Review

Special Issue: Specificity of plant–enemy interactions

Unifying concepts and mechanisms in the specificity of plant–enemy interactions

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Host ranges are commonly quantified to classify herbivores and plant pathogens as either generalists or specialists. Here, we summarize patterns and mechanisms in the interactions of plants with these enemies along different axes of specificity. We highlight the many dimensions within which plant enemies can specify and consider the underlying ecological, evolutionary and molecular mechanisms. Host resistance traits and enemy effectors emerge as central players determining host utilization and thus host range. Finally, we review approaches to studying the causes and consequences of variation in the specificity of plant–enemy interactions. Knowledge of the molecular mechanisms that determine host range is required to understand host shifts, and evolutionary transitions among specialist and generalist strategies, and to predict potential host ranges of pathogens and herbivores.

The importance of specificity in plant–enemy interactions

Herbivores and plant pathogens make use of only a subset of the plant species and organs to which they are exposed. Such specialization is ubiquitous in plant–enemy interactions (see Glossary) and can have important consequences for their ecological and evolutionary dynamics. In a broad sense, specialization to the many different niches represented by plant communities has facilitated the evolution of the enormous diversity of herbivorous animals and microbial pathogens [1,2]. In turn, the specialization of plant enemies can influence rates of encounter with hosts [3] and with competitors or members of the third trophic level [4–6], and the local coexistence of plant species [3,7,8]. Specialization is also important from a broad array of applied perspectives. In particular, questions concerning the potential host range of plant enemies become crucial in a world in which both plants and their enemies have highly increased mobility, mainly because of human activities [9].

Despite the general importance of specificity in plant–enemy interactions, clearly defining the term 'specialization' is surprisingly challenging, and understanding the causes and consequences of specialization on ecological or evolutionary timescales remains an even more difficult task. In part, this is because specialization can evolve along multiple axes, often simultaneously [10]. Specialization is usually considered as the process of adaptation to a limited spectrum of potential resources, although evidence is accumulating that the adaptations required by generalists might be as complex as those required by specialists [3]. Research into the molecular mechanisms of host utilization by plant enemies and the ecology and evolution of specificity has progressed mostly independently. Likewise, plant–herbivore and plant–pathogen interactions have only rarely been subject to general synthesis [11]. This is despite many commonalities: herbivores and pathogens often exploit the same plant species or plant organs, must overcome the same defence mechanisms, have similar effects on plant fitness and share clear demographic similarities.

In this review, we identify concepts and mechanisms of general importance to the evolution of specificity in interactions between plants and their enemies. We first

Glossary

Effector: a molecule secreted by a plant enemy to manipulate host resistance. Effectors are commonly polymorphic among strains of the same species of pathogen or herbivore.
Effector-triggered immunity (ETI): a plant resistance response that is activated upon recognition of enemy effectors by NB-LRRs.
Enemy (plant enemy): used here to denominate herbivores and plant pathogens; that is, animals and microorganisms that form the second trophic level.
Host range (potential): the host species or organs that could be used by an enemy in the absence of all other (usually geographical, behavioural or temporal) barriers.
Host range (realized): the current host range of a plant enemy.
Microbe-associated molecular pattern (MAMP): synonym to PAMP.
Nucleotide-binding and leucine-rich repeat protein (NB-LRR): plant resistance proteins that act as receptors for effector molecules. NB-LRRs are often polymorphic among races or populations of plants.
Pathogen-associated molecular pattern (PAMP): phylogenetically conserved molecular motifs, such as chitin and flagellin, that are recognized by plants as indicators of attacking pathogens.
Pattern recognition receptor (PRR): proteins serving the perception of PAMPs, usually conserved within a host species or larger taxonomic group.
Specificity (geographic): differences in host ranges among populations of an enemy.
Specificity (ontogenetic): specificity in host use among different developmental stages of the enemy.
Specificity (phylogenetic): specificity concerning the phylogenetic distances among host species.
Specificity (structural): specificity concerning different structures or organs of the host.

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highlight the many axes along which specificity can evolve and discuss the fitness benefits of different strategies. We then review the common ecological, evolutionary and molecular mechanisms that determine patterns of host specificity. Only a mechanistic understanding of the determinants of host range will identify the reasons behind host shifts, and transitions between specialist and generalist strategies, and enable researchers to predict the potential host ranges of geographically isolated enemies. Effectors emerge as a common molecular concept that determines the host spectra of pathogens and herbivores [12–15]. Specialist pathogens and many specialist herbivores have highly specific effectors that facilitate the exploitation of specific hosts, but which in turn are often recognized as 'avirulence' (avr) genes by resistant hosts [14,16]. By contrast, generalist enemies commonly have multiple or promiscuous effectors or digestive enzymes that successfully suppress or overcome resistance responses in many different hosts [14,17–22].

Thus, host resistance traits and enemy strategies to overcome these traits are central players in defining host range. Based on these observations, we question the general hypothesis that specialists are more adapted than generalists and suggest that generalists are better understood as 'multi-host specialists'. We finish with concrete suggestions as to how next-generation sequencing techniques can be used to investigate natural host ranges of herbivores and plant pathogens and to understand the molecular mechanisms that explain why certain plant enemies utilize specific organs of specific hosts.

The multifaceted nature of specificity

The specificity of the interactions between plants and their enemies can range from tightly coupled associations among species pairs, through to diffuse relationships among diverse communities of prospective partners (for recent reviews, see [3]). However, the number of host species that an enemy can exploit (Figure 1) is only one aspect of its specificity. Specificity can manifest in different ways, and simple similarities in the overall number of host species attacked can mask fundamental differences in the biology of the organisms involved (Box 1).

Potential and realized host ranges

The host species utilized by a plant enemy in nature (the realized host range) does not necessarily reflect the species that it could attack in principle (its potential host range). A modern history of repeated invasions by plant enemies attests to the importance of geographical barriers in limiting realized host ranges [9]. As a consequence of geographical and behavioral limitations, plant enemies in nature seldom utilize all potential hosts. For example, the recent arrival of a single genotype of the endemic Brazilian rust pathogen *Puccinia psidii* in Australia (to which a wide range of species in the family Myrtaceae are potential hosts) has added more than 100 species to the realized host range of this pathogen (http://www.outbreak.gov.au/pests_diseases/pests_diseases_plant/myrtle-rust/national_host_list.html). However, despite being a growing problem worldwide, researchers currently lack the ability to predict potential host spectra accurately. Pathogens in particular

