The evolution of coevolution in the study of species interactions

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The study of reciprocal adaptation in interacting species has been an active and inspiring area of evolutionary research for nearly 60 years. Perhaps owing to its great natural history and potential consequences spanning population divergence to species diversification, coevolution continues to capture the imagination of biologists. Here we trace developments following Ehrlich and Raven’s classic paper, with a particular focus on the modern influence of two studies by Dr. May Berenbaum in the 1980s. This series of classic work presented a compelling example exhibiting the macroevolutionary patterns predicted by Ehrlich and Raven and also formalized a microevolutionary approach to measuring selection, functional traits, and understanding reciprocal adaptation between plants and their herbivores. Following this breakthrough was a wave of research focusing on diversifying macroevolutionary patterns, mechanistic chemical ecology, and natural selection on populations within and across community types. Accordingly, we break down coevolutionary theory into specific hypotheses at different scales: reciprocal adaptation between populations within a community, differential coevolution among communities, lineage divergence, and phylogenetic patterns. We highlight progress as well as persistent gaps, especially the link between reciprocal adaptation and diversification.

KEY WORDS: Chemical ecology, evolutionary ecology, microevolution–macroevolution, plant–herbivore interactions, reciprocal natural selection.

The study of coevolution (here defined as reciprocal adaptation in interacting species) has a long and venerable history. Ehrlich and Raven (1964) popularized coevolution with their seminal study integrating chemical ecology, adaptive evolution, and macroevolutionary hypotheses based on detailed natural history of butterflies and flowering plants. The notion that ecological interactions could spur the generation of biodiversity at the grandest of scales was bold and inspiring. In the following decades, natural history continued to inform the increasingly flourishing field of coevolution. For example, Passiflora and Heliconius emerged as a model system where reciprocal adaptations were observed in the field (Benson et al. 1975, Turner 1981, Gilbert 1982). Heliconius larvae are specialized herbivores of Passiflora, and Passiflora evolved potent morphological and chemical defense such as cyanogenic glycosides against herbivory by Heliconius. In turn, some Heliconius spp. evolved the ability to tolerate or sequester high amount of these toxins (Nahrstedt & Davis 1981, Cavin & Bradley 1988).

As research methodologies advanced, coevolution began to diverge into distinct research traditions. Development of phylogenetic tools empowered researchers to more quantitatively study the macroevolutionary patterns laid out by Ehrlich and Raven (1964). Janzen’s (1980) commentary initiated a more population-level microevolutionary approach that focused on measuring the agents and strength of selection. At the beginning of this expansion of the field, two foundational contributions by Dr. May Berenbaum (1983, Berenbaum et al. 1986), considered both macro-coevolutionary patterns and experimentally testable microevolutionary processes. A set of hypotheses has subsequently emerged, spanning 1) key innovations of defenses and counter-defenses, whose evolution leads to elevated rates of diversification, 2) reciprocal selection, leading to matching phenotypes in communities of two interacting populations, and 3) geographically separated communities, among which interacting populations coevolve to different degrees (Figure 1, Table 1).
This perspective article celebrates the second bloom of research beginning in the 1980s and how it framed our current understanding of coevolution. In this light, although our focus is on plant-herbivore interactions, we note other systems where coevolutionary hypotheses are being tested at multiple scales (Table 1), often employing methods not readily amenable to plant-herbivore systems. Our overall goal is to highlight where substantial progress has been made and to suggest research that will bridge gaps in rapidly advancing areas of coevolutionary research.

**Macroevolutionary Origins**

**THE PRE-PHYLOGENETIC ERA**

Ehrlich and Raven’s coevolutionary hypothesis predicted a macroevolutionary pattern that is bold, yet challenging to test. It proposed, without specifying mechanisms, that a plant lineage that evolved a novel defense trait against its herbivores would escape into an enemy-free “adaptive zone” and subsequently diversify; herbivores were predicted to then evolve counter-defenses and diversify along existing plant lineages, thus creating sequential bursts of speciation in both plants and their insect herbivores. As some of the first pattern-driven evidence for macro-scale coevolution, Berenbaum (1983) outlined the relationship between plants in the parsley family (Apiaceae) and swallowtail butterflies (Papilionidae). She broke down the sequential steps laid out by Ehrlich and Raven and evaluated evidence for each. This study combined chemical and taxonomic knowledge of host use and proposed a scenario whereby plants sequentially evolved hydroxycoumarins, linear furanocoumarins and ultimately angular furanocoumarins to increasingly defend against herbivory; each step resulted in expansion of the toxic plant lineage and was met by counter-adaptation and diversification in a resistant lineage of butterflies.

