Review

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An ecosystem approach to understanding and managing within-host parasite community dynamics

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Hosts are typically coinfected by multiple parasite species, resulting in potentially overwhelming levels of complexity. We argue that an individual host can be considered to be an ecosystem in that it is an environment containing a diversity of entities (e.g., parasitic organisms, commensal symbionts, host immune components) that interact with each other, potentially competing for space, energy, and resources, ultimately influencing the condition of the host. Tools and concepts from ecosystem ecology can be applied to better understand the dynamics and responses of within-individual host–parasite ecosystems. Examples from both wildlife and human systems demonstrate how this framework is useful in breaking down complex interactions into components that can be monitored, measured, and managed to inform the design of better disease-management strategies.

What is ecosystem ecology and how can it be applied to studying host–parasite interactions?

Hosts are typically coinfected by multiple parasite species, broadly defined to include any infectious disease-causing agent (see Glossary), concurrently and over the course of their lifetime, and they combat these infections through a highly-diverse array of responses. This results in potentially overwhelming levels of complexity. Historically, ecologists have developed theories and experimental tools to break down complex systems into components that can be measured, manipulated, and compared across different habitats. In particular, ecosystem ecology has developed concepts and tools to understand the complexity arising from interactions between members of a community and the biotic and abiotic components of their environment.

An ecosystem is defined as an assemblage of organisms and the biotic and abiotic environment in which they occur [1]. Within an ecosystem, multiple species interact with each other, both directly and indirectly, via their shared environment and resources. The structure of the ecosystem and the many species interactions within it determine the abundance and dynamics of each species, the flow of energy through the system, and ecosystem functions such as stability and productivity. Ecosystem ecologists have developed a range of tools and concepts that can help us too understand these complex interactions, and to predict how the ecosystem will respond to different perturbations (e.g., species loss or gain, environmental/anthropogenic change). Importantly, this concept of an ecosystem can be applied to different scales. For example, we may naturally think of a large geographical region (e.g., the Serengeti) as an ecosystem. Alternatively, an ecosystem may be defined at a smaller spatial scale, comprising a discrete and clearly delineated habitat (e.g., a lake or cave). Similarly, in this respect, for parasites, the individual host can be considered as an ecosystem. We therefore explore the application of concepts and tools from ecosystem ecology to understand the assembly, dynamics, and management of within-individual host–parasite ecosystems.

Hosts are commonly coinfected by multiple parasite taxa. These coinfections can be concurrent or consecutive, and occur over the course of the lifetime of the host [2–4]. Furthermore, nearly all organisms are also 'infected' with diverse, commensal microbial communities [5–8]. As in free-living ecosystems [9,10], these diverse communities of co-occurring pathogenic and non-pathogenic species may interact directly and indirectly, both with each other and with their environment (the host). Interactions among these species can have major consequences for the presence of a species (the ability of a parasite to infect a host [11]), species abundance (parasite burden/levels of parasitaemia [12]), the invasibility of a host to a novel parasite (i.e., host susceptibility to secondary infection [13]), and host response to infection (e.g., clinical impact of infection or consequences for other coinfecting species [14]). Interactions among parasites and the microbiome can also impact upon bacterial community composition [15], the success of parasite infections (e.g., mediate successful hatching of helminth eggs [16]), and the development of immune phenotypes (e.g., pathogen recognition and susceptibility to autoimmune disorders [17]). Furthermore, the within-host

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Glossary

Assembly rules: the processes determining the colonization of a habitat or ecosystem by the component species within it, as defined by the ecological relationships between the organisms, such as competition or resource needs.
Bottom-up: a process initiated by changes in resources, commonly food or space, used by other organisms (e.g., grass in traditional ecosystems, or red blood cells in a within-host ecosystem).
Carrying capacity: the population size for a given species that can be supported by an ecosystem, considering available resources and competitive interactions with the same and other species.
Coinfection: when a host is infected with more than one parasite.
Ecosystem: the biological members of a community with the biotic and abiotic components of their environment.
Ecosystem engineer: species that modify their environment and influence resource availability to other members of the community by actively changing biotic or abiotic components of the habitat.
Ecosystem properties: the term encompasses productivity (how much living matter can be produced), the services provided to living organisms (e.g., conversion of essential nutrients to usable forms) and to the environment (e.g., sequestration of carbon or nitrogen). We use this term to describe how multiple components influence the within-host environment, or 'host condition' (see definition below).
Functional group: a category of (usually) closely related organisms (phylogenetically) with similar roles in the ecosystem (i.e., feeding on the same resources and consumed by the same predators).
Habitat: where an organism lives (eats, sleeps, breeds).
Host condition: measures of host health, survival, and reproductive output. When the host is the ecosystem, host health, survival, and reproductive output can all be metrics of emergent ecosystem properties owing to the diverse impacts parasites can have on their host.
Homeostasis: regulation of internal conditions of an organism.
Immune phenotype: describing the multiple components of the host immune response in particular conditions. This concept is most useful when multiple cell types or immune factors change in response to an infection (e.g., cytokines and cell types involved in a Th1-type response).
Niche: the role of an organism in the environment defined by how it uses resources in time and space. Niche segregation occurs when two species diverge in what niche they occupy, resulting in reduced competition.
Parasite: an organism which relies on another organism for food or space resources and ultimately negatively impacts on its host. We use this term in a broad sense: in other words, this refers to macroparasites (e.g., worms, fleas) and microparasites (e.g., bacterial or viral pathogens) which cause disease in their host.
Parasitemia: the amount of parasite infectious stages in a host, most often used for internal parasites. This may also be termed parasite load for internal or ectoparasites.
Pathology: detrimental effects that the host incurs due to a parasite infection or overactive immune response, such as weight loss, anemia, or cell/tissue damage.
Perturbation: a disruption to a community or ecosystem.
Productivity: the amount of biomass (living matter) produced from a unit of space.
Stability: the ability of a community to return to an initial equilibrium following a perturbation.
Structure (ecological usage): an assessment of how random or non-random the composition of a community is.
Top-down: changes induced by direct actions of another organism; in other words, predation in traditional ecosystems or the immune system in within-host ecosystems