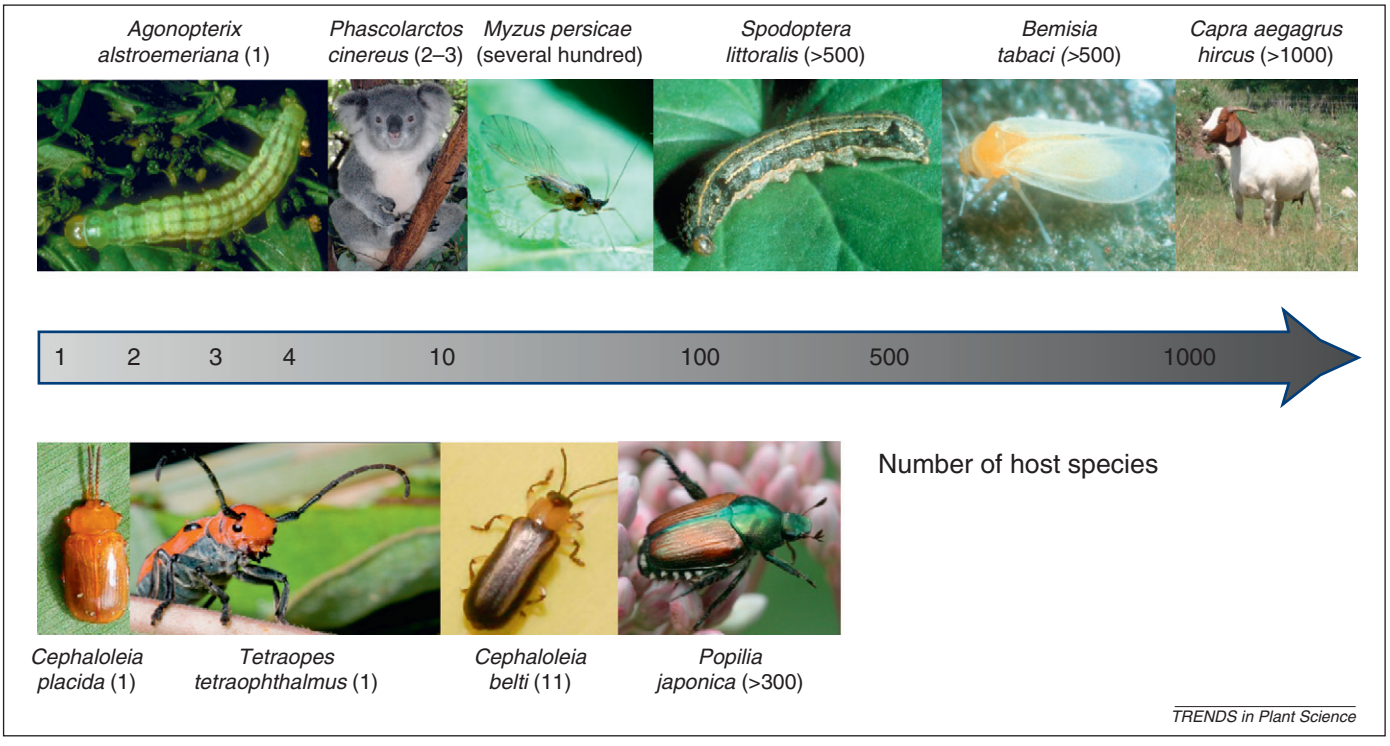

Figure 1. Host species number as the conventional concept of specialization. The number of host species (in parentheses after species names) that can be utilized by a plant enemy rank from a single one to over 1000. Neither feeding mode nor size or taxonomic position appear to be good predictors of the position of a plant enemy on this first axis of specialization. Images reproduced, with permission, from: Eric M. Coombs (*Agonopterix alstroemeriana*); W. Billen (*Bemisia tabaci*); Biologische Bundesanstalt für Land- und Forstwirtschaft (*Spodoptera littoralis*); S. Bauer (*Myzus persicae*); and David Cappaert (*Popilia Japonica*) (all published at http://www.bugwood.org under Creative Commons Attribution 3.0 License); Edith Freitag and Matthias Goeke (*Capra aegagrus hircus;* http://www.tiere-der-heimat.de); Marc E. Ellis at H2O pictures (http://www.h2opictures.com) (*Phascolarctos cinereus*); [62] and Carlos García-Robledo (*Cephaloleia* beetles); and Anurag A. Agrawal (*Tetraopes tetraophthalmus*).

are being increasingly discovered in association with novel host species, and often cause either unfamiliar or no obvious symptoms [23]. Humanopathogenic bacteria, such as *Salmonella*, *Escherichia coli* or *Klebsiella pneumoniae*, can also develop in plants [17,24,25], and endophytic fungi that were isolated from surface-sterilized, symptom-free leaves of diverse hosts commonly comprise multiple strains of the plant-pathogenic genera *Alternaria* [26–28], *Colletotrichum* [26,28,29] and *Fusarium* [26,28,30]. Are such endophytes non-pathogenic relatives of common pathogens, or are they pathogenic only under certain conditions, and what are the consequences of these alternative life stages for disease establishment and spread? In the 'Perspectives' section, we discuss how next-generation sequencing can be applied to investigate realized host ranges of herbivores and pathogens and how research into the molecular mechanisms used by plant enemies to overcome the resistance of their hosts will help to understand and reliably predict potential host ranges.

Axes of specificity

As stated by Daniel H. Janzen [31], plant enemies do not simply 'eat latin binomials'. Rather, they are adapted to exploit selected parts of selected organs of selected plants, and the evolutionary relationships among host species commonly affect the probability that a given plant species can be attacked by a particular enemy species. This poses the question 'to what is the specialist specialized?'

Specialization may manifest along various axes (Box 1). First, it can vary throughout the development of the enemy or host. Larval and adult stages of many insect herbivores [32,33] and different pathogenic spore stages [34] often have only partially overlapping or even completely separate host ranges ('ontogenetic specificity'), and most plant enemies can utilize only defined host developmental stages or organs ('structural specificity'). If the traits that make a host suitable for a particular enemy tend to be distributed among phylogenetically related hosts, then the 'phylogenetic specificity' of the enemy is high. In fact, the capacity of most herbivores and pathogens to exploit multiple hosts decreases with the phylogenetic distance among host species [14,35–39]. By contrast, few species are 'true generalists' that are capable of exploiting numerous completely unrelated host taxa. Examples include *Phytophthora cinnamomi*, which attacks more than 1000 plant species in numerous families, including Myrtaceae, Coniferales and Fagaceae [40]; the Japanese beetle, *Popillia japonica*, whose adults feed on the foliage, fruits and flowers of over 300 species of plants from at least 79 plant families (http://pubs.ext.vt.edu/2909/2909-1411/2909_1411_pdf.pdf), and classic 'model' generalists, such as whitefly (*Bemisia tabaci*) and *Spodoptera littoralis*. Finally, host ranges might differ according to specific environments or habitats ('geographic specificity') [41]. Importantly, specialization can vary almost independently along all the axes that we describe here. Thus, simply counting susceptible species limits one's ability to understand the processes that drive the evolution of specialization in plant–enemy interactions.

Generalist species as conglomerates of specialized genotypes

The observation that different populations of plant enemies can attack different host species indicates the potential importance of within-species genetic structure to an understanding of the evolution of host range [10,41]. In

Box 1. How can one define a specialist? The conceptual part of the problem

We identify an urgent need for obtaining standardized (and, hence, comparable) methods for the quantification of host ranges along different axes of specialization. Ideally, one would quantify how well every developmental stage of the enemy does on any of its potential host organs, or species. Useful data in this context would be growth rates, population densities of pathogens, time required to conclude certain developmental stages or, ultimately, fitness. As suggested in [36], specificity then can be quantified using classic diversity indices (e.g. Simpson and Shannon-Weaver indices) along at least four axes as ontogenetic, structural, phylogenetic and geographic specificity (Figure I). Their quantitative comparison is important when considering ecological and evolutionary consequences of specialization and will provide the basis for the understanding of underlying mechanisms.

For the enemy, ontogenetic specificity denominates specificity at different developmental stages (Figure Ia) such as, for example, beetle larvae and adults that perform differently on the same plant species [32]. Structural specificity means the degree of specialization on a specific host organ, or developmental stage of the host (Figure Ib). For example, *Peronospora* downy mildews, which are solely restricted to infecting flowers [76], have a higher structural specificity than does the Japanese beetle, *Popillia japonica*, whose adults feed on foliage, fruits and flowers (http://pubs.ext.vt.edu/2909/2909-1411/2909_1411_pdf.pdf). Phylogenetic specificity (Figure Ic) expresses the general tendency of herbivores and plant pathogens to restrict their host range to related species and can be high even for enemies that utilize multiple host species. Examples are *Puccinia psidii*, which attacks a large number of species that all belong to the family Myrtaceae [112] and the beetle *Cephaloleia belti*, which attacks 15 host species but only from the Zingiberales [62]. By contrast, the above-mentioned *Peronospora* downy mildews have been isolated from diverse plants belonging to the Orobanchaceae, Lamiaceae, Asteraceae and Dipsacaceae [76] and so showed higher structural than phylogenetic specialization. Finally, geographic specificity quantifies the differences among host uses of geographically disparate populations of a plant enemy (Figure Id) [41] and will be particularly strong for apparent generalist species that in fact represent groups of locally adapted cryptic specialists.

Importantly, the consequences of these different levels of specificity for the realized and potential host range and geographic range of a species are very different, as can easily be illustrated in terms of set theory (Figure Ie). The host range of an individual comprises the host ranges of all of its ontogenetic stages, meaning that its niche represents the intersection of the niches of its life stages (Figure Iei). At least one host for every ontogenetic stage must be present at the same site and in the correct temporal order to allow an individual to express positive fitness. As stated in [32], 'the breadth of environments in which a species can succeed is ultimately determined by the full pattern of its vital rates in each environment'. By contrast, the host range of a species is the sum of the host ranges of all of its geographic populations or genetic races (i.e. Figure Ieii). These aspects are important for invasion biology, for example, where an enemy that utilizes different hosts during its different developmental stages can invade only a region in which all hosts are present. By contrast, all individual populations or genetic lines of an enemy represent the source of potentially invasive founder individuals; therefore, species with very different host ranges in their different habitats, or genetic lines, have a higher level of releasing invasive progeny.

