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IDEA AND PERSPECTIVE

An integrated framework to improve the concept of resource specialisation

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Abstract

Resource specialisation, although a fundamental component of ecological theory, is employed in disparate ways. Most definitions derive from simple counts of resource species. We build on recent advances in ecophylogenetics and null model analysis to propose a concept of specialisation that comprises affinities among resources as well as their co-occurrence with consumers. In the distance-based specialisation index (DSI), specialisation is measured as relatedness (phylogenetic or otherwise) of resources, scaled by the null expectation of random use of locally available resources. Thus, specialists use significantly clustered sets of resources, whereas generalists use over-dispersed resources. Intermediate species are classed as indiscriminate consumers. The effectiveness of this approach was assessed with differentially restricted null models, applied to a data set of 168 herbivorous insect species and their hosts. Incorporation of plant relatedness and relative abundance greatly improved specialisation measures compared to taxon counts or simpler null models, which overestimate the fraction of specialists, a problem compounded by insufficient sampling effort. This framework disambiguates the concept of specialisation with an explicit measure applicable to any mode of affinity among resource classes, and is also linked to ecological and evolutionary processes. This will enable a more rigorous deployment of ecological specialisation in empirical and theoretical studies.

Keywords

Distance-based specialisation index, feeding niche, null model, phytophagous insects, species interactions.

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INTRODUCTION

Ecological specialisation, the diversity of resources used by a given species (Colwell & Futuyma 1971), is one of the most pervasive concepts in ecology. It is a fundamental element of niche theory that by way of resource use and overlap, extends to the functional organisation of communities (Winemiller *et al*. 2001). Specialisation features among preferred explanations for high tropical diversity (e.g. Lewinsohn & Roslin 2008) and species coexistence (Chase & Leibold 2003). However, ecological specialisation is defined and evaluated in very disparate ways, which impairs the interpretation and comparison of theoretical and experimental results. With regard to specialisation in resource use, a specialist/generalist dichotomy is often employed without any clear definition of these categories, either for potential or realised use (sensu DeVictor *et al*. 2010).

In this paper, we build on recent advances in ecophylogenetics and null model analysis to propose an explicit concept of specialisation that comprises affinities among resources as well as their co-occurrence with consumers. This concept should be useful at the theoretical level, by meshing with evolutionary and ecological mechanisms to explain various levels of specialisation and their effects, but also operationally by providing a consistent and replicable way of measuring and comparing levels of specialisation. Our approach is applicable to any system in which there are different resource categories whose similarities can be assessed. Thus, it bears directly on consumer-resource as well as mutualistic interactive systems; in both cases, interactions comprise pairings of lineages, species or their equivalents. In fact, most studies of ecological specialisation focus on one of these modes of species interactions. Henceforth, we will refer particularly to plant–herbivore interactions, which also include the data set on which we demonstrate the proposed concept. However, it should be noted that the same approach can also be employed for non-trophic resources, such as habitat categories, as long as their similarities can be gauged in an objective way.

Most species tend to consume a small fraction of the kinds of resources available (Strong *et al*. 1984; Bernays & Chapman 1994; Thompson 2005; Barrett & Heil 2012); in this sense, virtually every species would be a specialist. Hence, in commonplace use (e.g. Scriber 1973; Beaver 1979; Kitahara & Fujii 1994), categories of specialisation are established arbi-

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trarily for a given number or range of host taxonomic units (species or higher levels) used by a consumer species. In most cases, operational definitions for levels of specialisation are simple counts of resource classes; in network representations of ecological interactions, they appear as the number of links (L), its average per species (L/S) and its distribution over some or all species in a community (Dunne 2006). This approach is also often applied in other kinds of interaction, such as pollination networks (Ollerton *et al.* 2007) or entire food webs (Dunne 2006).

The taxonomic scaling of resources used makes sense because most consumers feed on related species, due to the phylogenetic conservation of traits that enable or restrain consumption, such as chemical defences or cues (Rasmann & Agrawal 2011). With the increasing availability of phylogenies, host phylogenetic relatedness is being incorporated in newer measures of host range, although these are not yet widely used (Symons & Beccaloni 1999; Beccaloni & Symons 2000; Morse & Farrell 2005; Pellissier *et al.* 2012; Davis *et al.* 2013). Further progress was made by Weiblen *et al.* (2006) who employed measures of phylogenetic community structure to assess herbivore specialisation relative to the entire set of host plant species in the system studied. However, their null model does not take into account differences in host abundance nor in the frequency of herbivore–plant interactions, so that any plant is equally likely to interact with any herbivore.

The selective use of resources among the potential range available to each consumer species is a key feature of ecological specialisation (Colwell & Futuyma 1971; Manly *et al.* 1993). We propose to integrate the phylogeny of resource species with the co-occurrence between consumer and consumed species groups in our concept of specialisation. Other, non-phylogenetic criteria, e.g. functional (Junker *et al.* 2013) or habitat/spatial (Chazdon *et al.* 2011), can also be used to gauge affinities among potential and used resources, but such alternatives are not pursued here.

In the following sections, we formalise this concept of specialisation and outline a null modelling approach, in which host range is measured as phylogenetic relatedness and compared with the null expectation of random use of the plants with which the herbivore co-occurs. A progression of null models correspond to increasingly constrained ecological processes. This framework is then applied to a large database of endophagous flowerhead herbivores of Asteraceae in Brazil, in which we can examine its behaviour and results at different spatial scales.

