



Disentangling an entangled bank: using network theory to understand interactions in plant communities

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Disentangling an entangled bank: using network theory to understand interactions in plant communities



Photo by Ray Blick

Ray A.J. Blick

Thesis submitted for the degree of Doctor of Philosophy

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School of Biological, Earth and Environmental Sciences
University of New South Wales

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**THE UNIVERSITY OF NEW SOUTH WALES
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Network analysis can map interactions between entities to reveal complex associations between objects, people or even financial decisions. Recently network theory has been applied to ecological networks, including interactions between plants that live in the canopy of other trees (e.g. mistletoes or vines). In this thesis, I explore plant-plant interactions in greater detail and I test for the first time, a predictive approach that maps unique biological traits across species interactions.

In chapter two I used a novel predictive approach to investigate the topology of a mistletoe-host network and evaluate leaf trait similarities between Lauranthaceous mistletoes and host trees. Results showed support for negative co-occurrence patterns, web specialisation and strong links between species pairs. However, the deterministic model showed that the observed network topology could not predict network interactions when they were considered to be unique associations in the community.

Network analysis has revealed similarities between mistletoe- and vine-host interactions. In this thesis I investigated the role of chance in structuring these interactions. Results showed that mistletoes and vines use host trees in very different ways even though network topology suggest similarities. In chapter three I show that the dispersion of individual mistletoes (i.e., clumping of mistletoe on certain trees) was not dependent on tree availability; however and perhaps more importantly, I show in chapter five that coincidental associations between vines and trees are sufficient to generate similar network patterns to that found in mistletoes. Each of these studies explored untested hypotheses regarding the architecture of plant-plant interactions.

In chapter six I turned to plant-animal interactions to test whether temporal changes influenced network topology. That is, I quantified variability among species interactions and I tested the role of species turnover in structuring a frugivore network spanning six sequential years. Results showed that frugivore interactions (i) changed from year-to-year, (ii) showed inconsistent patterns of nestedness, and (iii) novel interactions occurred even after six years of data collection.

In conclusion, a modified approach to the traditional randomisation procedure has allowed me to test explicit factors that determine interactions in plant communities. Network analysis is a useful measure of structure that can depict structural differences among arboreal plant communities in Australian ecosystems.

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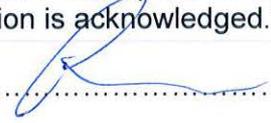
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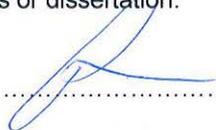
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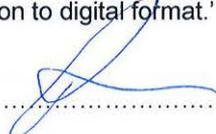
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During my time at UNSW I was given many opportunities to experience the less advertised side of science. For example, science communication in social media was an area of science I had not considered before. I demonstrated six different ecology courses, presented at four internationally recognised conferences, published three peer-reviewed articles and in-turn reviewed 14 articles from eight internationally recognised journals. I believe these activities are important for anyone considering a scientific career and I urge students to pursue these opportunities when they arise.

Two researchers shared their hard earned data. Thank you Rachael Gallagher and K.C. Burns, I am extremely grateful to have this opportunity. Such a phenomenal amount of work has been shared by these two researchers with no expectation in return. Thank you so much for your help.

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“The beautiful thing about learning is that no one can take it away from you”

Abstract

Network analysis maps interactions between entities to reveal complex associations between objects, people or even financial decisions. Network theory has been applied in a wide variety of disciplines to map city infrastructure, detect neural pathways and determine the organisation of social groups. Recently network theory has been applied to ecological networks, including interactions between plants that live in the canopy of other trees (e.g. mistletoes or vines). In this thesis, I explore plant-plant interactions in greater detail and I develop and test for the first time, a predictive approach that maps unique biological traits across species interactions.

In chapter two I used a novel predictive approach to investigate the topology of a mistletoe-host network and evaluate leaf trait similarities between Lauranthaceous mistletoes and host trees. Results showed support for negative co-occurrence patterns, web specialisation and strong links between species pairs. However, the deterministic model showed that the observed network topology could not predict network interactions when they were considered to be unique associations in the community.