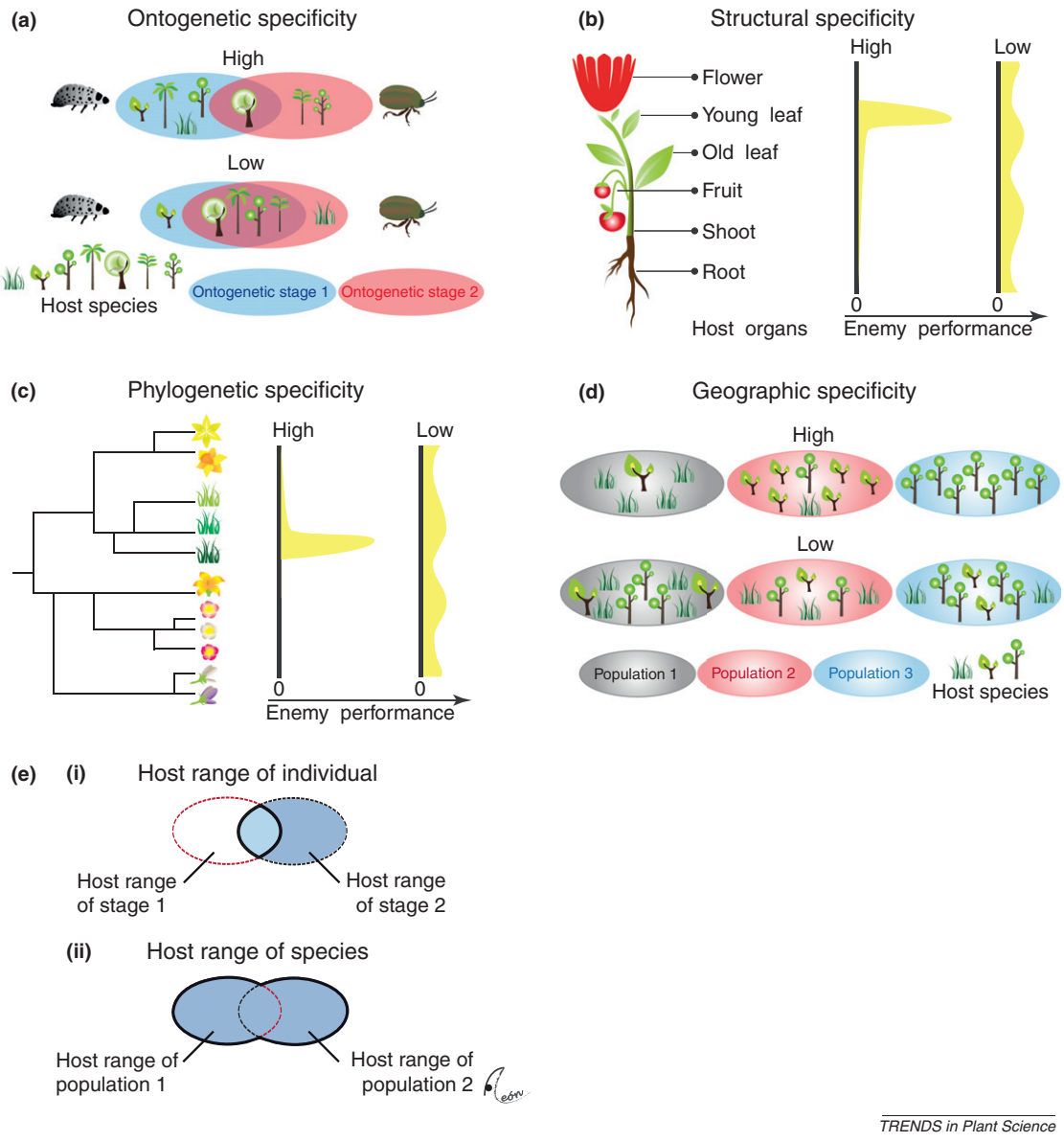

Figure I. Levels of specialization and their consequence for host ranges. Specialization can occur as **(a)** ontogenetic specificity, which describes specificity at different enemy developmental stages, **(b)** structural specificity towards certain host organs or developmental stages of the host, **(c)** phylogenetic specificity, and **(d)** geographic specificity, which describes different degrees of specialization among populations of the same species. Set theory **(e)** illustrates that the host range of an individual is formed by the intersection of the host ranges of all of its ontogenetic stages, whereas the overall host range of a species is represented by the union of the host ranges of all its populations.

particular, enemies recorded on a wide number of hosts (e.g. *P. cinnamomi* or *P. japonica*) should not be classified *a priori* as 'mega-generalists', because such species may exist as complexes of genetically discrete, host-specialized lineages. For example, a recent study reported a strong assortment of specific genotypes of the fungus *Beauveria bassiana* with specific hosts and other environmental conditions [42], and the generalist herbivore *B. tabaci* is also likely to represent a species complex [43]. In fact, many species with apparently wide host ranges exist as (often cryptic) complexes of closely related subspecies, host-associated lineages, locally adapted populations and individual specialists, all of which individually utilize a narrower host spectrum than does the species as a whole [41,44–47]. Such species can be termed 'generalists' only when considered as an entire, taxonomically defined unit [41] and might be better described and understood as a complex of functionally disparate, but closely related, specialists [41,48]. This intraspecific variability in host plant utilization is likely to be critical to understanding the emergence of true specialists, because specialization, similar to all adaptive processes, requires genetic variation within populations upon which evolution can act (see the section below on biotic heterogeneity and the emergence of specialists and the 'jack of all trades – master of none' principle).

Given such complexity, it is clear that categorically assigning enemies to generalist or specialist strategies is problematic. Rather, these terms need to be considered as end points along various continua of specificity. However, we argue that it remains important to differentiate among strategies, at least in a relative sense, because there are numerous ecological and evolutionary consequences of

specialist versus generalist life-histories. Although the terms are undoubtedly relative, evolution and ecology also act at relative scales (e.g. the same absolute number of offspring means a higher fitness in one environment, and lower fitness in a second environment). Therefore, distinguishing among strategies enhances one's ability to understand important processes and interpret patterns of specialization.

Ecological and evolutionary patterns

The ecological and evolutionary mechanisms that determine variation in host range mirror more general processes that drive the evolution of ecological specialization and the maintenance of biological diversity [10]. Here, we review the most widely cited hypotheses dealing with the phenomenon of specialization and how recent phylogenetic studies have challenged their universal applicability.

Adaptive radiation, ecological fitting and enemy-free space

The concept that is perhaps most widely cited to explain how the interactions of plants with their enemies enhance specialization is adaptive radiation, *sensu* Ehrlich and Raven [49]. Under this model, plants evolve new resistance traits in response to selective pressure exerted by herbivores; following herbivores then evolve specific counter-adaptations that enable them to overcome the new resistance, and so on. The same 'zigzag' model of coevolution of plant resistance with enemy counter-adaptations is also likely to be an important driver of the multiple layers of inducible resistance traits that plants exhibit against pathogens [14,16] and so potentially represents a major driving force in the process of adaptive radiation and ecological specialization. Thus, a common prediction is that coevolution will promote specialization via optimization of performance on a restricted subset of hosts. This outcome implies the existence of trade-offs between the capacity to attack a host and another component of fitness [50].

A necessary prerequisite of any host shift is that the plant enemy initially has the equipment to experience positive net fitness on the new host, before evolving specific adaptations that facilitate its utilization. The concept of ecological fitting was originally formulated by Daniel H. Janzen [51] to scrutinize the fact that observing a functioning plant–enemy interaction in nature does not necessarily indicate any coevolutionary history [52,53]. In fact, each of the thousands of invasive herbivores and plant pathogens represents independent empirical support of the importance of ecological fitting: all these species have initially experienced positive net fitness on new hosts that were probably never encountered before throughout their evolutionary history. At the ecological level, ecological fitting represents a 'black box' that is unlikely to be mechanistically understood, or let alone reliably predicted. However, recent studies have revealed molecular mechanisms that are likely to causally underlie ecological fitting and host shifts (see the section below on effectors and the determination of host specificity and host shifts).

Trade-offs

Low specificity increases the number of individual hosts available, probably enhances the geographic range that the enemy can occupy, reduces the time required to search for new hosts and lowers the risk of extinction should any one host be unavailable (Table 1). However, all plant enemies show some level of specialization and most are characterized by high phylogenetic conservatism [14,35–39]. The existence of trade-offs is perhaps the most frequently advanced hypothesis regarding the evolution towards specialization in plant–enemy interactions [6]. For example, adaptive radiation requires that a plant enemy that is well adapted to a specific new host will perform suboptimally on the ancestral species, and the concept of enemy-free space assumes different performances on the original versus the new host species [4,49].