Although there was a lack of some mechanistic details in the Apiaceae-Papilionidae system at the time of Berenbaum’s 1983 publication, later work filled these gaps. For example, in the proposed step where herbivores already feeding on plants with linear furanocoumarins evolved counter-defenses against novel angular furanocoumarins, little was known about the relevant biochemical mechanisms of detoxification, or whether it necessarily led to radiation in the insect lineage. Then Berenbaum et al. (1996) first identified and Krieger et al. (2018) later solidified cytochrome P450 monooxygenases as the key innovation that enabled
Table 1. Coevolutionary hypotheses, their predictions, and available empirical evidence stemming from Ehrlich and Raven (1964). We parsed evidence for each hypothesis into two categories: those for the predicted pattern (i.e., what is expected on a phylogeny or among populations) and those for the underlying processes (mechanisms generating the predicted pattern). The table is not exhaustive and does not consider predictions from the literature on host-parasite interactions or pollination biology, but rather aims to highlight classic systems where empirical tests of hypotheses have been particularly fruitful. When possible, process references match the study systems where corresponding patterns have been observed.

<table>
<thead>
<tr>
<th>Coevolutionary hypothesis</th>
<th>Predicted Pattern</th>
<th>Evidence for Pattern</th>
<th>Generating process</th>
<th>Evidence for process</th>
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<tr>
<td><strong>Plant-herbivore Systems</strong></td>
<td><strong>Non-Plant-herbivore Systems</strong></td>
<td><strong>Evidence for Pattern</strong></td>
<td><strong>Generating process</strong></td>
<td><strong>Evidence for process</strong></td>
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<tr>
<td>Through recombination or mutation, a lineage evolves novel defenses to its antagonists (Ehrlich and Raven 1964)</td>
<td>Host clades have enhanced and conserved defensive traits</td>
<td>Glucosinolates: Fahey et al. 2001</td>
<td>Tetrodotoxin: Brodie III and Brodie Jr. 2015</td>
<td>Genetic basis for the evolution of novel defense traits</td>
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<td>Alkaloids: Wink 2020</td>
<td>Acquired immunity in Bacteria (CRISPR-Cas9): Shmakov et al. 2017</td>
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<td>Released from its previous antagonists, a lineage with novel defense undergoes radiation in a new adaptive zone (Ehrlich and Raven 1964)</td>
<td>“Escape-and-radiate”; a key defense innovation escalates over time and is associated with enhanced diversification</td>
<td>Asclepias: Agrawal et al. 2009a, b</td>
<td>Plant and defensive mutualists: Weber and Agrawal 2014</td>
<td>Several hypotheses reviewed by Altoff et al. 2014; Marquis et al. 2016; and Maron et al. 2019</td>
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<td>Cytochrome P450s: Cohen et al. 1992; Li et al. 2004</td>
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<td>Nitrile-specifier proteins: Fischer et al. 2008</td>
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<td>The antagonist lineage evolves novel counter-defense traits (Ehrlich and Raven 1964)</td>
<td>Counter-defense traits evolve after host defenses and are conserved in clades that adapt to recently radiated hosts</td>
<td>Na⁺/K⁺-ATPase target site insensitivity to cardiac glycosides: Meyer et al. 2012</td>
<td>Genetic basis for the evolution of counter-defense traits</td>
<td>Na⁺/K⁺-ATPase target site insensitivity to cardiac glycosides: Karageorgi et al. 2019</td>
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<td>Bacteriophage tail fibers: Sousa et al. 2021</td>
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<td>Bacterial toxins: Schulte et al. 2010</td>
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<td>Snake sodium channels: Brodie III and Brodie Jr. 2015</td>
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<tr>
<td><strong>Antagonist lineage with novel counter-defense traits establishes on the previously radiated clade and diversifies (Ehrlich and Raven 1964)</strong></td>
<td>Major host shifts are associated with increased rates of diversification</td>
<td>Butterflies: Fordyce 2010, Edger et al 2015, Allo et al. 2021</td>
<td>Novel offense trait opens niche in new host species, increasing the chance of reproductive isolation</td>