environment contains a diverse suite of host-derived components (immune cells and molecules) that are intimately linked with the various pathogenic and non-pathogenic infecting species. Considering these immune components alongside the parasites and commensal species provides a view of interacting community members, existing within the environment of each individual host.

We review here the applicability of ecosystem ecology for understanding and managing within-host parasite ecosystems. Our specific goals are to (i) illustrate conceptual parallels between ecosystem ecology and the within-host processes that occur during coinfection, (ii) provide examples from wild animal and human systems where the application of ecosystem ecology tools and concepts could improve our understanding of within-host parasite dynamics, and (iii) suggest general guidelines for the application of ecosystem ecology concepts to studying wildlife disease.

The host as ecosystem

A beneficial aspect of within-host ecosystems is that the host is a clearly defined unit, and established methods exist for quantifying the component species infecting it and the immune components that respond to those infections. Indeed, the rapid, dynamic manner in which hosts respond to the species within it, coupled with the relatively short lifespan of an individual host (compared to that of traditional ecosystems), means that within-individual host–parasite ecosystem processes occur on rapid timescales, which can be observed with a high degree of replication. Furthermore, data on host condition, measures of host fitness, survival, and reproductive output, or heath (e.g., fat scores, behavioral metrics) can be obtained, providing an ecosystem-level metric of the outcome of the interaction between host and parasite [18].

How perturbations affect emergent ecosystem properties have been the focus of much research in free-living ecosystems [19,20]. When considering processes at the level of the individual host, the growing field of ecological immunology aims to measure how the host response to infection affects ultimately influences host fitness [21–23]. Studies suggest that immune function is costly: in other words, hosts increase energy consumption (increase metabolic rate) when fighting an infection [24], leading to overall higher energy needs. Such energy demands by the immune system may cause trade-offs or competition for limited hosts resources between immune function and other physiological processes such as reproduction and growth [25,26]. We focus here on host condition as our assessment of the impact of parasitic infection on the host because it likely to be affected by both increased exploitation by parasites and by immune processes induced in response to infection.

A key point when considering the host as an ecosystem is to recognize that individual hosts are not homogenous environments in which parasites, resources, and immune components are fully mixed. Free-living ecosystems are typically structured into connected 'patches', comprising subsets of potentially strongly-interacting species (e.g., via resource competition or predator–prey interactions) that are linked with other patches by flows of energy, nutrients, or species (e.g., salmon migration linking sea and freshwater stream habitats [27]; conversion of nutrients as in above- and below-ground plant–soil communities [28]). In the same way, the internal environment of an individual host will be structured into 'compartments' (e.g., organs, tissues, cells) such that parasites typically occupy different habitats within the host (e.g., the gut, blood, liver, or skin), but with some potential for movement of nutrients, energy, and 'species' (parasites, but also cell types and molecules that use or convert within-host energy and nutrients, including immune components) between compartments [6,29]). Within these compartments, species abundances and dynamics are affected by both 'bottom-up' interactions due to shared resources or space, and by 'top-down' interactions due to a shared immune response (Figure 1) [30]. Recognizing the compartmentalization of the host is