In other words, genotypes that perform well on one host should perform relatively poorly on alternate hosts, and specialists should outperform generalists on any given host species (i.e. the 'jack of all trades – master of none' principle) [54]. Perhaps the clearest empirical support for these predictions comes from studies of microbial pathogens. For example, a trade-off has been demonstrated between host range and the mean number of infective spores produced by the pathogen *Melampsora lini*, such that strains infecting a wider range of hosts were generally less fecund [55]. Studies of plant viral pathogens [56,57] further provide evidence that trade-offs can be important for the maintenance of different specialist pathogen lineages, such that experimentally passaged viral populations that evolved increased capacity to exploit novel hosts suffered negative effects on the original hosts. Another study found that less virulent strains of *Pseudomonas syringae* had a higher probability of survival in non-host conditions than did more virulent strains [58]. For black bean aphids (*Aphis fabae*), studies comparing different clones reported trade-offs in lifetime fitness between two different host plants

Table 1. Advantages and disadvantages of specialist versus generalist strategies

	Specialist	Generalist
Advantages	Higher optimal performance	Diet mixing can improve development (herbivores)
	Reduced interspecific competition	Decreased resource heterogeneity
	Better able to respond to changes in host resistance	Increased capacity to establish in new environments and exploit novel hosts (niche expansion)
Disadvantages	Reliance on fewer hosts increases potential for heterogeneity in terms of resource availability	Lower optimal performance (jack of all trades – master of none)
	Increased intraspecific competition	Metabolically costly
	Reduced capacity to establish in new environments and exploit novel hosts (niche contraction)	Promiscuous enzymes (e.g. for detoxification) are less efficient
	Increased chance of evolutionary constraint?	

[59]. Similarly, fitness of pea aphid (*Acyrthosiphon pisum*) races was negatively correlated among different hosts and, consequently, it has been argued that antagonistic pleiotropy is likely to be generally important [60]. However, strains of the bacterial pathogen *Salmonella typhimurium* that were passaged though multiple generations in plant hosts did not alter their virulence for animals cells [24], suggesting that pathogens can evolve to exploit different hosts without measurable reductions in their performance.

Specialization originating via trade-offs requires that the fitness of genotypes within populations be negatively correlated among different hosts. However, trade-offs at the interspecific or interpopulation level might be more likely to represent the consequence of adaptation to specific hosts rather than its cause [6,54]. Thus, the generality of trade-offs as mechanisms that limit the emergence of generalist enemies is the subject of much ongoing debate [10]. Indeed, many studies seeking empirical support for trade-offs within populations report results that conflict with expectations. For example, one study searched for performance trade-offs in a population of the moth *Rothschildia lebeau*, whose larvae feed on several host species [61], whereas another study compared the performance of various *Cephaloeia* leaf beetles on native and invasive Zingiberaceae [54]. Both studies found mainly positive rather than negative genetic correlations in cross-host performance, meaning that genotypes that performed particularly well on one host also performed better on the alternative host [54,61]. Surprisingly, this phenomenon applied to both generalist and specialist beetle species [54].

How can these contrasting results be explained? First, the above-cited studies might indicate that trade-offs are more relevant for enemies that are tightly associated with their host plant, such as pathogens and sap suckers. However, the available data are not sufficient to decide whether these contrasting results really indicate a general and biologically relevant difference among guilds of plant enemies. Second, it is possible that trade-offs might only be important under certain conditions and so are difficult to detect. For example, many plant–herbivore studies that find no support for trade-offs have commonly used comparisons among genotypes within populations [54]. Third, many studies have used generalists with an arguably narrow host spectrum. For example, the generalist beetle used in [62] feeds naturally on 15 host species from five families; however, all the species fall within the order Zingiberales. Fourth, because ecologically realistic studies depend on already established plant–enemy interactions, they are focused on enemies that exhibit ecological fitting towards the new host. Finally, detecting trade-offs requires picking the appropriate measure of performance, measured under appropriate ecological conditions; many experiments might fail to meet these criteria.

Further mechanisms independent of, or reinforcing, trade-offs

Although it might depend on the detailed experimental design whether the capacity to perform on one specific host can be demonstrated to be negatively correlated with the capacity to utilize another one, the empirical evidence is overwhelming: the 'jack of all trades – master of none' principle does not apply in all situations and a negative correlation among the performances of a given enemy genotype on different hosts is not sufficient as a sole explanation for the evolution of specificity. Indeed, trade-offs are by no means the only evolutionary mechanism proposed to influence variation in host range. In particular, demographic events and population-level processes, such as bottlenecks and assortative mating, might reinforce, and perhaps even drive, the evolution of specialization [10,63]. For example, populations of plant enemies that utilize different host plants have a reduced probability of encounter and mating even before any genetic isolation mechanisms can act, and this isolation can become total if hosts are geographically separated or exhibit non-overlapping phenologies [64]. Enemies that utilize geographically separated discrete host populations can secondarily specialize via genetic drift and assortative mating, evolve genetic differentiation and, consequently, divergent patterns of specificity towards these host populations [3,65]. The accumulation of deleterious mutations that degrade performance on alternate hosts might further reinforce the effects of drift and assortative mating to the point that specialization may evolve even in the absence of other selective processes [63].

Therefore, major patterns in the specialization of plant enemies can be explained by host–enemy coevolution. However, host shifts are common and can be favored if the new host represents an enemy-free space [4–6]. Insect herbivores might shift to nutritionally suboptimal host species when enemy encounters are less likely to occur on the new hosts. For example, caterpillars of the swallowtail butterfly (*Papilio machaon aliaska*) were found on new host plants that allowed for lower survival rates than the original hosts, more commonly in habitats with lower ant-mediated lethality [66]. Such observations indicate the importance of the third trophic level in host choice. Herbivores might also shift onto new hosts on which the encounter rates with competitors, rather than predators, are reduced [6]. These mechanisms also apply to microbial pathogens, whose performance often is impaired by plant endophytic fungi, which outcompete or directly attack the pathogens [23]. Thus, in principle, pathogens might also search for enemy-free space and specialize on new hosts when these contain lower competitor, parasite or predator loads.

Biotic heterogeneity and the emergence of specialists

As highlighted above, host range can be viewed as the result of a trade-off in the ability to exploit individual hosts optimally and the ability to utilize the maximum number of hosts encountered [10,50]. As soon as plant enemies exhibit at least some intraspecific variability of the underlying traits, selection can act upon the different genotypes and favor an evolution towards more specialized or more generalist species, depending on the detailed selective pressures. In the classic 'arms-race' model of adaptive variation, this selection should normally cause the evolution of a more generalist ancestor towards a group of closely related specialists.

However, heterogeneities in selective pressure experienced in complex plant communities might make it difficult

for generalist enemies to counteradapt to the emergence of new host resistance traits. Costs apply when enemies must search for suitable hosts or express specific effectors or digestive enzymes to invade or utilize changing host plants. Therefore, it might be expected that generalist strategies should evolve when suitable hosts are infrequent or ephemeral, whereas specialist strategies should be favored when susceptible hosts are abundant and predictable [67]. It has been argued that interactions between generalist pathogens and rare or ephemeral hosts should favor the host in any evolutionary arms race, because although common generalist enemies might be important to the population biology of rare hosts, the reverse is unlikely to be true [47].

In addition, the demography of enemy populations is likely to be tightly linked to variation in host community structure. In particular, enemies with a large population size have an increased probability of colonizing new hosts and areas, and of generating new mutations [68]. Thus, whereas extreme variability in host availability might favor generalist enemies, the emergence of such strategies may be more probable at intermediate levels of resource heterogeneity [69]. Although decreased population size is likely to be associated with a concomitant decrease in the supply of mutants from which capacity to attack new hosts might emerge, this should be countered by an increased selective advantage to those enemies capable of infecting novel hosts. Thus, the probability that a generalist enemy will evolve and then be maintained may be highest at some intermediate level of host community complexity.

Phylogenetic patterns

In summary, classic theories predict a general tendency to evolve towards a higher degree of specialization, but certain conditions might also favor a widening of host range. In fact, both scenarios have been reported in phylogenetic studies. Given the general propensity for host range conservatism (see above), many hypotheses advanced to explain the remarkable levels of specificity in interactions between plants and their enemies have been based on the assumption of tightly coupled, pairwise coevolution and subsequent co-speciation [70]. Furthermore, specialization is often predicted to be an evolutionary 'dead-end' because, due to the costly accumulation of host-specific adaptations, specialized enemies should have increasingly lower fitness on hosts to which they are not specialized (see [38] for a recent empirical example). In Box 2, we review evidence for such phylogenetic constraints on the evolution of different strategies. By contrast, phylogenetic studies using ancestral state mapping increasingly reveal evidence for host switching [71–77] and examples of generalists that have evolved from a specialist ancestor [78,79]. These observations make it increasingly apparent that life-history evolution in species interactions can be highly dynamic.