<td>Crossbills and lodgepole pines: Benkman 1993, Smith and Benkman 2007</td>
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<td>Flies: Winkler and Mitter 2008</td>
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<td>Many studies associate host shifts with the initiation of speciation: Forbes et al. 2017</td>
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<td>Gall-forming herbivores and their parasitoids: Nyman et al. 2007, Nicholls et al. 2018</td>
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<td>Bacteria and bacteriophages: Braga et al. 2018</td>
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<td>Distantly related herbivores diversify onto the same group of plants (or plants with similar chemistry)</td>
<td>Several insect groups on Apocynaceae: Karageorgi et al. 2019</td>
<td>Genetic basis of convergent adaptations leading to host shifts</td>
<td>Na^+ /K^+-ATPase target site insensitivity to cardiac glycosides: Dobler et al. 2012</td>
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<td>Several insect groups on Apiaceae: Berenbaum 2001 Lepidopterans on Inga: Endara et al. 2017</td>
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<td>Chemical bridge leads to host shifts in a butterfly: Murphy and Feeny 2006</td>
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<td><strong>Distantly related lineages convergently evolve counter-defenses and colonize existing hosts in parallel (Berenbaum 1983)</strong></td>
<td>Distantly related herbivores diversify onto the same group of plants (or plants with similar chemistry)</td>
<td>Squamate reptiles and their bufonid toad prey: Ujvari et al. 2015</td>
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<td><strong>Geographically separated populations of two coevolving species experience varying strengths of reciprocal selection (Thompson 1994)</strong></td>
<td>Coevolving populations will exhibit varying degrees of matching phenotypes across different communities</td>
<td>Parsnip’s furanocoumarins and webworm’s detoxification: Zangerl and Berenbaum 2003 Weevils and fruit shape: Toju and Sota 2006a,b Flowers and pollinating flies: Anderson and Johnson 2008</td>
<td>Within a discrete community, populations show reciprocal adaptations (Janzen 1980); selection can be diffuse depending on community contexts (Rausher 1996)</td>
<td>Bacteria and bacteriophage: Buckling and Rainey 2002, Perry et al. 2015</td>
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<td>Snakes and newts: Brodie and Ridenhour 2003</td>
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**Table 1. (Continued).**
Papilio to feed on plants with angular furanocoumarins. The mechanism of action and differential expression patterns of different cytochrome P450 variants were also elucidated later (Calla et al. 2020), yielding insights into potentially varying selective pressures posed by different toxic compounds.

CONVERGENCE AND COEVOLUTION BETWEEN PLANTS AND MULTIPLE HERBIVORES

One lesser discussed aspect of coevolutionary theory expounded on by Berenbaum (1983, 2001) is the notion that independent clades of herbivorous insects may show parallel adaptations to the same plant defenses and thus engage in a form of multi-species coevolution. She initially debunked the notion that closely related herbivores should only feed on closely related plants – pointing to occasional shared chemistry among distantly related plants that can act as a bridge for host shifts. She then went on to use convergent defenses of plants and counter-defenses of insects to explain how multispecies coevolution might proceed. The pattern would manifest as the convergent evolution of defense classes in distantly related plants, followed by insect colonization and speciation across those plant groups. In her case, the presence of coumarins in Rutaceae and Asteraceae facilitated host-shifts to the coumarin-rich Apiaceae in at least three groups of insects (Lepidoptera, Diptera, and Coleoptera). Berenbaum hypothesized multispecies reciprocity where similar chemistry among distantly related plants led to multiple insect host shifts, adaptation and speciation, and to reciprocal selection for escalated defense in plant lineages. As multiple herbivores act in concert on the same plant host, reciprocal defense in plants and subsequent counter-defenses in insect herbivores are potentially speeded up in multi-species coevolution.

