method may not be sufficient to describe or understand them. For this reason, future research should benefit from the expertise from different disciplines [30]. Responses to the questions of regulators will need the integration of concepts and techniques from epidemiology, behavioral and evolutionary ecology, insect molecular biology, neurobiology and physiology [3].

Beneficial microbes in the microbiome of plant roots improve plant health. Induced systemic resistance (ISR) has emerged as an important mechanism by which selected plant growth-promoting bacteria and fungi in the rhizosphere, prime the whole plant body for enhanced defense against a broad range of pathogens and insect herbivores. A wide variety of root-associated mutualists, including *Pseudomonas*, *Bacillus*, *Trichoderma* and mycorrhiza species sensitize the plant immune system for enhanced defense without directly activating costly defenses. The ISR are reported to be related to phytohormones in some cases and reviewed elsewhere [74, 75]. This will also be a good candidate to promote plant immune system against both insects and viruses pathogens simultaneously and therefore beneficial microbes could be the second genome of plants to be manipulated in sustainable protection against biotic stresses.

Despite the difficulty in performing such multidisciplinary approaches, these efforts will undoubtedly provide a much better basis to understand the evolution of parasitic manipulation in vectors but also how to apply this knowledge into sustainable Integrated Plant Protection to achieve global food security [76,77]. Although speculative in appearance, each of the scenarios mentioned above is legitimate from the sustainable and ecological viewpoints. Because of this and the likelihood that the above hypotheses would considerably change based on the method of control and the transmission model for the most harmful pathogens affecting crops, they should be verified at the population level.

As expected, exponential growth of big data is accumulated in such vast quantities that they defy conventional analysis techniques. These data include gene expression, plant dynamics volatile components, insect neurological responses and behaviors, plant and environmental properties in a field, such as soil nutrition condition, soil microbiota, and so on. Similarly, detailed insect community composition can allow for the precise targeting of controls in a field. New ways of thinking of plant pathology and plant protection are essential [76,77]. New algorithms and computational decision making system will also be essential for sustainable plant health management technology.

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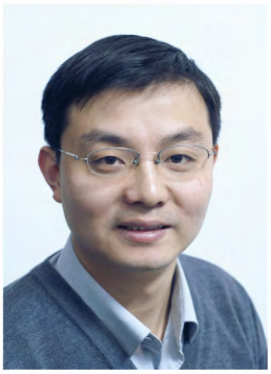

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