Molecular mechanisms determine specificity in plant–enemy interactions

As we have highlighted above, specialization along one or more axes is inherent to all plant enemies, and trade-offs are one of the key evolutionary mechanisms that are likely to underlie the maintenance of specialized strategies. A specialist that very efficiently utilizes one host is commonly less efficient on a second host, generalists are often less successful than specialists on highly defended plants, and the potential to encounter an enemy-free space favors shifts towards hosts that are less optimal as a food source than the original host.

However, why is it so difficult in a proximate sense to utilize multiple plant species, or host organs, and which traits promote ecological fitting? Rather than enemies being limited by primary metabolic demands, performance on a host is mainly determined by the interplay of host resistance and enemy counteradaptation. Thus, host resistance traits and the capacity of the enemies to deal with them emerge as central factors in the determination of host ranges. The use of any specific host assemblage (which can be narrow or broad) requires particular adaptations [80]. Due to their different size and mobility, pathogens and herbivores have different strategies to avoid being affected by host resistance traits. Here, we discuss how host resistance traits and the molecular basis of their suppression or avoidance by plant enemies are fundamentally involved in the determination of host spectra and, thus, the evolution of specialist versus generalist strategies.

Genetic and physiological trade-offs

In specialized interactions, constraints on the use of certain host plants might be evident as pleiotropic trade-offs in the performance on alternate hosts (i.e. genes that promote the ability to utilize one partner, impair the ability to utilize another) [10,81]. For example, in the interaction between the plant *Linum usitatissimum* and its fungal pathogen *M. lini*, several interacting host resistance (R) and pathogen effector gene loci, which provide alternate resistance and infectivity specificities, have been identified [82]. Importantly, allelic variants at the *AvrP123* effector locus that escape recognition from one R gene, usually confer recognition to a different R gene [82]. Thus, specialization via antagonistic pleiotropy is seemingly built into the system. Analogous trade-offs might also

Box 2. Phylogenetic constraints and the evolution of specialization

If specialization evolves due to trade-offs in performance, then optimized performance on one host should limit performance on others. Thus, host specialization has classically been thought to be an evolutionary outcome that strongly selects for further specialization [67,78]. If these predictions are correct, then phylogenetic reconstructions should reveal that host shifts are rare in specialists, that transitions from specialist to generalist strategies are uncommon, and that specialist lineages are phylogenetically derived. Despite the intuitive appeal of these predictions, it is becoming increasingly clear that transitions from specialist to generalist strategies are common, host shifts are frequent and generalist lineages are equally likely to be phylogenetically derived [14,70,113]. Several authors report generalists and specialists within the same phylogenetic line [14], and models assuming irreversible evolution of generalists to specialists are usually strongly rejected [78]. Genomic plasticity and rapid evolution of the mechanisms underlying specialization are emerging as key drivers of this evolutionary dynamism. Horizontal gene transfer can radically alter the genomes of microbial pathogens, and many host jumps involve horizontal transfer of large effector complements [14]. Furthermore, conserved effector loci often undergo strong diversifying selection and display unusually high sequence polymorphism, suggesting rapid evolution in genes underlying host specificity [106,114].

mediate broader patterns of resistance specificity, such as resistance to insect herbivores versus microbial pathogens [83,84], and biotrophic versus necrotrophic pathogens [85]. Interestingly, evidence is mounting that some enemies can directly exploit constraints imposed on hosts by such pleiotropic costs [86–88].

Genetic constraints can also limit the success of generalist strategies, via trade-offs between the capacity to utilize a wide range of hosts and optimal performance on any one host [81,88]. A mechanism that causally underlies the 'jack of all trades – master of none' principle would be the fact that specialist herbivores can utilize specialized enzymes for the detoxification of the ingested food whereas generalists require either multiple [20–22] or widely effective digestive enzymes [19,89]. Because promiscuous enzymes are less efficient than those that catalyze only one distinct chemical reaction [18], and the synthesis of multiple enzymes comes at a high metabolic cost, generalists are usually less efficient than specialists in utilizing any given host species.

Effectors are used to overcome host resistance

Pathogens and herbivores have evolved some common molecular mechanisms to evade or suppress host resistance. Perhaps most universal is the concept of the 'effector': a term used to denominate all molecules that are released from plant enemies for host manipulation [13,14]. The concept of effectors and their role in host invasion and activation of resistance is most advanced in the context of plant–pathogen interactions [14,16,82,90,91]. In general, plants have evolved the capacity to perceive two classes of molecule (elicitors) that indicate attack by a pathogen. Conserved microbial molecules, known as 'pathogen-associated molecular patterns' (PAMPs) or 'microbe-associated molecular patterns' (MAMPs), are perceived by host receptor proteins known as pattern recognition receptors (PRRs). PAMPS are typically common structural components of a class of pathogen, such as chitin and flagellin, and their recognition causes PAMP-triggered immunity (PTI). To overcome this problem, many successful pathogens have evolved the capacity to deliver effectors into host cells to suppress PTI and other defence responses. In turn, many hosts have acquired the capacity to recognize either the changes that are inflicted by the action of these elicitors ('modified-self recognition'), or to recognize directly and specifically the effectors via their interaction with a class of plant receptor proteins that contain nucleotide-binding (NB) domains and leucine-rich repeat (LRR) receptor kinases. The recognition of effectors by plant NB-LRR proteins results in further layers of more specific ('gene-for-gene') resistance responses denominated 'effector-triggered immunity' (ETI) [16,82].

Plant NB-LRR proteins confer resistance to both microbial pathogens and insects [82]. The emerging pattern is that R genes in general confer resistance to herbivores in a similar manner to that described above for pathogens [12,13,15], although it is likely that the relative importance of effectors for host utilization is higher for insect herbivores that are more intimately associated with their host plant, such as leaf miners, phloem feeders and single-cell feeders. By contrast, classic, leaf-chewing folivores are likely to depend more on behavioral, detoxification and sequestration strategies, although specific effectors, such as β-glucosidases and proteases, are likely to play a crucial role in this process.

As for pathogens, plants can recognize herbivore-derived effectors either directly, or by monitoring their action. In the first case, the recognition of 'herbivore-associated patterns' (HAMPs) induces relatively general resistance responses, which often depend, at least partly, on induction of a jasmonic acid (JA) pathway. Plants also recognize the action of herbivores, similar to the above-mentioned 'modified-self recognition' strategy, by perceiving fragmented or delocalized molecules associated with damage caused by their action. The responses that are elicited by plant 'damaged-self recognition' are very general ones and are commonly based on JA induction [92,93]. By contrast, sucking insects cause little mechanical damage. However, they are intimately associated with the plant cells and need multiple effectors to evade recognition of their HAMPs [13]. As such, herbivores such as the hessian fly (*Mayetiola destructor*) and aphids were the first for which resistance mediated by R genes was reported [13,15,94,95]. The targets of these R genes are likely to be insect-derived effectors. For example, based on their similarity to pathogen effectors, 48 effector candidates have been identified in the green peach aphid (*Myzus persicae*) [96]. At least one of these functions as the target of recognition in certain plant hosts [96]. Correspondingly, several NB-LRR proteins have been identified that are required for a successful resistance induction against insect herbivores, including whitefly and aphids ([15,97] and references therein).

Effectors and the determination of host specificity and host shifts

As essential microbial components, PAMPS are highly conserved within and among pathogen species and plant PRRs are also highly conserved [98]. By contrast, the overall effector repertoire of pathogens can be highly variable, particularly among species and host-specific lineages [14,90] and, when conserved, often displays unusually high levels of sequence polymorphism [3,75]. Similarly, NB-LRR resistance protein repertoires are variable among species and can be highly polymorphic [14] or deleted entirely within host species [99]. As we note above, there are well-established patterns of evolutionary conservatism in the range of hosts used by any given pathogen. It has been hypothesized that the interplay among highly conserved PRR-triggered immunity, and highly specific NB-LRR protein-triggered immunity can explain the phylogenetic specialization of plant enemies [14]. In particular, as phylogenetic distances among hosts increase, the effector repertoire carried by a pathogen becomes increasingly ineffective, first at suppressing specific NB-LRR-mediated resistance, followed by increasing basal PRR-mediated resistance. Together with trade-offs and ongoing antagonistic coevolution, such dynamics could strongly promote evolution towards increasing specialization in plant–enemy interactions.