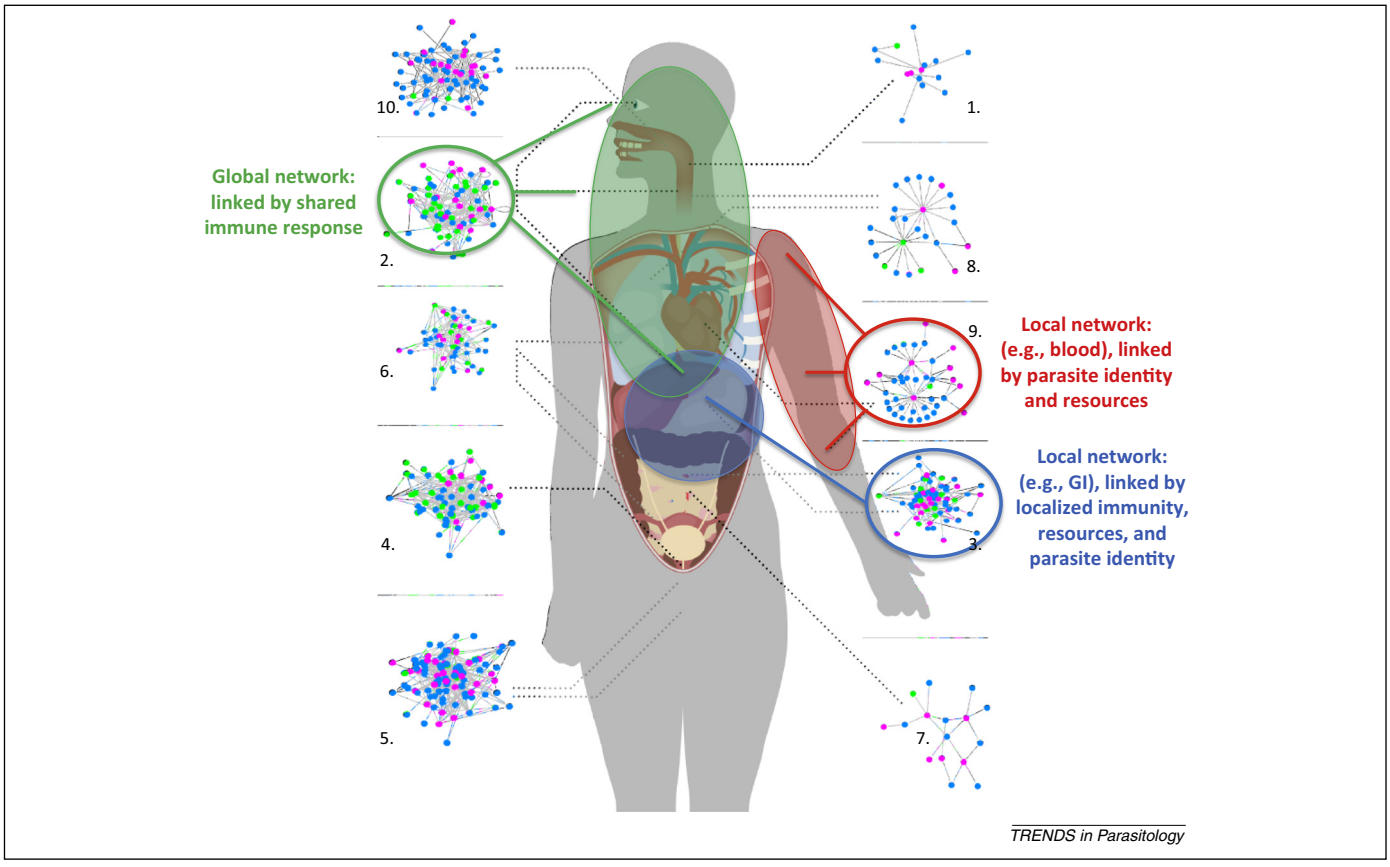

Figure 1. Network analysis of human parasites illustrating the compartmentalization of within-host communities [green, global network; red, a local network in blood; blue, a local network in the gastrointestinal (GI) tract] (modified from [30]). Parasites could be linked through parasite identity, resource use, or shared immune interactions. Local interactions, such as within the habitat of host blood (9, red), were largely linked through parasite identity and resource use, while interactions within the GI tract (3, blue) were linked by all three mechanisms. Global linkages between multiple habitats (2, green) were mostly linked by common immune responses.

fundamental to understanding and predicting the outcome of coinfection for parasite abundance and host condition.

Ecosystem ecology concepts applied to within-host ecosystems

We highlight four key ecosystem ecology concepts: stability, limiting resources, community assembly rules, and ecosystem engineering, as well as the scales at which these effects can occur (local vs global interactions) that provide theoretical, experimental, and observational foundations for elucidating mechanisms involved in within-host parasite community dynamics and their impact on host condition (Table 1). We focus on parasite coinfection and how to assess their impacts on disease, but recognize that the microbiome is a huge component of diversity within hosts and creates much of the biotic environment in which parasites interact.

Stability

Over the course of the lifetime of an individual host, its burden of infection will likely fluctuate due to both parasite-mediated mechanisms, such as competition for infection sites, and host-mediated mechanisms, such as an immune response targeting or eliminating a parasite. In contrast to conventional wisdom, that suggests an uninfected host is a healthy host, it may be that hosts that maintain a stable parasite community but suffer few ill-effects will sustain the best condition, as has been proposed for free-living ecosystems [31]. That is, it may be preferable for a host to adopt a 'tolerance' response [32,33], thereby maintaining health in the face of infection, because clearing an infection may be energetically more costly or result in increased damage due to immunopathology. While there are many ways to define stability, generally a stable community is one that cannot be easily moved from an equilibrium state (a 'resistant' community) or one that returns quickly after a perturbation (a 'resilient' community) [34,35], and these are important characteristics of a 'healthy' host. Community stability is influenced by the nature and number of linkages and types of interactions among species within a community [35,36], together with the level of compartmentalization and number of relatively weak links present among members [37,38]. As with free-living ecosystems, we can ask what the characteristics of a stable parasite community are, and how we can assess stability in terms of the parasite community and host condition.