DEFINING AND EVALUATING SPECIALISATION

We define a specialist as a species that selects a subset of the resources available, so that the components of the used subset are more related (or similar) than expected by chance. Conversely, a generalist selects resources less related (or similar) than expected by chance (Fig. 1). The inclusion of relatedness or similarity in the concept of specialisation builds on the premise that, the more similar a given set of resources, the more likely that shared adaptations will enable their use with equivalent costs. When the resources are species, this similarity can be approximated by phylogenetic relatedness, and in this case

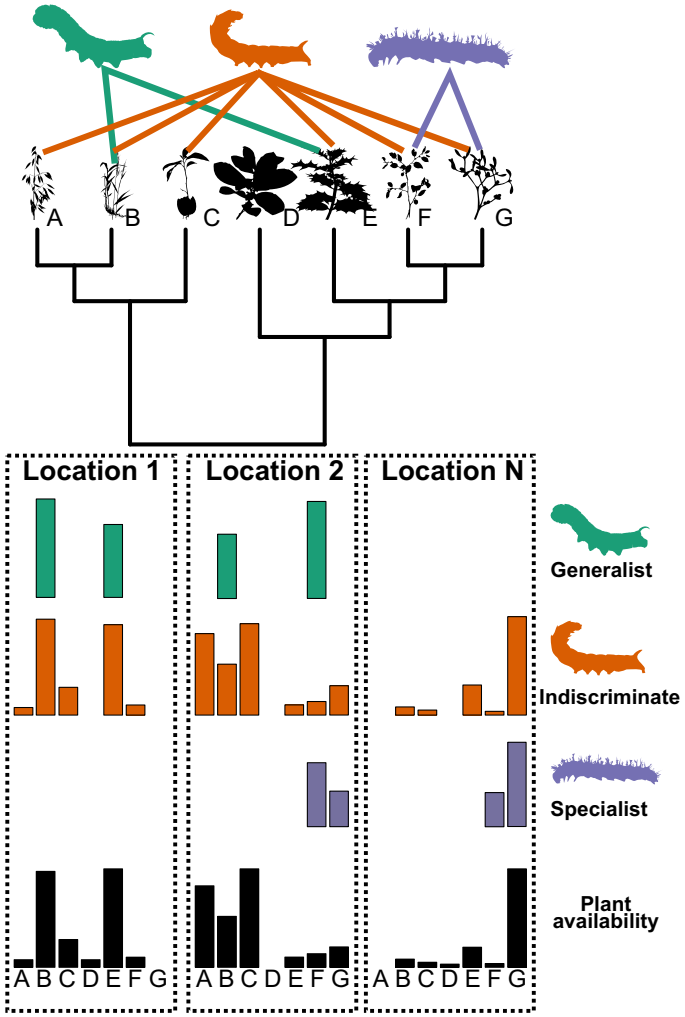

Figure 1 A schematic representation of the factors considered in the specialisation framework we propose. Resource use is assessed, and the similarity among used resources is calculated by means of the mean pairwise distance (MPD) calculated from a phylogeny or similarity dendrogram of the whole resource pool. This observed MPD is then compared to a null distribution obtained from sampling resources available for each consumer, taking both abundances and co-occurrence into account. Species that feed in an over-dispersed set of resources (green) are generalists, while species using clustered resources (purple) are specialists. When resources are used according to their availability (orange), consumers are considered indiscriminate feeders.

the problem is similar to the non-independence of species in the comparative method (Felsenstein 1985). We address this issue by measuring the phylogenetic scope of a consumer's diet. For this purpose, we employ a widely adopted metric for relatedness among species in communities: the mean pairwise phylogenetic distance – MPD (Webb *et al.* 2002). The MPD metric is the mean phylogenetic distance between all pairs of species in a given subset of the species pool in question. When applied to the list of resources used by a species, it is the mean phylogenetic distance between each pair of resource components in a diet. MPD is fairly independent from species richness and therefore also from sampling intensity, contrary to phylogenetic diversity (Faith 1992), which tends to be highly correlated with species richness (Clarke & Warwick

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2001; and also in our data set, see Fig. S1), MPD also reflects more properly the idea of relatedness within the resource use of a given group of species.

To evaluate if observed diet breadths deviate from expectation according to the phylogenetic relatedness of potential resources, we propose null models that sample the pool of available resources. Observed diet breadths are then standardised by the null model to gauge how clustered or dispersed they are. As in any null model approach, a crucial step is the definition of the sampling pool, which is the potential resource range for the consumer species. We propose that when studying a given set of consumer species, the full list of resource species recorded for that set of consumers be considered the resource pool. In this way, one can compare different levels of specialisation for a given set of consumers, without overestimating specialisation by including resources outside the known range for the group of species being studied. A less restrictive choice would also encompass similar or related available resources (e.g. in the same taxon or phylogenetic branch) whose use was not observed in that study but is known from other areas.