Despite the general appeal of the above scenario, some enemies do have truly wide host ranges, and host shifts can involve quite distantly related hosts [70,76]. What mechanisms facilitate the ecological fitting that underlies these

seeming anomalies? One strategy that may be important to the maintenance of wide host ranges is to suppress host resistance mechanisms at an early stage of induction. In pathogens, the type III secretion system is used to inject multiple effectors into host cells and helps *Salmonella* to colonize plant and animal hosts [17,24]. This system represents a common trait of numerous plant and animal pathogens, many of which are characterized by wide host ranges [100–104]. Because it can be also used by specialists to inject specific cocktails of coevolved effectors [105], the type III secretion system does not represent a 'generalist strategy' per se, but represents instead an apparatus that might facilitate a true 'generalists' strategy. For example, many *Pseudomonas* strains inject coronatine, a JA mimic. Coronatine manipulates the crosstalk between the JA and salicylic acid (SA) pathways [84], resulting in the suppression of SA-dependent responses. Thus, this process renders hosts generally susceptible to this pathogen [11]. Similarly, many insects and necrotrophic pathogens release hormones that suppress JA-dependent defence responses [11,84]. Such strategies mean that enemies can avoid the consequences of the expression of hundreds of defence-related genes, thereby greatly enhancing their ability to utilize a wide range of hosts. Other generalists may rely on carrying a broad spectrum of effectors, only a subset of which might be effective against any given host (e.g. *Botrytis cinerea*), and generalist herbivores often use multiple, or highly promiscuous, enzymes to detoxify their food [18–22], although such strategies presumably come at a cost [106]. Thus, host shifts and ecological fitting likely involve mechanisms that suppress resistance strategies that are shared between the old and new host. Moreover, shifts within and among closely related species may be achieved by mutations or deletion of single effector genes [106], whereas more distant jumps often seem to involve horizontal transfer of large complements of effectors [14].

Perspectives: new approaches to studying specificity in plant–enemy interactions

In the above section, we reviewed the most common molecular mechanisms that underlie the specificity in host use by herbivores and plant pathogens and discussed how recently developed molecular concepts can help to explain classic ecological and evolutionary hypotheses, such as adaptive radiation, phylogenetic conservatism and ecological fitting. However, molecular tools remain underutilized in the ecological and evolutionary disciplines and more could be done to identify the molecular determinants of specificity of host use by plant enemies.

As we highlight in Box 1, the various axes along which plant enemies evolve specificity are important because they provide insight into the underlying ecological and evolutionary mechanisms. However, there is a real deficiency of empirical data on host range under natural conditions. Such data will be required to inform theory and to develop capacity to predict host shifts and potential for invasion of plant enemies. For pathogens, one way to develop a better understanding of host utilization under field conditions might be intensive sampling and unbiased sequencing of microbial DNA resident in plants. This approach has been applied recently to discover asymptomatic endophytes, many of which are pathogens in crops [27–30]. However, to determine precise outcomes, such studies will need to be accompanied by experiments examining the effects of colonization under common environmental conditions. For herbivores, realized host ranges can be assessed by unbiased collection strategies (fogging, etc.) and food web construction, preferably accompanied by feeding trials (e.g. [107]). An alternative way to determine realized host ranges of herbivores will be DNA barcoding or another sequencing-based approach to determine species ranges of ingested food items in the digestive tracts of animals living in the wild.

The continued development of next-generation sequencing platforms will revolutionize research into the functional and evolutionary genetics of specialization in plant–enemy interactions. As well as the identification of realized host ranges of herbivores and pathogens, DNA barcoding and other sequencing-based strategies can be used to identify cryptic species and patterns in the association of certain genotypes of plant enemies with specific hosts [42,44]. Recently, these techniques have successfully been applied to understand the specificity and virulence of the over 50 pathovars of the 'generalist' pathogen, *P. syringae* [104,108].

Large-scale phylogenies are increasingly becoming available and can be subjected to ancestral trait mapping to identify host shifts and truly 'phylogenetically conservative' plant enemies [71–77]. Enemies from different populations, or species that have recently diverged and specialized onto different hosts [109], can be compared at the genomic, transcriptomic and phenotypic level, to investigate directly the genetic changes that are involved in host specialization. Perhaps the most powerful tool is represented by phylogenetically controlled comparisons among transcriptomes of specialists and generalists or in enemies that have recently been subject to a major shift in their host range. In particular, pathogens that have evolved higher specialization following a host shift [73,108], pathogens that have changed their life style from pathogen to asymptomatic endophyte [110] or vice versa [111], and related herbivores that represent the same feeding guild but differ strongly in host range [22], are promising models to screen for adaptations that allow generalists and specialists to fulfill successfully all the specific tasks that are required for their respective strategy. As is the case for many other disciplines, research into host ranges of plant enemies urgently requires multidisciplinary approaches to gain a causal understanding of why a particular enemy can, or cannot, successfully attack certain hosts and to predict potential host shifts and changes among specialist and generalist strategies.