Perturbations, such as novel species invasions and local extinctions, have been studied extensively in terms of understanding stability in free-living communities [39]. In particular, species removal/addition experiments have been conducted in all types of ecosystems to measure the impact that species loss/gain has on community structure and

Table 1. How the four ecosystem ecology concepts apply to the within-host ecosystem

Ecological concept	Free-living ecosystem approach to quantifying concept	Related eco-immunology or animal physiology concepts and approaches	Predicted effects on within-host ecosystem (host condition) due to parasite infection(s)	Influence on coinfection
Stability	Measure change in community composition and ecosystem function before and after a perturbation.	Homeostasis. Measure energy needed to maintain stable internal conditions in response to a perturbation (e.g., experimental infection).	Increased energy needs and demands on thermoregulation during infection.	Parasites may disrupt the balance of energy input, use, and output within a host. This may fundamentally change physiological stability after infection.
Limiting resources	Experimentally test the outcome of species competition under varying abiotic conditions (e.g., resource levels) and/or biotic conditions (e.g., competitor abundances).	Trade-offs between physiological processes will likely occur due to host being a semi-closed system (e.g., between reproduction and mounting immune response).	Total host resources are reduced, leaving less energy for general maintenance and metabolic needs. However, significant localization of resource competition occurs within habitats.	Resource needs of parasite(s) and immune response will influence the viability of the infection and how the host responds. Parasite identity and function may influence local resource availability and parasite abundance.
Community assembly, priority effects, ecosystem engineers	Measure effects on biotic and abiotic composition and/or dynamics in the presence/absence of putative pioneer species or ecosystem engineers.	Infection with a parasite can alter the host environment to significantly influence immune profile, resource availability, and/or susceptibility to subsequent infection.	Immune modification, resource use, and tissue damage by first and subsequent parasites will affect host resource use and reallocation during infection.	The outcome of coinfection and probability of host–parasite competition depends on the order in which the host becomes infected by the different species and by the immune profile generated in response to parasites.
Local versus global interactions	Monitor effects of a perturbation at multiple scales (i.e., individuals, populations, communities) or different spatial scales, (i.e., local, regional, continental).	Some within-host habitats are more connected than others owing to function, location, or blood flow.	Predicted influence on host resources or health will be made based on specific local or global behavior of the parasite and immune response, in single and coinfection cases.	Nature (direct, indirect) and direction of parasite interactions (positive, negative, neutral) will impact on how host condition is affected (i.e., damage to host tissue, facilitation of coinfection, energetically costly immune response).

function measured over time [40,41]. The same types of perturbations can be conducted in within-host parasite communities by either using drug treatments to remove/reduce a target parasite or through experimental infections. The impact of the initial perturbation and response of non-target parasites will indicate resistance or resilience in the community [29]. The within-host ecosystem may be considered to be stable if there is little change in parasite community composition or structure (presence/absence or burden of species within that community) after the perturbation. For example, in a study of the parasite community of wild wood mice, Knowles *et al*. [42] showed that although anthelmintic drug treatment reduced nematode infections (the target parasite), burdens quickly returned to pre-treatment levels. The temporary reduction of nematodes also caused a concurrent, dramatic increase in coinfecting, non-target intestinal coccidial protozoans. Interestingly, as the nematodes re-infected the treated hosts there was a parallel reduction in coccidia burdens, which also returned to pre-treatment levels 4 weeks following treatment [42]. Hence, these within-host communities appear to be highly stable to perturbation, demonstrating resistance [only one coinfecting parasite group (of ~20 species measured) responded to a reduction in nematodes] and resilience because both nematodes and coccidians rapidly returned to pre-treatment levels. Clearly understanding the factors that determine the response of hosts and their parasite communities to a perturbation is vital for our ability to design effective disease-control strategies in coinfected hosts.

In some cases, a pertinent question is not only how stable is an ecosystem but what types of stability are possible. Alternate stable states have been observed in free-living ecosystems [43] where communities are stable in alternative forms or species compositions, determined by starting conditions or resource levels. Once in one of these states, the community can only switch to an alternative composition with extensive changes in environmental conditions [34]. Within a host, the immune environment and microbiome can be very important in determining the outcome of coinfection [15,44] because they can be altered by parasite infection, and thereby change the likelihood of infection by other parasite types, effectively switching from one community state to another. Resident *Streptococcus pneumoniae* communities in the noses of humans and rodents, while diverse, have a predictable composition of strains, and are usually non-pathogenic (commensal) in their host [45]. However, cross-reactivity of the local immune response to *Haemophilus*

influenzae infection, including an increase in complement-mediated opsonization and phagocytosis, can have non-target effects on *S. pneumoniae*, with some strains being more susceptible than others [46]. This changes the competitive environment in the nose such that common *S. pneumoniae* serotypes are out-competed by rare ones, changing bacterial community composition and diversity [46]. These different compositions of nasal *S. pneumoniae* communities can be considered as alternative stable states, dependent on coinfection with *H. influenzae*. Considering alternative stable states of within-host communities may identify hosts that are more susceptible to infection by pathogenic serotypes, and help to predict the possible range of alternative configurations following species removal through treatment or after species addition through controlled colonization [47].

Because parasites directly affect their host by consuming host resources, it is important to consider the stability of host physiology ('homeostasis'), especially in the case of assessing the impact of multiple infections. Parasite infection can disrupt homeostasis by altering the energy needs of the host, in other words influencing metabolism and thermoregulation. Endothermic hosts incur temperature increases as part of their integrated response to infection [48]. Studies using metabolic chambers, measuring host CO_2 production or O_2 use, have shown that metabolism increases in immune-stimulated laboratory mice, providing evidence that the production of immune cells is energetically demanding [49]. For example, measuring metabolic markers in the blood of little brown bats showed that those infected with the causative agent of white-nose syndrome used twice as much fat, their main energy reserves, as control bats [50]. The authors suggest increased metabolism caused by infection may induce hyperventilation to remove the excess CO_2 in the blood of the bat, and subsequently increase activity and body temperature. This disruption to the physiological stability of the host is significant, as seen in the high rates of mortality of infected bats in North America [51]. Hence, measuring host physiological responses to infection can allow quantification of resistance or resilience to changes in its parasite community.