Network analysis has revealed similarities between mistletoe- and vine-host interactions. In this thesis I investigated the role of chance in structuring these interactions. Results showed that mistletoes and vines use host trees in very different ways even though network topology suggest similarities. In chapter three I showed that the dispersion of individual mistletoes (i.e., clumping of mistletoe on certain trees) was not dependent on tree availability; however and perhaps more importantly, I show in chapter five that coincidental associations between vines and trees are sufficient to

generate similar network patterns to those found in mistletoes. Each of these studies explored untested hypotheses regarding the architecture of plant-plant interactions. In chapter six I turned to plant-animal interactions to test whether temporal changes had a similar effect on network topology. That is, I quantified variability among species interactions and I tested the role of species turnover in structuring a frugivore network spanning six sequential years in a wildlife reserve in New Zealand. Results showed that frugivore interactions (i) changed from year-to-year, (ii) showed inconsistent patterns of nestedness, and (iii) novel interactions occurred even after six years of data collection.

In conclusion, a modified approach to the traditional randomisation procedure has allowed me to test explicit factors that determine interactions in plant communities. A wide variety of ecological processes may generate different network properties, such as disturbance and fragmentation; however the results from this thesis show that these factors can be identified and tested using network theory. I suggest that network analysis is a useful measure of structure that can depict differences among arboreal plant communities in Australian ecosystems.

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Chapter One

Introduction to Ecological Network Analysis

Understanding and describing species' interactions is a major focus for community ecologists (van der Maarel 2009 and references within). Over recent years, network theory has been applied to ecological and environmental sciences, providing a framework for investigating complex interactions. An ecological network is depicted as 'nodes' (species) and 'arcs' (interactions) which summarise the connectivity between organisms (Figure 1). However, identifying the processes responsible for different patterns of connectivity is a major challenge (Alcantara et al. 2012, Florent et al. 2012, Nuismer et al. 2013, Pires et al. 2013, Roy et al. 2012).

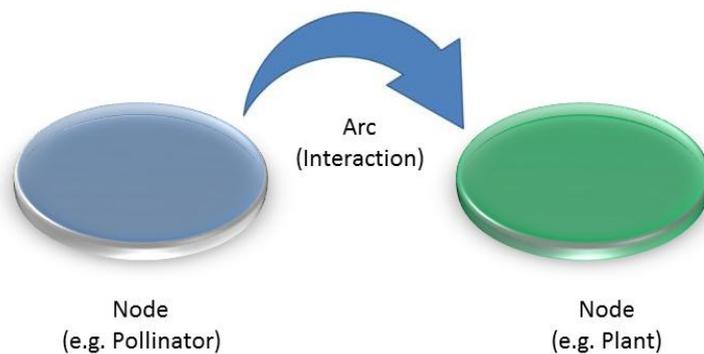


Figure 1 A simple network depicting two organisms (nodes) observed interacting together in a single community (arc).

In this thesis, I quantify a range of factors underpinning mutualistic (plant-visitor interactions) and antagonistic networks (mistletoe- and vine-host interactions) that are predicted to explain complex ecological patterns. This introduction is presented in two parts: first, I provide a synthesis of ecological networks that follows well-known concepts, including the individualistic and organismal concepts developed during the early twentieth century, to Diamond's (1975) controversial assembly rules and the advent of null models in ecology. Second, I develop questions and predictions specific to this thesis (see thesis outline).

Early research focussing on the structure of plant communities was centred around fixed laws and universal principles in structure (Tansley 1904 and 1913, Clements 1909). Clements (1916) proposed that vegetation acted as “an organic entity” with “a climax formation” (organism concept), while Gleason (1917) stated that vegetation was the maintenance of individuals (individualistic concept) such that “every species of plant is a law unto itself” (Gleason 1926). Both perspectives were challenged (Tansley 1920, 1935) and debate ensued over successional processes (Gleason 1926, Godwin 1929, Phillips 1934, Clements 1936). Support has shifted between the Clementsian and Gleasonian perspective during the last century (Watt 1947, 1964, van der Maarel 1996), however debate over which mechanisms govern community organisation are ongoing (Keddy and Weiher 1999, Hubbell 2001, Leibold et al. 2004, Hausdorf and Hennig 2007).