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References

1 Thompson, J.N. (2005) Coevolution: the geographic mosaic of coevolutionary arms races. *Curr. Biol.* 15, R992–R994

2 Dyer, L.A. *et al.* (2007) Host specificity of Lepidoptera in tropical and temperate forests. *Nature* 448, 696–699
3 Barrett, L.G. *et al.* (2009) Continua of specificity and virulence in plant host–pathogen interactions: causes and consequences. *New Phytol.* 183, 513–529
4 Berdegue, M. *et al.* (1996) Is it enemy-free space? The evidence for terrestrial insects and freshwater arthropods. *Ecol. Entomol.* 21, 203–217
5 Jeffries, M.J. and Lawton, J.H. (1984) Enemy free space and the structure of ecological communities. *Biol. J. Linnean Soc.* 23, 269–286
6 Futuyma, D.J. and Moreno, G. (1988) The evolution of ecological specialization. *Annu. Rev. Ecol. Syst.* 19, 207–233
7 Connell, J.H. (1978) Diversity in tropical rain forests and coral reefs. *Science* 199, 1302–1310
8 Janzen, D.H. (1970) Herbivores and the number of tree species in tropical forests. *Am. Nat.* 104, 501–528
9 Parker, I.M. and Gilbert, G.S. (2004) The evolutionary ecology of novel plant–pathogen interactions. *Annu. Rev. Ecol. Evol. Syst.* 35, 675–700
10 Poisot, T. *et al.* (2011) A conceptual framework for the evolution of ecological specialisation. *Ecol. Lett.* 14, 841–851
11 Pieterse, C.M.J. and Dicke, M. (2007) Plant interactions with microbes and insects: from molecular mechanisms to ecology. *Trends Plant Sci.* 12, 564–569
12 Erb, M. *et al.* (2012) Role of phytohormones in insect-specific plant reactions. *Trends Plant Sci.* 17, 250–259
13 Hogenhout, S. and Bos, J. (2011) Effector proteins that modulate plant–insect interactions. *Curr. Opin. Plant Biol.* 14, 422–428
14 Schulze-Lefert, P. and Panstruga, R. (2011) A molecular evolutionary concept connecting nonhost resistance, pathogen host range, and pathogen speciation. *Trends Plant Sci.* 16, 117–125
15 Walling, L.L. (2008) Avoiding effective defenses: strategies employed by phloem-feeding insects. *Plant Physiol.* 146, 859–866
16 Jones, J.D.G. and Dangl, J.L. (2006) The plant immune system. *Nature* 444, 323–329
17 Schikora, A. and Hirt, H. (2012) Plants as alternative hosts for Salmonella. *Trends Plant Sci.* 17, 245–249
18 Li, W. *et al.* (2003) Diversification of furanocoumarin-metabolizing cytochrome P450 monooxygenases in two papilionids: specificity and substrate encounter rate. *Proc. Natl. Acad. Sci. U.S.A.* 100, 14593–14598
19 Li, X. *et al.* (2004) Structural and functional divergence of insect CYP6B proteins: from specialist to generalist cytochrome P450. *Proc. Natl. Acad. Sci. U.S.A.* 101, 2939–2944
20 Pauchet, Y. *et al.* (2008) Mapping the larval midgut lumen proteome of *Helicoverpa armigera*, a generalist herbivorous insect. *J. Proteome Res.* 7, 1629–1639
21 Pauchet, Y. *et al.* (2010) Pyrosequencing the *Manduca sexta* larval midgut transcriptome: messages for digestion, detoxification and defence. *Insect Mol. Biol.* 19, 61–75
22 Ramsey, J.S. *et al.* (2010) Comparative analysis of detoxification enzymes in *Acyrthosiphon pisum* and *Myzus persicae*. *Insect Mol. Biol.* 19, 155–164
23 Partida-Martinez, L.P.P. and Heil, M. (2011) The microbe-free plant: fact or artefact? *Front. Plant Sci.* 2, 100
24 Schikora, A. *et al.* (2011) Conservation of *Salmonella* infection mechanisms in plants and animals. *PLoS ONE* 6, e24112
25 Winfield, M.D. and Groisman, E.A. (2003) Role of nonhost environments in the lifestyles of *Salmonella* and *Escherichia coli*. *Appl. Environ. Microbiol.* 69, 3687–3694
26 Fisher, P.J. *et al.* (1994) Fungal endophytes from the leaves and twigs of *Quercus ilex* L. from England, Majorca and Switzerland. *New Phytol.* 127, 133–137
27 Albrectsen, B.R. *et al.* (2010) Endophytic fungi in European aspen (*Populus tremula*) leaves: diversity, detection, and a suggested correlation with herbivory resistance. *Fungal Divers.* 41, 17–28
28 Ghimire, S.R. *et al.* (2011) Biodiversity of fungal endophyte communities inhabiting switchgrass (*Panicum virgatum* L.) growing in the native tallgrass prairie of northern Oklahoma. *Fungal Divers.* 47, 19–27
29 Frohlich, J. *et al.* (2000) Endophytic fungi associated with palms. *Mycol. Res.* 104, 1202–1212
30 Gazis, R. and Chaverri, P. (2010) Diversity of fungal endophytes in leaves and stems of wild rubber trees (*Hevea brasiliensis*) in Peru. *Fungal Ecol.* 3, 240–254
31 Janzen, D.H. (1979) New horizons in the biology of plant defences. In *Herbivores: Their interactions with Secondary Plant Metabolites* (Rosenthal, G.A. and Janzen, D.H., eds), pp. 331–350, Academic Press
32 García-Robledo, C. and Horvitz, C.C. (2011) Experimental demography and the vital rates of generalist and specialist insect herbivores on native and novel host plants. *J. Anim. Ecol.* 80, 976–989
33 Altermatt, F. and Pearse, I.S. (2011) Similarity and specialization of the larval versus adult diet of European butterflies and moths. *Am. Nat.* 178, 372–382
34 Jin, Y. (2010) Century-old mystery of *Puccinia striiformis* life history solved with the identification of *Berberis* as an alternate host. *Phytopathology* 100, 432–435
35 Novotny, V. and Basset, Y. (2005) Host specificity of insect herbivores in tropical forests. *Proc. R. Soc. B* 272, 1083–1090
36 Poulin, R. *et al.* (2011) Host specificity in phylogenetic and geographic space. *Trends Parasitol.* 27, 355–361
37 Gilbert, G.S. and Webb, C.O. (2007) Phylogenetic signal in plant pathogen–host range. *Proc. Natl. Acad. Sci. U.S.A.* 104, 4979–4983
38 Rasmann, S. and Agrawal, A. (2011) Evolution of specialization: a phylogenetic study of host range in the red milkweed beetle (*Tetraopes tetraophthalmus*). *Am. Nat.* 177, 728–737
39 Ness, J.H. *et al.* (2011) Phylogenetic distance can predict susceptibility to attack by natural enemies. *Oikos* 120, 1327–1334
40 Zentmyer, G.A. (1983) The world of *Phytophthora*. In *Phytophthora, its Biology, Taxonomy, Ecology and Pathology* (Erwin, D.C. *et al.*, eds), pp. 1–8, American Phytopathological Society
41 Fox, L.R. and Morrow, P.A. (1981) Specialization: species property or local phenomenon? *Science* 211, 887–893
42 Ormond, E.L. *et al.* (2010) A fungal pathogen in time and space: the population dynamics of *Beauveria bassiana* in a conifer forest. *FEMS Microbiol. Ecol.* 74, 146–154
43 Brown, J.K. *et al.* (1995) The sweetpotato or silverleaf whiteflies: biotypes of *Bemisia tabaci* or a species complex? *Annu. Rev. Entomol.* 40, 511–534
44 Hebert, P.D.N. *et al.* (2004) Ten species in one: DNA barcoding reveals cryptic species in the neotropical skipper butterfly *Astraptes fulgerator*. *Proc. Natl. Acad. Sci. U.S.A.* 101, 14812–14817
45 Garrido, E. *et al.* (2012) Local adaptation: simultaneously considering herbivores and their host plants. *New Phytol.* 193, 445–453
46 Loxdale, H.D. *et al.* (2011) The evolutionary improbability of 'generalism' in nature, with special reference to insects. *Biol. J. Linnean Soc.* 103, 1–18
47 Kniskern, J.M. *et al.* (2010) Maladaptation in wild populations of the generalist plant pathogen *Pseudomonas syringae*. *Evolution* 65, 818–830
48 Bolnick, D.I. *et al.* (2003) The ecology of individuals: incidence and implications of individual specialization. *Am. Nat.* 161, 1–8
49 Ehrlich, P.R. and Raven, P.H. (1964) Butterflies and plants: a study in coevolution. *Evolution* 18, 586–608
50 Thrall, P.H. *et al.* (2007) Coevolution of symbiotic mutualists and parasites in a community context. *Trends Ecol. Evol.* 22, 120–126
51 Janzen, D.H. (1985) On ecological fitting. *Oikos* 45, 308–310
52 Agosta, S.J. (2006) On ecological fitting, plant–insect associations, herbivore host shifts, and host plant selection. *Oikos* 114, 556–565
53 Agosta, S.J. *et al.* (2010) How specialists can be generalists: resolving the 'parasite paradox' and implications for emerging infectious disease. *Zoologia* 27, 151–162
54 García-Robledo, C. and Horvitz, C.C. (2012) Jack of all trades masters novel host plants: positive genetic correlations in specialist and generalist insect herbivores expanding their diets to novel hosts. *J. Evol. Biol.* 25, 38–53
55 Thrall, P.H. and Burdon, J.J. (2003) Evolution of virulence in a plant host–pathogen metapopulation. *Science* 299, 1735–1737
56 Agudelo-Romero, P. *et al.* (2008) The pleiotropic cost of host-specialization in *Tobacco etch potyvirus*. *Infect. Genet. Evol.* 8, 806–814
57 Wallis, C.M. *et al.* (2007) Adaptation of plum pox virus to a herbaceous host (*Pisum sativum*) following serial passages. *J. Gen. Virol.* 88, 2839–2845
58 Barrett, L.G. *et al.* (2011) Cheating, trade-offs and the evolution of aggressiveness in a natural pathogen population. *Ecol. Lett.* 14, 1149–1157
59 Mackenzie, A. (1996) Trade-off for host plant utilization in the black bean aphid, *Aphis fabae*. *Evolution* 50, 155–162