Limiting resources

Because energy inputs into an ecosystem are not unlimited, this can lead to competition for available resources, and ultimately limit the carrying capacity of an ecosystem (how many organisms can be supported). A fundamental concept in ecology is competitive exclusion, such that if species compete directly for the same resources then one of the species will be excluded. Diversity is then maintained by organisms using resources in different ways [52]. Furthermore, competing species are able to coexist if the strength of competition among individuals of a given species is greater than the strength of competition between species [1]. As in free-living communities, parasite species tend to aggregate into conspecific groups at specific sites of infection, for instance those organized by the internal organs of the host [30], and this niche segregation will tend to reduce competition between parasite species, facilitating their coexistence within the individual host [53,54].

However, unlike free-living ecosystems, the host environment and its parasites can directly compete for resources, including those needed for host metabolism, growth, and reproduction, and for confronting further parasite challenges. This resource competition between parasite and host means that the damage caused by parasite infection can feed back to affect host condition and the environment experienced by subsequent parasites, possibly affecting susceptibility to further infection [55,56]. For example, only specific ratios of iron and glutamine lead to stable 'coexistence' between host and parasite in laboratory mice experimentally infected with bacteria [57]. All other conditions lead to failure of the infection to establish or death of the host. Similarly, there is evidence of resource-mediated competitive interactions between disease vectors [58] and plant pathogens [59], and these types of interactions likely occur among other within-host parasite and microbial communities (e.g., [44]).

Theoretical approaches have begun to assess how parasites and host can interact through shared resources, including how variation in host resource availability affects how one parasite influences the transmission dynamics of another [60]. Recently, Cressler *et al*. [61] built a mathematical model that included multiple pathways for within-host energy allocation as well as a gradient of competition between the immune response and the parasite that ranged from little overlap in the shared resource pool to complete overlap with no prioritization of resources for the host. The results suggest that the level of overlap in resources leads to very different outcomes for parasite load, the density of immune factors, and host energy reserves (their proxy for host condition), including parasite elimination from the host and an overwhelmed immune response. Therefore, considering host and parasite as competitors may help explain the unexpected outcomes of host–parasite interactions and give insight into how these interactions affect host condition.

In parallel with such theoretical studies, empirical studies are now seeking to assess the relationships between infection, immune response, and host metabolic needs. In a recent study by Hawley *et al*. [62], house finches infected with a pathogenic bacterium and kept at cooler temperatures had lower parasite burdens but higher levels of interleukin 6 (IL-6), a proinflammatory cytokine, compared to control birds. This is in contrast to the expectation that birds at cool temperatures would need to expend more energy on thermoregulation, leading to a reallocation of resources away from immunity, and therefore higher parasite burdens and a lower immune response. However, the cooler temperatures may have created competition between the host and parasite for energy (host thermoregulation vs parasite infection/growth), with the host immune response outcompeting the parasite for host resources, allowing IL-6 levels to remain high and reduce parasitemia [62]. Further experimental studies of resource overlap and competition between immune response and parasites will be necessary to test the patterns predicted by Cressler *et al*. [61].

The next step of these models and experiments will be to include multiple parasites and variation in the ability of the host to use energy resources, for example, variable

immunocompetence [63]. Furthermore, although ultimately resource availability will be determined by host nutrient intake (as in [61]), actual resource levels for individual parasites and the host immune response will likely be determined by host energy allocation to different organs and functions, such that resource availability will differ substantially across within-host habitats. In addition, local resource levels can also be actively altered by the parasites themselves, as seen when specific strains of tumor-causing plant pathogens increase in abundance by using metabolites produced by the tumor they create [59]. Understanding how and which resources are shared between host and parasites will help improve how we treat and manage parasite infection and understand changes in host condition.

Community assembly rules, priority effects, and ecosystem engineers

There is great taxonomic diversity in the parasite community with which an individual host may become infected with throughout its life. However, it is well known from natural populations that all hosts do not become infected with all possible parasites, and some parasites seem to co-occur more or less often than expected by chance [64]. The order in which hosts become infected by different parasites may be predictable and repeatable, much like ecological succession in free-living ecosystems, suggesting there are 'assembly rules' within the communities. Ecology theory suggests that community assembly is determined by a balance of deterministic processes (e.g., niche-based processes such as competitive interactions between species), and stochastic, dispersal-based processes [65,66]. These concepts can be applied to the assembly and structure of within-host parasite communities, where interactions among parasite species and between parasite species and the host immune system can be considered as niche-based structuring forces, and infection events are akin to dispersal-based processes [67,68]. Recognizing how these processes interact and which is dominant under different circumstances is important for determining if parasite management strategies should target 'dispersal', the infection process, or attempt to manipulate individual parasite species via treatment and/or components of the immune response through vaccination.