Current thinking would suggest that such parallel adaptations in plants and insects do not necessarily imply coevolution for all lineages, as some insects may simply be “chasers” of the diversified plants or other species with similar chemistry without necessarily imposing strong selection on the plants. Nonetheless, patterns of parallel defense-offense radiations are an important corollary to diversifying coevolution as they point to the predictable role of specific plant defenses in counter-defense innovations in herbivores and why these interactions may escalate over macroevolutionary time. Furthermore, this notion distances coevolutionary processes from a pattern of congruent phylogenies of plants and herbivores (co-diversification, discussed below). Extreme convergence in defense-offense interactions has recently been demonstrated between plant-produced cardenolide toxins and insect tolerance mechanisms across distantly related taxonomic groups (Dobler et al. 2012, Petschenka et al. 2017, Karageorgi et al. 2019, Yang et al. 2019). These findings confirm Berenbaum’s notion of convergent reciprocal adaptations, but have yet to demonstrate elevation in diversification rates as a result of the emergence of defense or offense traits. The finding of chemical similarity impacting host use has also inspired a growing line of research where phylogenetic relatedness and chemical similarity are tested as alternative explanatory factors for herbivore community similarity (Becerra et al. 2009, Endara et al. 2017, Volf et al. 2018).

THE PHYLOGENETIC ERA

Initiated by Felsenstein (1985), the expansion of phylogenetics generated a wealth of well-sampled phylogenies, updated systematic hypotheses, and phylogenetic comparative methods. Advances on these fronts have enriched our understanding far beyond resolving the relatedness of species. Resolving phylogenies has generated new questions by revealing associations among the evolution of defense and offense traits, changes in host association, and increased diversification rates. First, insect host shifts onto new plant clades are frequently associated with enhanced rates of diversification (Janz et al. 2006, Fordyce 2010, Allio et al. 2021). For a specialist herbivore, host shifts can happen through 1) switching directly from one host to the next, or 2) an intermediate stage of host range expansion followed by subsequent specialization. A few well-documented examples are in support of the former scenario—termed “musical chairs hypothesis” and of its role in generating lineage diversity (Hardy and Otto 2014). For example, Murphy and Feeny (2006) documented an on-going host shift of Papilio machaon aliaska from ancestral Apiaceae hosts to novel Asteraceae hosts. They identified hydroxycinnamic acids as the driving chemical bridge that enabled this host shift. The latter scenario, termed “the oscillation hypothesis” (Janz et al. 2006, Janz and Nylin 2008), has garnered much more attention in recent decades, but its generality has also been questioned (Hardy and Otto 2014, Wang et al. 2017).

The expansion of phylogenies and comparative methods has also enabled us to test the role of putative key innovations in plant lineage diversification. However, only in a few systems have the traits enabling host shifts and accompanying radiation been unequivocally identified. Perhaps the best studied is the Brassicales-Pieridae system where, concordant with host switching to glucosinolate-containing plants, Pierid butterflies evolved nitrile-specifier proteins that detoxify glucosinolates (Wheat et al. 2007). The emergence of nitrile-specifier proteins was associated with enhanced diversification rates and this innovation was lost when the butterfly lineage shifted to host plants lacking glucosinolates. More recently, the same research group has shown that gene duplication events in the Brassicales substantially increased the diversity of glucosinolate compounds and resulted in two distinct bouts of enhanced plant lineage diversification (Edger et al. 2015). Temporally concordant with the fast plant innovation, two butterfly tribes (Anthocarisidini and Pierina) independently evolved genes for detoxifying glucosinolates and
subsequently radiated. This elegant combination of genetic mechanism, trait function, and macroevolutionary patterns in the chemical arms race of the Brassicales-Pieridae system is perhaps the most compelling case for key innovation-driven diversification (Table 1).