60 Hawthorne, D.J. and Vian, S. (2001) Genetic linkage of ecological specialization and reproductive isolation in pea aphids. *Nature* 412, 904–907
61 Agosta, S.J. and Klemens, J.A. (2009) Resource specialization in a phytophagous insect: no evidence for genetically based performance trade-offs across hosts in the field or laboratory. *J. Evol. Biol.* 22, 907–912
62 García-Robledo, C. *et al.* (2010) Larval morphology, development, and notes on the natural history of *Cephaloleia* 'rolled-leaf' beetles (Coleoptera: Chrysomelidae: Cassidinae). *Zootaxa* 2610, 50–68
63 Kawecki, D. (1994) Accumulation of deleterious mutations and the evolutionary cost of being a generalist. *Am. Nat.* 144, 833–838
64 Matsubayashi, K.W. *et al.* (2010) Ecological speciation in phytophagous insects. *Entomol. Exp. Appl.* 134, 1–27
65 Sicard, D. *et al.* (1997) Genetic diversity and pathogenic variation of *Colletotrichum lindemuthianum* in the three centers of diversity of its host, *Phaseolus vulgaris*. *Phytopathology* 87, 807–813
66 Murphy, S.M. (2004) Enemy-free space maintains swallowtail butterfly host shift. *Proc. Natl. Acad. Sci. U.S.A.* 101, 18048–18052
67 Jaenike, J. (1990) Host specialization in phytophagous insects. *Annu. Rev. Ecol. Syst.* 21, 243–273
68 Normark, B. and Johnson, N. (2011) Niche explosion. *Genetica* 139, 551–564
69 Benmayor, R. *et al.* (2009) Host mixing and disease emergence. *Curr. Biol.* 19, 764–767
70 Janz, N. (2011) Ehrlich and Raven revisited: mechanisms underlying codiversification of plants and enemies. *Annu. Rev. Ecol. Evol. Syst.* 42, 71–89
71 de Vienne, D.M. *et al.* (2009) Phylogenetic determinants of potential host shifts in fungal pathogens. *J. Evol. Biol.* 22, 2532–2541
72 Desprez-Loustau, M-L. *et al.* (2011) Interspecific and intraspecific diversity in oak powdery mildews in Europe: coevolution history and adaptation to their hosts. *Mycoscience* 52, 165–173
73 Raffaele, S. *et al.* (2010) Genome evolution following host jumps in the Irish potato famine pathogen lineage. *Science* 330, 1540–1543
74 Roy, B.A. (2001) Patterns of association between crucifers and their flower-mimic pathogens: host jumps are more common than coevolution or cospeciation. *Evolution* 55, 41–53
75 Van Der Merwe, M.M. *et al.* (2008) Coevolution with higher taxonomic host groups within the *Puccinia/Uromyces* rust lineage obscured by host jumps. *Mycol. Res.* 112, 1387–1408
76 Voglmayr, H. (2003) Phylogenetic relationships of *Peronospora* and related genera based on nuclear ribosomal ITS sequences. *Mycol. Res.* 107, 1132–1142
77 Woolhouse, M.E.J. *et al.* (2005) Emerging pathogens: the epidemiology and evolution of species jumps. *Trends Ecol. Evol.* 20, 238–244
78 Nosil, P. and Mooers, A.O. (2005) Testing hypotheses about ecological specialization using phylogenetic trees. *Evolution* 59, 2256–2263
79 Branca, A. *et al.* (2011) Intraspecific specialization of the generalist parasitoid *Cotesia sesamiae* revealed by polyDNAvirus polymorphism and associated with different *Wolbachia* infection. *Mol. Ecol.* 20, 959–971
80 Gomez, J.M. *et al.* (2010) Ecological interactions are evolutionarily conserved across the entire tree of life. *Nature* 465, 918–921
81 Forister, M.L. *et al.* (2012) Revisting the evolution of ecological specialization, with emphasis on insect–plant interactions. *Ecology* DOI: 10.1890/11-0650.1
82 Dodds, P.N. and Rathjen, J.P. (2010) Plant immunity: towards an integrated view of plant–pathogen interactions. *Nat. Rev. Genet.* 11, 539–548
83 Erb, M. *et al.* (2011) Synergies and trade-offs between insect and pathogen resistance in maize leaves and roots. *Plant Cell Environ.* 34, 1088–1103
84 Thaler, J.S. *et al.* (2012) Evolution of jasmonate and salicylate signal crosstalk. *Trends Plant Sci.* 17, 260–270
85 Kliebenstein, D.J. and Rowe, H.C. (2008) Ecological costs of biotrophic versus necrotrophic pathogen resistance, the hypersensitive response and signal transduction. *Plant Sci.* 174, 551–556
86 Lorang, J.M. *et al.* (2007) Plant disease susceptibility conferred by a 'resistance' gene. *Proc. Natl. Acad. Sci. U.S.A.* 104, 14861–14866
87 Faris, J.D. *et al.* (2010) A unique wheat disease resistance-like gene governs effector-triggered susceptibility to necrotrophic pathogens. *Proc. Natl. Acad. Sci. U.S.A.* 107, 13544–13549
88 Loiseau, C. *et al.* (2008) Antagonistic effects of a MHC class I allele on malaria-infected house sparrows. *Ecol. Lett.* 11, 258–265
89 Marsh, K.J. *et al.* (2006) The detoxification limitation hypothesis: where did it come from and where is it going? *J. Chem. Ecol.* 32, 1247–1266
90 Arnold, D.L. and Jackson, R.W. (2011) Bacterial genomes: evolution of pathogenicity. *Curr. Opin. Plant Biol.* 14, 385–391
91 Pieterse, C.M.J. *et al.* (2009) Networking by small-molecule hormones in plant immunity. *Nat. Chem. Biol.* 5, 308–316
92 Heil, M. *et al.* (2012) How plants sense wounds: damaged-self recognition is based on plant-derived elicitors and induces octadecanoid signaling. *PLoS ONE* 7, e30537
93 Heil, M. (2009) Damaged-self recognition in plant herbivore defence. *Trends Plant Sci.* 14, 356–363
94 Foster, J.E. *et al.* (1991) Effects of deploying single gene resistance in wheat for controlling damage by the Hessian fly (Diptera: Cecidomyiidae). *Environ. Entomol.* 20, 964–969
95 Hogenhout, S.A. *et al.* (2009) Emerging concepts in effector biology of plant-associated organisms. *Mol. Plant Microbe Interact.* 22, 115–122
96 Bos, J.I.B. *et al.* (2010) A functional genomics approach identifies candidate effectors from the aphid species *Myzus persicae* (green peach aphid). *PLoS Genet.* 6, e1001216
97 Dogimont, C. *et al.* (2010) Host plant resistance to aphids in cultivated crops: genetic and molecular bases, and interactions with aphid populations. *C. R. Biol.* 333, 566–573
98 Zipfel, C. (2009) Early molecular events in PAMP-triggered immunity. *Curr. Opin. Microbiol.* 12, 414–420
99 Stahl, E.A. *et al.* (1999) Dynamics of disease resistance polymorphism at the Rpm1 locus of *Arabidopsis*. *Nature* 400, 667–671
100 Coombes, B.K. (2009) Type III secretion systems in symbiotic adaptation of pathogenic and non-pathogenic bacteria. *Trends Microbiol.* 17, 89–94
101 Grant, S.R. *et al.* (2006) Subterfuge and manipulation: Type III effector proteins of phytopathogenic bacteria. *Annu. Rev. Microbiol.* 60, 425–449
102 Killiny, N. and Almeida, R.P.P. (2011) Gene regulation mediates host specificity of a bacterial pathogen. *Environ. Microbiol. Rep.* 3, 791–797
103 Marlovits, T.C. and Stebbins, C.E. (2010) Type III secretion systems shape up as they ship out. *Curr. Opin. Microbiol.* 13, 47–52
104 O'Brien, H.E. *et al.* (2011) Next-generation genomics of *Pseudomonas syringae*. *Curr. Opin. Microbiol.* 14, 24–30
105 Dean, P. (2011) Functional domains and motifs of bacterial type III effector proteins and their roles in infection. *FEMS Microbiol. Rev.* 35, 1100–1125
106 Barrett, L.G. *et al.* (2009) Diversity and evolution of effector loci in natural populations of the plant pathogen *Melampsora lini*. *Mol. Biol. Evol.* 26, 2499–2513
107 Novotny, V. *et al.* (2010) Guild-specific patterns of species richness and host specialization in plant-herbivore food webs from a tropical forest. *J. Anim. Ecol.* 79, 1193–1203
108 Baltrus, D.A. *et al.* (2011) Dynamic evolution of pathogenicity revealed by sequencing and comparative genomics of 19 *Pseudomonas syringae* isolates. *PLoS Pathog.* 7, e1002132
109 Stukenbrock, E.H. *et al.* (2010) Whole-genome and chromosome evolution associated with host adaptation and speciation of the wheat pathogen *Mycosphaerella graminicola*. *PLoS Genet.* 6, e1001189
110 Freeman, S. and Rodriguez, R.J. (1993) Genetic conversion of a fungal pathogen to a nonpathogenic, endophytic mutualist. *Science* 260, 75–78
111 Romao, A.S. *et al.* (2011) Enzymatic differences between the endophyte *Guignardia mangiferae* (Botryosphaeriaceae) and the citrus pathogen *G. citricarpa*. *Genet. Mol. Res.* 10, 243–252
112 Rayachhetry, M.B. *et al.* (2001) Host range of *Puccinia psidii*, a potential biological control agent of *Melaleuca quinquenervia* in Florida. *Biol. Control* 22, 38–45
113 Roy, B.A. and Kirchner, J.W. (2000) Evolutionary dynamics of pathogen resistance and tolerance. *Evolution* 54, 51–63
114 Chen, M-S. *et al.* (2010) Unusual conservation among genes encoding small secreted salivary gland proteins from a gall midge. *BMC Evol. Biol.* 10, 296