Recognizing the ways in which the presence of one species alters the environment (either favorably or not) for subsequent species is key to understanding and manipulating parasite community assembly. In free-living communities, the order in which species colonize a new habitat, 'priority effects', is important for determining subsequent community composition and stability. An extreme form of such priority effects is where species actively change the biotic or abiotic components of the habitat, thereby modifying their environment: such species are termed ecosystem engineers [69]. Beavers are a classic example because they dramatically alter ecosystem structure by building dams and changing the hydrologic landscape, creating advantageous conditions for their persistence while affecting (either adversely or beneficially) many other species within that habitat [70]. Similarly, leaf-cutter ants facilitate forest succession in the tropics by altering soil structure and chemistry, creating gaps, and increasing light availability [71]. Similar processes may exist for parasite communities, where the outcome of infection is significantly influenced by the order in which parasites infect and how each one influences the characteristics of their immediate environment during and following infection. These changes to the host could take place through changes in available resources, the physical characteristics of infection sites, immune phenotypes, or a combination of factors. For instance, the immune, chemical, and physical effects of gut inflammation following infection with a specific strain of *Salmonella enterica* can facilitate subsequent growth of the bacterium in the lumen of the host gut, whereas otherwise it is outcompeted by resident gut microbes [72].

Changes in the immune phenotype of the host in response to infection can affect the establishment and growth of a subsequent parasite. Altering components such as cytokine profiles and dominant T-helper (Th) cell types leads to cascading effects on how the immune system responds and, subsequently, the intensity and duration of infection of other parasites [73–75]. Infection with gut helminths can facilitate viral infections through downstream effects on the host immune phenotype. Helminth infections typically activate Th2 cells, which induce changes in the cytokine profile of the host, including increased levels of IL-4 and of alternatively activated macrophages (AAM) involved in the elimination of helminths. In hosts previously infected with viruses that can remain latent in macrophages (i.e., γ-herpesviruses), the skew towards a Th2 response leads to an increase in viral reactivation and titers, due to both a reduction in the proinflammatory Th1 response as well as to IL-4 and transcription factor-dependent activation of a viral gene that induces reactivation [76]. In addition, the same helminth-activated macrophages can reduce clearance efficiency of an active viral infection, increasing viral titer following helminth infection [77].

Furthermore, many parasites, particularly helminths, modify the immune response of their host to facilitate their own infection or transmission [78]. These effects can alter the immune environment for other, coinfecting species, and such immunomodulatory parasites may be considered to be 'ecosystem engineers'. For example, helminths can downregulate inflammatory responses and increase their own survival while reducing host tissue damage; however, this can lead to increased susceptibility to other infectious agents, such as with other helminth species and malaria [79]. Chronic, sublethal infections may provide the best examples of such ecosystem engineering because they can have long-lasting effects on the environment, creating conditions for significant ecological and evolutionary responses from other coinfecting parasites and the host [13,80]. Using contemporary immunological tools it may be possible to assess how the environment is altered by infection, and which immune factors or other within-host habitat components are affected [81–83], thereby allowing predictions about the role of parasite ecosystem engineers on host condition.

Local versus global interactions

Discrete habitats within an ecosystem can become linked by the flow of energy and nutrients. Habitats within a host

are intrinsically linked because they are all part of one organism. However, as in free-living ecosystems [35], there is a high degree of compartmentalization in infection sites within individual hosts, and in the localization of host immune responses to infection [84,85]. Hence, infection by one parasite species may not necessarily influence another, even within the same host. An analysis of a human coinfection network showed dense clusters of closely-interacting parasites and localized immune components, organized around specific and relatively discrete habitats (i.e., organs or organ systems) within the body of the host. Interestingly, parasites with broader interactions (i.e., those occurring between habitats) were more often linked by shared immune responses (Figure 1) [30]. Interactions between parasites, therefore, may be most likely within habitats (i.e., for space or local resources), while among-habitat interactions may only occur through systemic mechanisms.

The localization of such interactions is illustrated by evidence of competitive release between the nematode *Heligmosomoides polygyrus* and coccidial parasite *Eimeria hungaryensis* in wild mice [42]. Experimental removal via drug treatment of the nematode resulted in a dramatic increase in *E. hungaryensis* intensity, which inhabits the same section of the small intestine as *H. polygyrus*. Such competitive release was not seen for another *Eimeria* species (*Eimeria apionoides*) that infects further down the gastrointestinal tract. However, unlike direct competition for infection sites, effects of locally stimulated immune responses can remain local and only affect a single habitat, as seen with bacterial infections in the nose [46], or have a global effects, when intestinal helminths influence viral load circulating in the host blood [76,77]. As in the study by Griffiths *et al*. [30], a network approach can help to simplify the analysis of complex communities by grouping parasite or microbial species by their interaction mechanism (e.g., immune- or resource-mediated) [15,47]. Analysis within compartments or by interaction mechanism can help predict the impact of parasites on each other and on host condition via consumption of resources or by damaging host tissues.

Suggestions for the application of ecosystem ecology concepts to studying coinfection

Breaking down the complexity of the impact of coinfection on parasite community dynamics and host condition can be simplified using an ecosystem ecology framework. In particular, it is important to recognize that within-host ecosystems are highly compartmentalized [30], with interactions occurring at multiple scales within the host. Hence, identifying within-host foci of interactions would be a major step in facilitating this understanding. Network theory, and associated network analysis tools, may be particularly valuable in achieving this [30,47]. Furthermore, such tools may help to define parasite 'functional groups' [86] in which parasites are grouped not taxonomically, but functionally, in terms of their infection characteristics (infection site, resources consumed) and how the host immune system responds to their infection. This could allow predictions of likely direct and indirect effects of one parasite on another, or could direct treatments to parasites of specific function instead of taxonomic identity [47].