After defining the potential resource species pool, the next step is assigning different weights to the resource species based on the availability of each resource class for different consumers. Our initial null model assigns equal probability to all resource kinds, and is equivalent to shuffling the resources' phylogeny while maintaining its richness (number of resource species) for each consumer. In a second, improved model, resources are also weighted by their abundance or commonness; in this way, the use of a single phylogenetically unique resource represents a stronger specialisation signal, the scarcer that resource is (Feinsinger *et al.* 1981; Manly *et al.* 1993). Finally, a more comprehensive model also includes co-occurrence between resources and consumers. In this complete null model, sampling of the resources in the potential pool is weighted differentially for each consumer species, both by the resources' regional commonness (frequency of collection) and by the frequency of the consumer species in the different localities, so that only resources that actually co-occur with the consumer are selected, and resources are represented in proportion to the abundance of each consumer in the localities where it occurs (Fig. 1).

As many resource items as the observed frequency of the consumer (fixing the observed interaction frequency of each consumer species) are sampled and the null expectation for MPD is calculated. The observed MPD for the consumer is then standardised by the null model's mean and standard deviation, producing a Z-score. This index is equivalent to the net relatedness index (NRI) (Webb *et al.* 2002; Weiblen *et al.* 2006) widely used in community ecology, and has a similar interpretation. However, the null model we employ is more restricted, and represents potential resources more accurately than the unweighted sampling of the list of potential resources. Note that by incorporating resource-consumer co-occurrences, this null model is not directly comparable to the models in current use for phylogenetic community structure (Kembel 2009). We name this Z-score transform of the MPD the distance-based specialisation index (DSI).

To assess the contribution of different components of this null model and investigate the biases created by excluding them from specialisation assessments, we also calculated specialisation by using the abovementioned less-restricted models: (1) a weighted richness index (WRI) using the same null model, but employing resource richness (e.g. number of host species) instead of MPD as the measure of diet breadth. In this case, the number of resources each consumer feeds upon is also standardised by the resampling results, but the corresponding specialisation index leaves out resource relatedness. It is equivalent to calculating DSI assuming all species are equally related to each other (i.e. a star phylogeny). (2) A simple species list-based DSI_S with unweighted sampling of all potential resource species, regardless of their commonness or their local co-occurrence with consumers. In this model, only the number of resource species recorded for each consumer is maintained and that number is sampled without replacement from the entire resource list to produce the null model. This index is equivalent to the NRI as applied to herbivore host ranges by Weiblen *et al.* (2006) and is the same as null model 3 in Kembel (2009). (3) A DSI_S that incorporates commonness or abundance of resources but not their observed co-occurrence with consumers (DSI_A), which can be used when no local interaction data are available. Here, the entire set of resources is weighted by resource commonness, and then sampled for each consumer species for their resource frequency in samples. This model follows the same rationale as Kembel's (2009), null model 5; however, the latter if adapted to interaction studies would use interaction frequencies instead of resource availabilities as used in our model.

Both DSI_S and DSI_A are calculated in the same way as DSI. Additionally to analysing these continuous metrics, we also used them to classify species in the widely used categories of specialist and generalist. Species whose index values exceed 1.96 (i.e. in the upper 2.5% of the fitted normal distribution) are considered specialists; conversely, values below −1.96 indicate generalists, which feed on resources less related than expected by the available pool. The remaining species with an index between these cut-off values, are classed as indiscriminate feeders. Note that the latter category includes scarce species for which statistical power is too low to detect a deviation from the null expectation, together with well-represented species that are true indiscriminate feeders (Fig. 1).

This framework can be applied at a single scale, which can be a local assembly or encompass a larger spatial extent. However, if data span multiple scales, the analysis can be enriched by including the frequency of interacting species in local communities within the studied region. A species can be more specialised locally than in its entire geographical range (a local specialist, Fox & Morrow 1981). In the framework we propose, a local specialist should have significantly higher values for local DSI than for its regional DSI. To inspect local patterns taking regional differences among species into account, we measured local specialisation as the difference between the local and regional DSI for the species. This local distance-based specialisation index (DSI-L) is interpreted in the same manner as DSI, so that a species is a local specialist when this value exceeds 1.96. In this case local co-occurrence is not applicable, and DSI_A becomes identical to DSI. All models were built in the R statistical environment (R Core Team 2013), using functions from the ape package (Paradis *et al.* 2004) to

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deal with phylogenies and the picante package (Kembel *et al.* 2010) to measure relatedness of resources. The models built to measure DSI are available with a worked example in file S5.

MATERIAL AND METHODS

Sampling methods and database

We used a large data set on endophagous herbivore insects that breed in Asteraceae flowerheads, reared out from plants collected over eight years in four regions in Brazil – montane grasslands in the Espinhaço and Mantiqueira ranges, *Cerrado* vegetation in São Paulo state and coastal lower to montane grasslands in southern Brazil. In these regions, spanning 15° latitude, 26 localities were sampled (Table S1), most of them more than once and in different seasons. We followed the plant sampling and insect rearing procedures described in Prado *et al.* (2002): flowerheads in different developmental stages of all flowering Asteraceae species in each locality were collected and kept in plastic vials covered with a mesh cap to await adult emergence.