**DETECTING RELATIONSHIPS BETWEEN TRAITS AND DIVERSIFICATION**

Although there has been substantial debate and controversy in how to detect diversification rate shifts on phylogenies, recent advances are beginning to address past limitations of the stochastic birth–death models employed (Laudanno et al. 2021). In the plant-herbivore coevolution literature, however, most of the work on diversification has taken a different route. For candidate key innovations in host plants, for example, distinct approaches have been developed to address quantitative versus discrete defensive traits. For a quantitative trait, researchers often set out to detect correlations between the trait expression levels and the species’ phylogenetic positioning (Harvey and Pagel 1991, Freckleton et al. 2008). For example, quantitative evolution of defense traits was associated with speciation in the milkweeds (*Asclepias* spp.), although different defense traits showed divergent patterns (Agrawal et al. 2009a,b, Agrawal and Fishbein 2008). Glucosinolate production in *Streptanthus* (Brassicaceae) followed a de-escalating phylogenetic trend which was independent of plant resource availability or stress (Cacho et al. 2015). And finally, terpene production increased in both richness and diversity as *Bursera* (Buseraceae) diversified, albeit at a slower rate than species accumulation (Becerra et al. 2009). Despite these strong correlative patterns of directional change in defense expression on phylogenies, cause and effect are especially difficult to disentangle in such analyses. Indeed, the question of whether traits drive speciation or speciation drives trait evolution remains remarkably understudied (Futuyma 1987, Agrawal et al. 2009b).

As for discrete traits, the association between trait evolution and changes in diversification is often detected at a higher taxonomic scale, typically among genera. Here we can cautiously infer causality because repeated evolution of such traits provides independent evolutionary replication. The evolution of latex and resin exudation in plants has long been held as the classic case of defense innovation spurring plant diversification (Farrell et al. 1991). Nonetheless, a recent revaluation of this pattern with updated phylogenies and improved trait characterization questioned its generality (Foisy et al. 2019). In a study of the evolution of extra-floral nectaries (EFN; Weber and Agrawal 2014), the authors found a consistent association between EFN and higher diversification rates among botanical families. When they zoomed in to six plant genera, the pattern of enhanced diversification rate was more variable and sometimes lagged behind the emergence of EFNs. Nonetheless, the case for EFNs provides the strongest evidence for the role of plant defenses facilitating diversification and is largely consistent across phylogenetic scales.

**CURRENT AND FUTURE DIRECTIONS IN MACROEVOLUTIONARY COEVOLUTION**

Following from the original coevolutionary hypotheses at the macroevolutionary scale (Table 1, first four rows), there is substantial evidence that plants evolve defensive biochemical novelty, often along a repeatable molecular path. The impact of plant defensive innovation on adaptive radiation is best known from a few case studies (EFNs, cardenolides and latex in milkweeds, glucosinolates in Brassicaceae). A substantial body of work shows that hosts shifts onto novel plant groups are associated with increased diversification. But overall, the evolution of insect resistance to novel plant defenses and its macroevolutionary consequences for insect herbivores has been studied in a coarse, non-quantitative manner. Too often do we couple a clade of specialist insects with a clade of host plants without identifying the underlying functional traits that enabled the initial host shift or quantifying the subsequent radiation, with the notable exception of the Pieridae-Brassicales system. As a result, causal analyses that connect specific innovations with diversification remain rare.

The patterns discussed in this section have been recapitulated in other systems (Table 1), suggesting some generality beyond plant-herbivore interactions. In sum, many pieces of the macro-scale coevolutionary hypothesis have been satisfied, albeit in few systems; but how often reciprocal adaptation is the cause of reciprocal radiations and what functional traits underlie this causation remain unclear. Although better resolved phylogenies, new comparative methods, and detailed knowledge of natural history and chemical mechanisms will further enhance our ability to address macro-scale coevolutionary hypotheses, it is unclear how much effort should be devoted to this area. We suggest that finer scale analyses (e.g., within large genera with well-resolved phylogenies) are likely the most profitable as they can be more easily coupled with mechanistic studies of adaptive traits, host shifts, and changes in diversification rates.