Overall we suggest that identifying the parasite species, infection location and duration, previous and current infections, and host resource availability will all help to determine if a parasite will successfully infect a host and how that infection will affect host condition. With this in mind, we suggest several key issues, inspired by the ecosystem ecology concepts described in the previous sections, which should be considered to help predict the outcome of coinfection both for the parasites and host condition (Figure 2). We illustrate the application of our framework using the coinfection study between the helminth *H. polygyrus* and coccidia *E. hungaryensis* in wild wood mice (Figure 3) [42]; while this example is with only two parasite species, a stepwise approach of adding

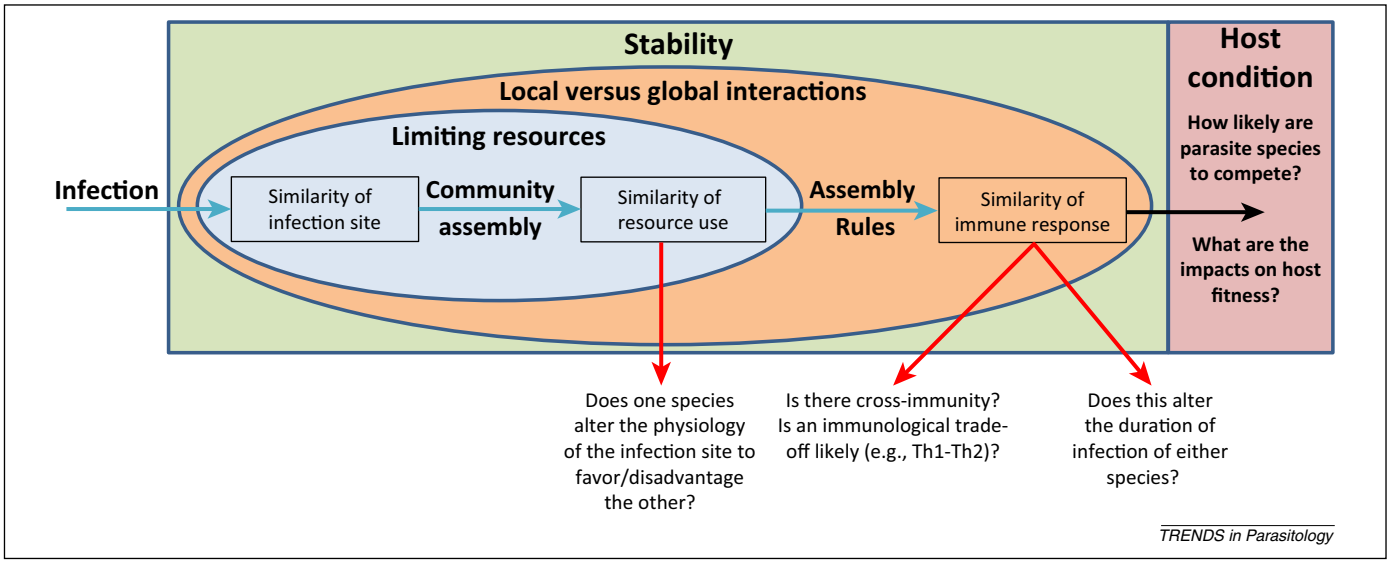

Figure 2. A framework with specific questions to ask when developing predictions of parasite–parasite and parasite–host competition and their impact on host health. These questions should be asked when the parasite infects a host alone and with other parasite species, and the answers to these questions should highlight the probable avenues of interaction between parasites and between host and parasite, specifically shared infection sites, resources, and immune response, and the limiting energy and nutrient resources available in the within-host ecosystem.