The sampling unit in the data set is a population of a given Asteraceae species in a site, and each entry is a recorded interaction, for which we have both plant and herbivore identity, location (with exact geographic coordinates in most cases), number of reared individuals and total weight of flowerheads. The data set comprises 3309 interactions among insects from two orders: Diptera (especially Tephritidae and Agromyzidae) and Lepidoptera (Tortricidae, Pterophoridae, Pyralidae, Gelechiidae, and three other occasional families); and Asteraceae belonging to 372 species and 83 genera; other reared herbivorous groups were more difficult to identify or separate into morphospecies. We selected the four insect families that are taxonomically better resolved at the species level: Tephritidae (TE, 106 species), Tortricidae (TO, 32), Agromyzidae (AG, 20) and Pterophoridae (PT, 10). This subset of the database comprises 2690 interactions and 337 plant species from 66 genera.

Analysis

To apply our analytical framework, we built a hybrid tree for all plant genera in our data set (Fig. S2). This was produced by combining the information from a composite tree of the whole Asteraceae family (Funk *et al.* 2009) for most of the genera, with the taxonomy as a surrogate for phylogenetic relationships of nodes for which no information was available. When even the taxonomy was unable to provide relationships, unresolved nodes were left as polytomies. On the basis of this tree, we calculated MPD both for the herbivore species' observed diets and in the four null models as described above. In all cases we ran each of the different null models for each herbivore species with 1000 repetitions, calculating the indices separately for each.

We also measured specialisation in local communities of the four localities in the Espinhaço Range with the most samples and the highest richness of plants and herbivores. We ran the null model for these local communities also with 1000 repetitions. For each locality we measured DSI-L for the occurring species, classifying their level of local specialisation according to the criteria proposed above. All analyses were performed in the R statistical environment (R Core Team 2013).

RESULTS

DSI was highly variable among the 168 herbivore species, with a strong bias towards specialisation. When classifying these herbivores as specialists (DSI > 1.96), generalists (DSI < −1.96) or indiscriminate feeders (−1.96 < DSI < 1.96), there are marked differences among families in the proportion of species assigned to each specialisation category (Table 1). As a continuous specialisation metric, DSI showed substantial disagreement with three host range measures in current use: S, the number of host–plant species, MPD among hosts and d' (Blüthgen *et al.* 2006) (Fig. 2). DSI was correlated with the number of host species (Fig. 2a, $\rho = 0.34$, $P < 0.001$) but, contrary to expectation, the correlation was positive. The relationship with MPD was negative as expected (DSI, similar to NRI, increases with phylogenetic clustering, contrary to MPD which is a distance metric). Nonetheless, DSI values were highly variable in herbivorous species feeding on closely related plants (Fig. 2b, $\rho = -0.55$, $P < 0.001$). The specialisation metric d', proposed by Blüthgen *et al.* (2006) showed no relationship with DSI (Fig. 2c, $\rho = 0.04$, $P = 0.67$). Even when adjusted for potential host plants, specialisation as evaluated against less-restricted null models was biased in relation to DSI (Fig. 3, Table 2). The WRI index, which ignores phylogenetic relatedness, showed higher levels of specialisation, especially in Tortricidae and Agromyzidae (Fig. 2a and b). Similarly, DSI_S, which gives equal probability to all potential host species, also estimated higher specialisation than DSI, although to a lesser degree. The degree of overestimation was not constant among herbivore groups; it was weaker for Tephritidae compared to the other families (Fig. 3c and d, 14% species of Tephritidae misclassified as specialists, against 30% for Tortricidae and 42% for Agromyzidae). On the other hand, DSI_A, whose model considers host commonness but not co-occurrence, gave very similar estimates to the full model, with almost no incongruence among them (Fig. 3e and f).

When applying these models at the local scale, local specialisation was very uncommon. With the exception of two species of *Trupanea* (Tephritidae) in Grão Mogol, all other species were not significantly more specialised locally than regionally (Fig. 4). These two species bred solely on *Trixis* in this locality, feeding on other genera elsewhere. Most species in different localities show less specialisation at the local than

Table 1 Classification of host range of 168 species of Asteraceae flowerhead endophagous insects belonging to four families according to the null modelling approach proposed in this study

Family	Specialists	Indiscriminate	Generalists	Singletons
Agromyzidae	2	12	0	6
Pterophoridae	4	3	0	3
Tephritidae	62	21	0	23
Tortricidae	7	15	1	9

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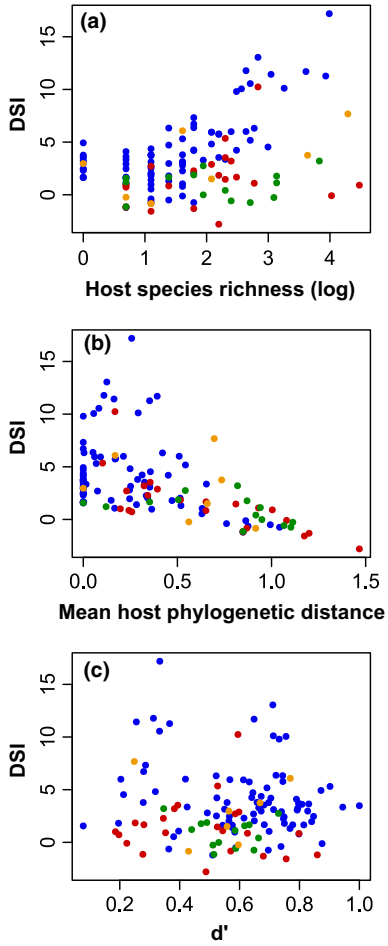

Figure 2 Relationship between the specialisation metric proposed in this study (DSI), and three measures of host range widely employed in the literature. (a): Host plant richness, (b): Mean phylogenetic distance among the host plants, (c): Blüthgen *et al.*'s (2006) *d'*, a metric of interaction diversity weighted for interaction frequency of the resources. Different colours represent species belonging to the four herbivore families included in the study: Agromyzidae (green), Pterophoridae (orange), Tephritidae (blue) and Tortricidae (red).

the regional level, with many differences in specialisation between localities for species occurring in more than one site.