Finally, it bears reiterating that we should not interpret phylogenetic patterns, particularly the pattern of congruent phylogenies, as sufficient evidence for coevolution between interacting species (Brooks 1979). Congruent phylogenies have often been invoked as evidence for co-speciation in tightly interacting host-parasite pairs, particularly in obligate endosymbiotic species like *Buchnera* (Xu et al. 2018) and *Wolbachia* (Balvín et al. 2018). But as has long been known, joint vicariance can yield patterns of co-diversification without requiring any reciprocal adaptation (Brooks 1979). And as mentioned above, a “chaser” lineage may diversify onto a recently radiated host clade, but the chaser may play no role in the initial natural selection for
host plant defense traits or diversification of the host clade. Despite the fact that many species pairs that are co-speciating are likely to experience reciprocal selection, co-speciation does not require a change to lineage diversification rates, as envisioned in escape-and-radiate coevolution. Recent advances have improved analytical methods that account for phylogenies in trait correlations (Adams et al. 2018), allowing for more robust studies of co-diversification, host switching, and extinction in potentially coevolving species. Nonetheless, novel comparative methodologies that explicitly test coevolutionary hypotheses (Table 1) within the context of co-diversification are sorely needed.

The Microevolutionary Side of Coevolution

MEASURING SELECTION

Despite its conceptual appeal in explaining the staggering diversity of flowering plants and their insect herbivores, Ehrlich and Raven (1964) provided no explicit definition of coevolution, nor did they lay out the processes that might enable plant radiation following escape from herbivory. Janzen (1980) defined strict coevolution as evolutionary changes in two interacting populations through reciprocal adaptation. He distinguished species pairs with matching phenotypes due to coevolution from those merely interacting, the latter potentially generated by one-way adaptation (e.g., insects chasing plants). This emphasis on the process of coevolution necessitated tests of the essential elements of adaptive evolution. That is, to demonstrate reciprocal adaptation, we first needed to measure additive genetic variance for functional traits and test how fitness varies with variation in trait values. At the time of Janzen’s commentary, statistical and experimental methods to measure natural selection were only beginning to emerge. With the popularization of Falconer’s (1960) classic text and Lande and Arnold’s (1983) insight into the measurement of selection, the study of microevolutionary process gradually acquired its necessary toolkit.

During this bloom of quantitative genetics, Berenbaum et al. (1986) published a classic study on parsnip-webworm coevolution. The authors showed heritable genetic variance of furanocoumarins in wild parsnips (Pastinaca sativa), verified their defensive properties in functional assays, and detected selection imposed by parsnip webworms (Depressaria pastinacella) on furanocoumarin production and composition. This paper left an enduring legacy as an early empirical example of constrained evolution in which variation in an adaptive phenotype (defensive phytochemicals) is maintained by balancing selection on its benefits in the presence of herbivores and costs in their absence. Moreover, detailed analysis of furanocoumarin biosynthesis revealed a mechanistic basis for trade-offs among compounds and ultimately among components of fitness. Herbivory imposed strong selection for the production of bergapten in seeds at the cost of another furanocoumarin (sphondin) and nutrients for vegetative growth. Additive genetic variance of cytochrome P450-mediated metabolism of two parsnip furanocoumarins, bergapten and xanthotoxin, and their corresponding targets of selection were identified in parsnip webworms a decade later (Zangerl and Berenbaum 1997). This series of papers set the archetype of combining quantitative genetic and functional evidence to test for the presence of plant-insect reciprocal selection for more than two decades of fruitful research (synthesized in Rausher 1996, Geber and Griffen 2003).