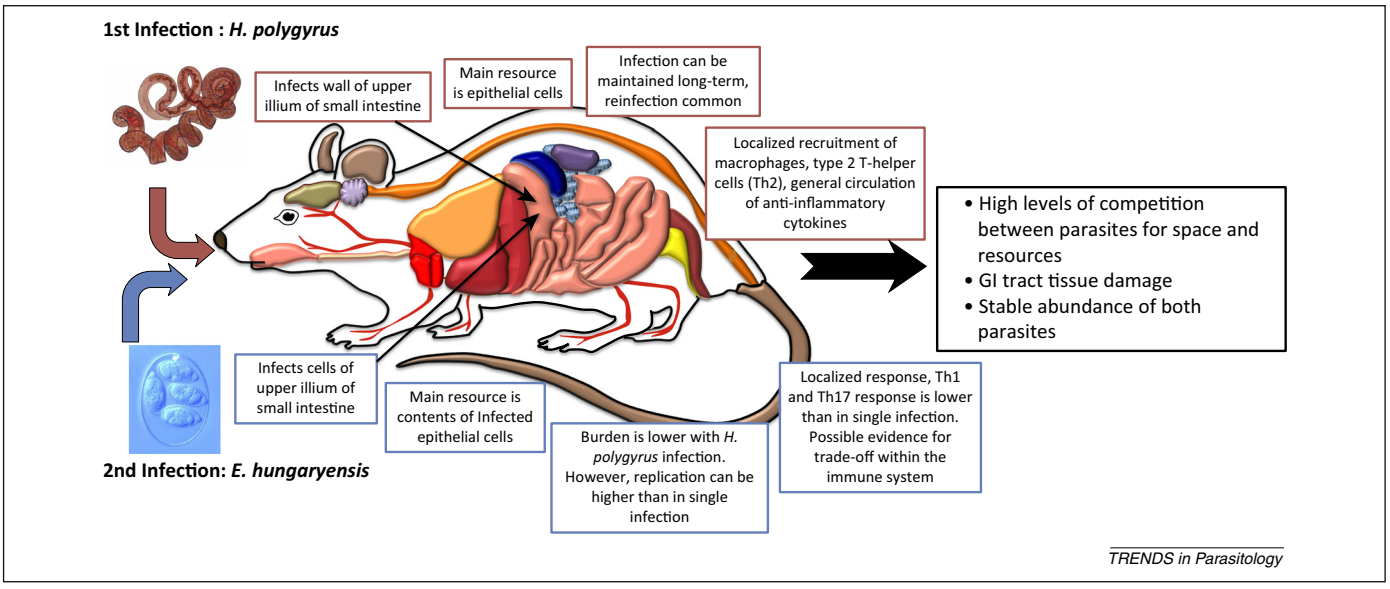

Figure 3. We illustrate how to begin applying our framework to a wildlife system with an example of coinfection between the helminth worm *Heligmosomoides polygyrus* and the coccidial parasite *Eimeria hungaryensis*. We assume the helminth infects first, followed by the coccidia, which is common in models of these parasite infections. Because these two parasite species share space, resources, are maintained at constant abundance when in coinfection, and influence the immune environment experienced by the other, they are expected to experience high competition and may have significant negative effect on host condition, a metric of within-host ecosystem properties. Abbreviation: GI, gastrointestinal.

parasite species can be done for more diverse communities. *H. polygyrus* and *E. hungaryensis* share an infection site within the host small intestine and may compete for space and nutritional resources because *E. hungaryensis* replicates in intestinal epithelial cells, the same cells which *H. polygyrus* ingests [80]. When *H. polygyrus* infects first, the evidence suggests that it modulates the host immune response, inhibiting the ability of the host to clear the helminth infection [78]. This may stabilize the system, keeping both helminth and *E. hungaryensis* burdens constant [42]. However, it is likely that the outcome of this coinfection could be very different if *E. hugaryensis* infected first, because of fewer initial changes to the immune environment that might influence helminths, but further experiments are needed. Similarly, as discussed previously, if *H. polygyrus* was coinfecting with *E. apionodes*, which occurs in a different part of the gut, we would expect reduced competition between these species and little influence on the infection success of the other. Little is known about how these parasites affect host condition in the wild, in terms of survival, reproduction, or health, and it is therefore difficult to predict the actual outcome of coinfection on the host. This is true of many wildlife systems, and this emphasizes the need for a combination of intensive monitoring in the field, ideally with experimental manipulation of parasite burdens, coupled with controlled experiments in a laboratory setting.

Limitations of the ecosystem approach for studying within-host parasite communities and further directions

While the ecosystem approach provides a framework for analyzing interactions between parasites and between parasites and their host, there are areas where the analogy breaks down. In particular, it is important to recognize that each individual host ecosystem is embedded within a larger ecological ecosystem. Clearly, the physiology and behavior of an individual animal will be influenced by extrinsic factors in its external environment, such as temperature, rainfall, and resource quality, as well as by its social environment. These variables can impact upon energy budgets [87], within-host resource allocation [62,88], and what nutrients are available to the host and parasites [44]. Conversely, not only will the external environment influence the infection status of an individual, but the combination of infections across multiple individuals will influence disease dynamics at the population level. To scale these individual-level processes up to populations, a necessary step towards managing disease spread, we need to utilize this hierarchical structure of ecosystems to understand how interactions among hosts affect parasite transmission dynamics [89]. Such an approach was recently illustrated through a combination of long-term monitoring and large-scale experimental manipulations of helminth burdens in wild African buffalo. Anthelmintic treatment, which reduced helminth infection, was predicted to actually increase the basic reproductive number (R_0) of bovine tuberculosis (BTB) to eightfold that of untreated individuals because of the ninefold increase in lifespan found in anthelmintic-treated, BTB-infected buffalo [81].

A major issue in wildlife disease management is to appropriately assess the cause and effect relationship between host condition and parasite infection: in other words, does parasite infection reduce host condition or are hosts in poor condition more likely to be infected? This can be difficult to determine [55,56] because investigating the causes of variation in the outcome of coinfection requires controlled experimental studies. Ideally these experiments would also manipulate the external environmental characteristics that are hypothesized to impact on host susceptibility and/or exposure to one or multiple parasites. Bringing animals into a laboratory or semi-natural condition (e.g., outdoor aviaries) where the external environment and parasite

exposure can be controlled, and changes in host condition monitored (e.g., temperature [49], fat stores [90], reproductive success [91]), will be necessary to tease apart the cause and effect relationship of individual condition, susceptibility, and levels of infection. These experiments then need to be fully integrated with long-term intensive and extensive studies, incorporating experimental manipulations, to truly understand and predict coinfection dynamics and impacts in natural populations.

Concluding remarks

The host is a unique type of ecosystem that can directly and indirectly compete with the species that live within it because it is simultaneously the environment, resource, and predator for parasites. This leads to a wide range of potential outcomes for host condition and the within-host parasite community. The occurrence and strength of parasite–host and parasite–parasite interactions are greatly determined by location, local resource levels, and localized immune responses. Therefore it is no longer sufficient to consider hosts as merely being 'coinfected'; instead it is essential to consider the identity of the parasites, where those coinfections occur, and through what potential mechanism they may interact. We suggest that ecosystem ecology provides us with many of the tools and concepts necessary to improve our ability to measure and assess how parasites impact upon hosts, potentially leading to better individual-level predictions and treatment of disease.

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