DISCUSSION

We developed a null modelling framework to define and measure specialisation phylogenetically, taking the pool of potential resources, their commonness and co-occurrence between resources and consumers all into account. By applying this framework, the concept of specialisation is disambiguated and its measure becomes more explicit and comparable. Our example data set is illustrative in this regard, because it concerns a herbivore guild whose members, as far as known, are entirely restricted to a single plant family (Asteraceae); therefore, in accordance with conventional standards, all of them should be considered highly specialised (c.f. Scriber 1973; Beaver 1979). According to our DSI index, host range is highly overestimated by established approaches; specialisation

Table 2 Congruence between the classification of Asteraceae flowerhead endophagous insects as specialists, generalists and indiscriminate feeders according to the most restricted model proposed in this study (DSI) and three simpler models, that do not incorporate all host pool attributes

	WRI		
DSI	Specialist	Indiscriminate	Generalist
Specialist	**72**	3	0
Indiscriminate	25	**26**	0
Generalist	0	1	**0**

	DSI_S		
DSI	Specialist	Indiscriminate	Generalist
Specialist	**65**	0	0
Indiscriminate	24	**24**	0
Generalist	0	1	**0**

	DSI_A		
DSI	Specialist	Indiscriminate	Generalist
Specialist	**71**	4	0
Indiscriminate	2	**49**	0
Generalist	0	0	**1**

DSI takes into account phylogenetic relatedness and abundance of hosts and also plants and herbivore co-occurrence. WRI is equivalent to DSI without relatedness, measuring numbers of host plants instead. DSI_S measures relatedness but considers neither abundance nor co-occurrence. DSI_A is similar to DSI without plant–herbivore co-occurrence. Values in the diagonal (in bold), are congruences between the models. Values below the diagonal are overestimates of specialisation from the simpler models, while values above the diagonal are underestimates.

was highly variable within this guild and local specialisation was very rare. These results enable us to reappraise our understanding of this particular system in the light of the proposed approach, and furthermore to examine new paths of investigation of specialisation in various kinds of interactive assemblages, both antagonistic and mutualistic.

Specialisation in flowerhead endophages

The null modelling approach we employed clarifies the patterns observed in the Asteraceae-flowerhead endophage assemblage, first, by disentangling the effect of host commonness and actual co-occurrence on observed host ranges; second and most importantly, it tests explicitly the effect of phylogenetic constraints on the host ranges of phytophagous insects. The DSI_S showed some unexpected results in this herbivore guild. By comparing the specialisation evaluated under a fully restricted model (DSI) with simpler models that exclude some of the processes at play in host-plant selection, we were able to show that the detection of specialisation is strongly sensitive to the phylogenetic relatedness among plant species (excluded from WRI) and their geographical commonness (excluded from DSI_S). Conversely, in our data set the co-occurrence of hosts with herbivores (excluded from DSI_A), had no substantial effect, so that values of DSI and DSI_A were largely similar.

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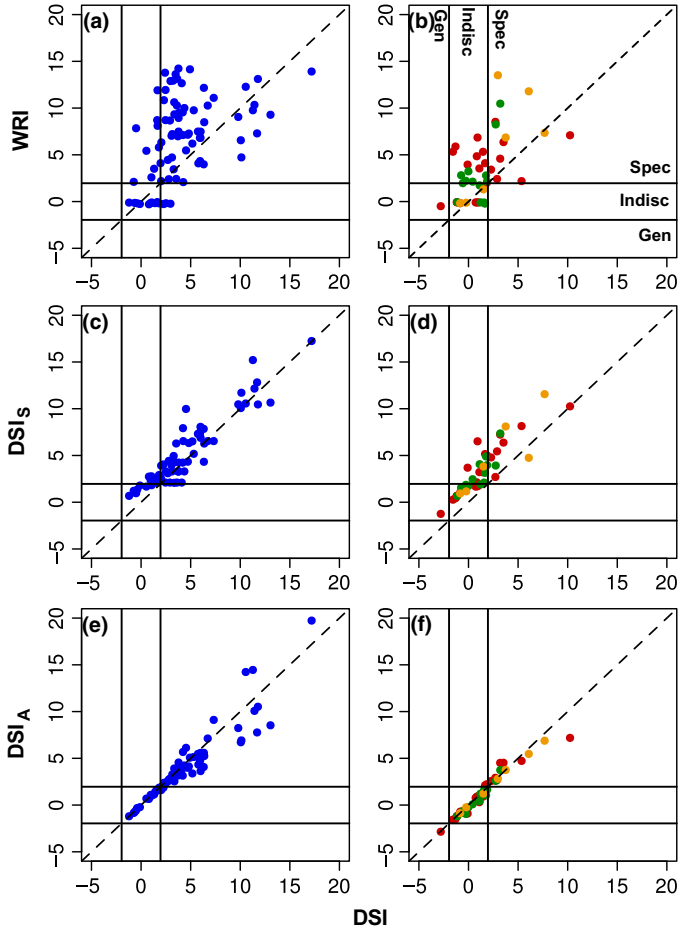