In his 1980 commentary, Janzen also noted “diffuse coevolution”, which he defined as when either or both co-evolving populations are experiencing collective selection from an assemblage of species. In the context of plant-herbivore interactions, diffuse coevolution occurs when a focal plant species is fed on by an herbivore community or when a generalist herbivore feeds on multiple species of plants. Diffuse coevolution was later defined more quantitatively as a change in the strength or direction of selection in a pairwise coevolutionary interaction in the presence of additional plants or insects (Rausher 1996). As discussed above, simultaneous selective pressures imposed by a community of herbivores may have macroevolutionary consequences such as intensified defense escalation in plants; however, methods to distinguish and test for the collective effects of an entire herbivore community have not been widely developed. Coevolutionary outcomes of when a plant evolves in response to an herbivore community, and similarly, when a generalist herbivore evolves in response to multiple host plants, are likely more complex than predicted by the additive effects of simple pair-wise interactions, but further work is needed in this area (Lachin 2002, Wise and Rausher 2013, Hall et al. 2020).

Experimental (co)evolution has proven to be a fruitful method in the study of coevolution at the community and population scales, notably in microbial and host-microparasite systems (Table 1, Brockhurst and Koskella 2013). Regrettably, the relatively long generation times of plants and the mobility of insects have prohibited widespread adoption of this method in plant-herbivore systems. The few studies using experimental evolution in plant-herbivore interactions evolved only one of two coevolution populations for relatively few generations (Agrawal et al. 2012, Gompert and Messina 2016, Ramos and Schiestl 2019, Magalhães et al. 2007). The lack of long-term direct observations and temporal sampling of coevolving populations has thus limited our ability to unequivocally assess reciprocal responses to selection over time. Nonetheless, an alternative approach comparing different communities emerged in the 1990s that filled this gap.
GEOGRAPHICALLY STRUCTURED COEVOLUTION

Building on the quantitative genetics era marked by Berenbaum et al. (1986) was a wave of coevolutionary studies that focused on the outcome of selection between two interacting populations within a single community. However, the bridge between local selection within a community, population divergence, and ultimately the formation of new species was largely unnoted until the publication of Thompson’s geographic mosaic model of coevolution (1994). As pointed out by Thompson (1994, 1999), the strength and direction of reciprocal local adaptation differs spatially and temporally across the landscape, resulting in varying degrees of phenotype matching between coevolving populations in different communities. Factors such as how long the local pair has been coevolving, the extent of gene flow between distant communities, and the presence of alternative interactions in communities likely contribute to such variable dynamics. In the classic parsnip-webworm system, Zangerl and Berenbaum (2003) found frequent plant defense-insect detoxification matches among 20 communities, with exceptions being explained by the presence of an alternative host plant. As another example, specialist weevil seed predators (Curculio camelliæ) that feed on Camellia japonica showed strongly matched rostrum length (the agent to penetrate fruits and access seeds) to pericarp thickness (the barrier to access seeds) among 17 populations (Toju and Sota 2006a,b). Nonetheless, the strength and balance of the interaction varied latitudinally, suggesting that other biotic factors are influencing this interaction.

The study of Lithophragma (Saxifragaceae) and their pollinating herbivores (Greya moths) has yielded a substantial body of evidence for the geographic mosaic model. In a study of the pairwise interaction between L. parviflorum and G. politella among 12 communities, the effect of moths on seed capsule development spanned the spectrum from beneficial to detrimental (Thompson and Cunningham 2002). This study indicated that the evolutionary outcome of the interaction between the same two species differs significantly between locations, likely due to varying abiotic conditions and the presence of other insect herbivores or pollinators. When surveyed across multiple Lithophragma and Greya spp., Lithophragma populations varied significantly in floral morphologies and volatile production in relation to the composition of Greya spp. present (Thompson et al. 2017, Friberg et al. 2019). These studies indicate that communities of coevolving populations fine-tune their traits in reference to their local abiotic and biotic context, and this will yield a heterogeneous landscape of co-evolving communities whose complexity exceeds that predicted from the interaction within any single locality. It is worth noting that not all coevolving species will exhibit geographically structured interactions. Dispersive species with high rates of gene flow may have genetically homogeneous meta-populations. For example, Yucca elata (Agavaceae) and Yucca filamentosa both show little to no within or between population variation in floral scents (Svensson et al. 2005, 2006). This result may be due to purifying selection imposed by pollinating Tegeticula moths across the landscape, or it could be attributed to rampant gene flow facilitated by long-range Tegeticula movement.