Figure 3 Specialisation in four families of herbivores feeding on Asteraceae flowerheads in Brazil, according to four different null models. In all figures, the most restricted model, measuring phylogenetic relatedness and accounting for plant commonness and co-occurrence with herbivores (DSI, abcissa) is matched to simpler models: a similarly restricted model with host species numbers instead of relatedness (a, b; WRI), a model based on relatedness in simple species lists (c, d; DSI$_S$) and a phylogeny-based model including plant commonness but not co-occurrence with herbivores (e, f; DSI$_A$). Left figures (a, c, e) are for Tephritidae and right (b, d, f) are for Agromyzidae (green), Tortricidae (red) and Pterophoridae (orange). Both WRI and the DSI measures are Z-deviates, with higher values representing higher specialisation. The continuous lines represent the boundaries for considering specialists (values above 1.96) or generalists (values below −1.96); intermediate species, generalists *s.l.*, are better designated as indiscriminate feeders. The dashed lines represent equal degrees of specialisation in both models.

The contrast of DSI and WRI shows that the widespread use of the number of host species as a measure of specialisation produces frequent overestimates. In fact, although most herbivores do feed on a smaller number of plants than would be expected by chance, for about 20% of the studied herbivorous species their hosts are not more closely related than expected by chance. If phylogeny is indeed a good surrogate for the plant traits that mediate host-plant selection (Rasmann & Agrawal 2011), herbivores feeding on small but phylogenetically random host groupings are limited by other factors that constrain the set of potential host-plants for each species. For this to happen, availability must not be phylogenetically structured; for example, it might reflect seasonal or microhabitat mismatches. In systems where the assumption that phylogenetic similarity is a key mediator of resource selection does not hold, the contrast between WRI and DSI may serve as a test of the importance of resource similarity in the resource selection process.

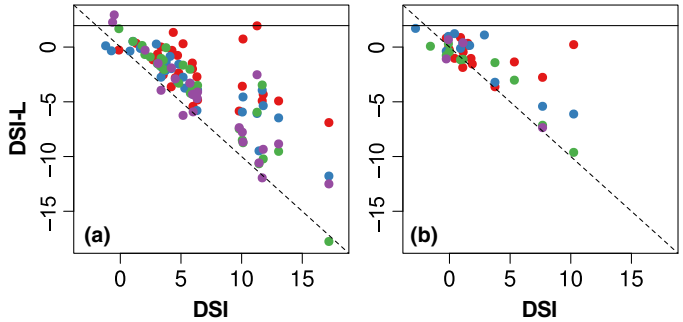

Figure 4 Relationship between regional specialisation and the change in specialisation at local scale (DSI-L) in four families of herbivores feeding on Asteraceae flowerheads in four localities at the Espinhaço Mountain Range in Brazil: Diamantina (red), Serra do Cabral (blue), Serra do Cipó (green) and Grão Mogol (purple). Negative values represent lower local than regional specialisation, while species with higher local than regional specialisation have positive values, being significant local specialists with values above the continuous line. The dashed line represents a DSI of 0 at the local level without taking regional DSI into account. (a): Tephritidae, (b): Agromyzidae, Tortricidae and Pterophoridae.

The second alternative model, DSI$_S$, incorporates another widely employed assumption, that herbivores are equally likely to feed upon all potential hosts regardless of their commonness and degree of co-occurrence. In our data set, many herbivore species feeding on several genera are nonetheless considered specialists according to DSI$_S$. This overestimation of specialisation occurs because rare unrelated plant species strongly raise the average phylogenetic distance among available plants, inflating the null expectation for the host ranges. Moreover, this model is unable to assess specialisation for apparent monophages, reducing the number of species for which a comparison is possible.

In the last comparison, which isolates the effect of host-herbivore co-occurrence in the complete model, there was a surprisingly high congruence between DSI$_A$ and DSI. With only six divergences, there is also no signal of bias in the estimation, as there were both over- and underestimates of specialisation, with very symmetric deviations (Fig. 3e and f). This indicates that for the majority of insect species most of the phylogenetic diversity (although not the species diversity) in the regional set of hosts is available to the insects in any locality.

The larger proportion of specialists among the Tephritidae compared to the other families (especially Tortricidae and Agromyzidae) indicates that within the specialised guild of endophagous flowerhead herbivores there are some strong discrepancies. As shown in Figure 2, even for similar numbers of host species or unstandardised MPD, Tephritidae tended to have higher DSI values than the other families. Note that some of the simpler null models do not show such marked differences. WRI and DSI$_S$ overestimate specialisation to a

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much higher degree for Tortricidae and Agromyzidae than Tephritidae (11% and 14% species misclassified as specialists for Tephritidae vs. 39% and 30% for Tortricidae and 50% and 43% for Agromyzidae – Table S2) when compared to DSI. This shows that the exclusion of phylogenetic relatedness and/or resource commonness can hide important differences in resource use by different lineages of herbivores.

The variation in specialisation within species ranges and its scale dependency was proposed by Fox & Morrow (1981) and has been widely applied to date. According to our framework, in the test data local specialisation was unexpectedly scarce, being restricted to only two related species in the same locality. However, these two species correspond precisely to the expectation for a local specialist: they use several unrelated hosts in their entire range, but feed selectively on a single genus in one locality. Thus, on the local scale our criterion for specialisation is much stricter than simpler criteria in common usage.

Perspectives and conclusions

The incorporation of relatedness among host plants to assess and contrast host ranges was an important step in the comprehension of insect–plant associations. However, only recently this is becoming feasible with the gradual availability of phylogenies for major groups and the development of metrics for relatedness of subsets of a species pool (Webb *et al.* 2002; Weiblen *et al.* 2006). By integrating this metric with statistical models that take plant commonness (geographical spread and/or local abundance) and spatial co-occurrence into account, we were able to define and measure specialisation in phytophagous insect communities in a more rigorous and theoretically sounder way. We should note that here we use DSI as a comparison standard, not because we consider it an intrinsically "true" measure of specialisation, but because it is more explicit and comprehensive, with higher power to detect differences and hence more useful than other measures of specialisation.

We designed different models to reflect distinct combinations of evolutionary and ecological processes. Comparison of these models reveals that, with the incorporation of phylogenetic information, the concept of specialisation becomes much more effective than the simple count of host species to differentiate degrees of specialisation. Most importantly, it is clear that measuring either relatedness or the number of host species without reference to the set of potentially available resources has a pronounced effect on the measurement of specialisation. Most previous endeavours to incorporate phylogenetic relatedness in measures of specialisation did not take the resource pool into account (e.g. Morse & Farrell 2005; Pellissier *et al.* 2012), even when, for instance, independent data for plant communities were available.

A similar problem besets studies that investigate the role of phylogeny in structuring interactions without direct reference to specialisation (Ives & Godfray 2006; Rafferty & Ives 2013). Using linear mixed models with a phylogenetic component in the errors, these approaches disregard species abundances and co-occurrences; moreover, they cannot be employed when a consumer phylogeny is not available. In these cases, our framework could improve the comprehension of the processes at work, and is likely to change the inferred patterns of specialisation. An improvement is also to be expected in studies that measured specialisation by the number of species or other taxonomic levels (e.g. Aizen *et al.* 2012), as in this case both sources of bias in the estimation of specialisation apply. Even when the resource pool was included in phylogenetic measures of specialisation (Weiblen *et al.* 2006; Vamosi *et al.* 2014), the exclusion of resource commonness and of frequency of consumers may still affect the estimates of specialisation, as we showed in our data set. Likewise, previous attempts to consider species frequencies and abundances (Novotny *et al.* 2004) did exclude relatedness and in turn did not compare the observed patterns with random expectations.

Other specialisation metrics that do not measure relatedness have been applied in interaction network studies, such as Blüthgen *et al.*'s (2006) d'. In this case, local resource availability is taken into account; nonetheless, these methods use only frequencies of interaction within the network, regardless of sampling intensity, resource commonness or the absolute number of observations of the interactions. As shown in the analysis of our data set, the exclusion of these factors can overestimate specialisation of scarce species. By using the number of interactions of each species as a surrogate for its availability, host usage cannot be decoupled from abundance; in this sense, the metric of Blüthgen *et al.* (2006) deviates from the classical concepts of resource availability and selection (Colwell & Futuyma 1971; Manly *et al.* 1993). The absence of relationship between DSI and d' (Fig. 2c), fuels the current discussion on the inadequacy of using interaction frequencies as a proxy for abundance (Vizentin-Bugoni *et al.* 2014), reinforced by the fact that d' also showed a very weak relationship with WRI (Fig. S3, $\rho = 0.20$, $P = 0.02$). As d' and WRI both disregard phylogenetic relatedness of resources and use a similar rationale for resource diversity and availability, the key difference between them is the use of interaction frequencies for d' and resource commonness for WRI.

Our framework has the advantage of being quantitative, because DSI is a continuous measure. Since, by consensus, specialists and generalists are extremes of a specialisation gradient, this might obviate the need for classifying species into specialisation categories. All comparisons of specialisation both among and within species would then be quantitative, with a continuous metric applied to spatial and temporal gradients or different ecological settings. Nevertheless, the specialist/generalist dichotomy continues to thrive in the recent literature, both theoretical and empirical (McCann 2012; Börschig *et al.* 2013). In the Web of Science© database, a combined search for specialists and generalists returned over 1,000 publications. Given that the dichotomy will continue to be used, a firmer basis for this classification is certainly useful.