Community ecology clearly matters for evolutionary outcomes, but the specific processes through which local community-level interactions trigger feedbacks to macroevolutionary patterns are not well-explored (Fig. 1). In each of the three population-level cases described above, deviations from the expected phenotype matching were broadly explained by varying abiotic or biotic contexts, but none specifically measured how the strength and direction of natural selection varied in relation to those changing environmental contexts. Thus, we are still left with the missing link between the causes of variation in selection which produce geographic structure in coevolving populations and whether this leads to the formation of new species.

CURRENT AND FUTURE DIRECTIONS IN MICROEVOLUTIONARY COEVOLUTION

Berenbaum et al. (1986) provided an archetype for testing microcoevolutionary outcomes based on Janzen’s strict definition of coevolution. The integration of measuring trait function, deciphering the genetic basis of traits, and measuring selective strength has been widely adopted. This approach has been used so widely, in fact, that reports of additive genetic variance, selection coefficients, and functional assays of putative defensive traits plateaued in the 2000s and have not provided many qualitatively new insights in recent years. It is now abundantly clear that reciprocal selection does indeed occur, but the burden of proof required to detail all of the essential elements for reciprocal adaptation can be onerous. Ultimately we are still left with pressing questions: Does coevolution simply generate increasingly exquisite adaptive fits between interacting populations without spurring speciation, or do coevolving populations spin off distinct locally adapted populations, a subset of which become new species (Hembry et al. 2014)? How do environmental and community variation affect the strength and direction of reciprocal selection, and what combinations of environmental and community contexts push co-evolving lineages towards speciation and enhanced rates of lineage divergence (Maron et al. 2019)?

Concluding Remarks

The grandness of Ehrlich and Raven’s vision for the study of coevolution has percolated through time and well beyond plant-herbivore interactions (Table 1). The two studies by May Berenbaum that inspired our article advanced the field by laying out
and testing explicit macro- and microevolutionary hypotheses on coevolution. For more than half a century, methodological advances have continued to spur empirical studies that elaborated and expanded upon aspects of these ideas; progress in testing other parts of their vision remains stagnant. Phylogenies and comparative methods have advanced our ability to detect macrocoevolutionary patterns and generate causal hypotheses between traits and diversification. Within-population adaptive processes now have rigorous genetic and functional support, but their repercussions for speciation and landscape-level variation still have not been resolved. The outstanding empirical gap linking microevolutionary processes and macroevolutionary patterns in coevolutionary interactions has persisted for decades, despite persistent calls for research in previous reviews (Janz 2011, Althoff et al. 2014, Suchan and Alvarez 2015).

Revisiting the core missing link between population-level microevolution and macroevolutionary radiation has yielded several hypothesized mechanisms (Althoff et al. 2014, Marquis et al. 2016, Maron et al. 2019), but these await empirical testing. Exclusion of herbivory has been shown to lead to locally relaxed defenses (Stenber et al. 2006, Agrawal et al. 2012), but it is still unclear whether novel defense leads to an adaptive zone for subsequent plant radiation. Furthermore, little evidence exists for how local adaptation and resulting geographic mosaics of defense-defense traits will yield reproductive barriers via reduced gene flow, assortative mating, or other mechanisms explicitly conducive to lineage divergence (but see Smith and Benkman 2007, Parchman et al. 2016). The best evidence to date that coevolution can promote adaptive divergence comes from microbial systems where the process can be experimentally manipulated and observed over many generations (Table 1). From the perspective of herbivores, diversified plant lineages can be reasonably considered empty niches and subsequent insect diversification onto these new niches is straightforward to imagine (Nyman et al 2010, Winkler and Mitter 2008). Insects in the middle of an ongoing host shift allow us to observe natural lineage divergence in real-time, and systems where the underlying mechanism of that host shift have been identified (e.g., Papilio machaon alaska, Murphy and Feeney 2006) are promising places to fill our long-lasting knowledge gap of the link between micro- and macro-coevolutionary processes. Similar processes may be observable in other host-parasite interactions, providing a window to the generality of host-switching and species divergence (Benkman 1993).

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