As our specialisation measure is compared to a null model, one can place observed values in the tails of the corresponding distribution. The cut-off points we employ to assign species to specialisation categories follow the common practice of using a normal distribution for reference. Although it is as arbitrary as any division of a continuous variable into discrete classes, this approach has the advantage of being familiar and replicable, improving the quality of comparisons within and among

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studies. Other alternative statistics are applicable, for instance the proportion of the null distribution exceeding the observed value for each species. We chose to use a Z-score to maintain comparability with currently used phylogenetic structure metrics (NRI and similar). Moreover, values of DSI measured as a proportion are forced to vary between 0 (all null model iterations above observed MPD) and 1 (all null model iterations below observed MPD). These limits are dependent on the number of iterations used, and for our data both measures are nearly identical for species whose DSI measured as a proportion is between 0 and 1 (Fig. S4). Furthermore, when the proportion of values above and below the observed MPD are used to classify consumers into specialists or generalists, only three species out of the 168 change category when compared with a Z-score. Even when considering only the 127 species that are not singletons and hence for which a classification is possible, only 2.4% of the species have mismatched classifications among both metrics.

By defining three categories of specialisation, we highlight that so-called generalists comprise two very different kinds of resource use: indiscriminate use, which is the usual meaning of generalist, and over-dispersed use of resources. This disambiguation opens up the possibility of investigating which processes cause each of these kinds of generalism. Also, by categorising undersampled species as indiscriminate feeders we improve the assessment of specialisation, given that most studies would classify these species as highly specialised. A further advancement in this direction would require a quantitative method to sort indiscriminate feeders from undersampled species, in a similar manner to the criterion proposed by Chazdon *et al.* (2011) for a classification in a simple two-habitat situation.

The importance of adequate regional species pools to test for phylogenetic structure has already been advocated in the context of community structure (Cavender-Bares *et al.* 2009), and this is also crucial for specialisation measures, such as DSI_A and DSI, that evaluate usage with respect to the set of available resources (see also Forister *et al.* 2012). To measure specialisation in interactive assemblages, the spatial scale, extent of the study, and the effective range of usable resources are all choices to be considered carefully, preferably when designing the study.

Local specialisation has become a popular concept, very often unrelated from the scale dependency of host–plant selection as proposed by Fox & Morrow (1981). Many studies discussing local specialisation neither test for specialisation at different spatial scales, nor consider differences in availability of host-plants among localities as an alternative explanation for local patterns of host use. The framework we propose has an explicit measure of local specialisation accounting for these neglected factors, allowing to test for scale dependency in specialisation. Under these explicit restrictions, local specialisation was shown to be very unusual in our data set. It should now be tested whether this is an idiosyncrasy of our system or if it is a broader pattern that remains hidden by ignoring differences in host availability, or by other concepts of specialisation used in most studies.

Generalists in the proposed sense were also rare in our test study. Significant phylogenetic overdispersion of used resources could have different causes. If spatial differentiation of host use is involved, we might expect regional generalists to be local specialists, which did not occur in our data set; if so, they might be an extreme case of a geographic mosaic of interactions (Thompson 2005). However, for a more economical explanation, generalists are born candidates to be investigated whether they are not in fact unresolved cryptic species or host races. On a finer scale, populations may comprise individuals whose use of distinct resources is due to phylopatry or idiosyncratic preference (e. g. grasshoppers, Ben Halima *et al.* 1985). Larger-bodied organisms often combine unrelated resources to complement nutritional requirements or to avoid intoxication, and these could appear as generalists in community surveys (Belovsky 1984).

Our approach can be applied more broadly, as for instance, to any other kind of bipartite interaction, such as mutualistic networks of pollination and seed dispersal (Aizen *et al.* 2012). As long as there is knowledge about local resource communities and interaction networks, allied with a phylogeny for the resource group, this specialisation metric can be applied. When resources and consumers, or mutualistic partners, are two sets of species, specialisation becomes a two-way phenomenon that can be evaluated for any set with regard to the other (Bascompte *et al.* 2006).

Restrictions in other aspects of the resource range can be equally measured with this approach, by using different metrics instead of phylogenetic relatedness. For example, diversity of any functional trait (as in Junker *et al.* 2013) can be measured and compared to the null expectation. It can also be applied for specialisation beyond the interaction context, if the resources or even habitats for which specialisation is measured can be characterised and classified in a hierarchical manner; for instance, the habitats in a landscape whose attributes can be used to produce a similarity matrix and a cluster analysis. Habitat selection studies would benefit from this approach when there are many habitat classes to be selected, as the problem is simplified and tests are more powerful by habitat clustering instead of proportion of habitats available (Davies *et al.* 2004).

Other methodological advances in this framework are possible. Ideally, a highly resolved phylogeny with stem lengths is recommended. However, given that such trees are hard to produce, an important further step would incorporate uncertainties in phylogenetic knowledge into null models (Huelsenbeck *et al.* 2000). Another fairly straightforward addition to these models would be the detectability of both consumers and resources in a hierarchical model when measuring co-occurrences, instead of assuming there is no sampling error (Dorazio *et al.* 2006). Further on, a logical next step is the derivation of a specialisation measure for the community level, integrating the information from co-occurring species into a single measure that can be compared among communities and related to biotic and abiotic variables in macroecological or metacommunity studies. By highlighting neglected aspects in the measurement of ecological specialisation and proposing an integrated framework to apply them, we expect that our contribution will enable a more rigorous application of one of the most important ecological concepts in empirical and theoretical studies.

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AUTHORSHIP

L.R.J. and T.M.L. conceived the paper; T.M.L., P.I.P. and M.A.N. collected data; all authors designed the null models, L.R.J. performed analyses; L.R.J. and T.M.L. wrote the manuscript.

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