At the heart of Gleasonian and Clementsian perspectives is the dichotomy between two schools of thought described by Hubbell (2001) as the *niche-assembly perspective* and *dispersal-assembly perspective*. The niche-assembly perspective states that there is strict organisation among species in the exploitation of available resources, while the dispersal-assembly perspective states that an ecological community is largely open, with regular changes from random speciation, drift and extinction events. MacArthur and Wilson’s (1960) ‘Theory of Island Biogeography’ is an example of the dispersal-assembly perspective (see Hubbell 2001). Significantly, it was MacArthur and Wilson’s research that introduced a new idea to ecology: that chance could play a viable and predictable role in assembling ecological communities. Subsequently, many branches of

ecology have stemmed from these ideas, including co-occurrence indices and null models.

The question of whether species co-occur randomly, or according to assembly rules, is an extremely controversial topic in ecology (Strong 1984). This debate was initiated by Diamond (1975), who stated that a set of assembly rules (interspecific competition) could determine the co-occurrence of birds on the Bismarck Archipelago. Diamond's model was later challenged by Connor and Simberloff (1979), who sought a null hypothesis to explain the divergence of observed patterns from a random distribution (by using Monte Carlo simulations). Null models and pattern detection have remained an important area of research (Manly 1995, Gotelli and Graves 1996, Sanderson et al. 1998, Gotelli and McCabe 2001, Gotelli and Rhode 2002, Gotelli 2000, 2001, Gotelli and Entsminger 2003, Ulrich 2004, Ulrich and Gotelli 2013), and form the basis for hypothesis testing in ecological network analysis.

A common approach to testing species co-occurrence patterns is to arrange the observed data in a binary matrix where rows represent species and columns represent sites. An index is then calculated and compared statistically with the randomly generated values using a standardised z score. The result shows whether the observed community has significantly more, or fewer, co-occurring species than would be expected by chance. For example, C scores are used to quantify the degree of segregation between species pairs (Stone and Roberts 1990). C scores are calculated for each pair of species as $(R_i - S)(R_j - S)$ where R_i and R_j are the matrix row totals for species i and j , and S is the number of sites in which both

species occur. An observed C score greater than expected by chance indicates the presence of assembly rules such as interspecific competition (Diamond 1975).

A primary assumption of the null model approach is that a community with segregated species pairs that is significantly different to a randomised pattern can be considered deterministic (Stone and Roberts 1990). That is, an index significantly greater (or less, in the case of nestedness; see Atmar and Patterson 1986) than predicted by chance would indicate that an ecological community is determined by an unspecified ecological process (Connor and Simberloff 1979). Previously, 'segregation' (pairs of species that are never found together) was inferred to mean interspecific competition (Diamond 1975), but it is now widely agreed that segregation could be determined from ecological drift, environmental filtering, spatial turnover, turnover-independent species sorting, and stochastic processes (Hubbell 2001, Holyoak et al. 2005). However, real ecological communities are undoubtedly assembled by multiple rules of coexistence (see Chase et al. 2005) and deliberation over an appropriate theoretical model continues to foster interesting research (Veech 2013).

Null models are not the only avenue for determining community structure or hypothesis testing. For example, the neutral perspective of the metacommunity concept (spatially separated communities linked by dispersal) forms a basis for comparing ecological processes such as patch dynamics, mass effects and species sorting (Holyoak et al. 2005). Here the neutral model predicts that community composition changes with distance, not environment, while the species sorting perspective predicts that community composition changes with environment, not distance (Chase et al. 2005). Current empirical evidence suggests that community

composition is not explained by distance alone (Terburgh et al. 1996, Tuomisto et al. 2003, Condit et al. 2002, Leibold and Mikkelsen 2002) further emphasizing the uncertainty in community-level co-occurrence patterns.

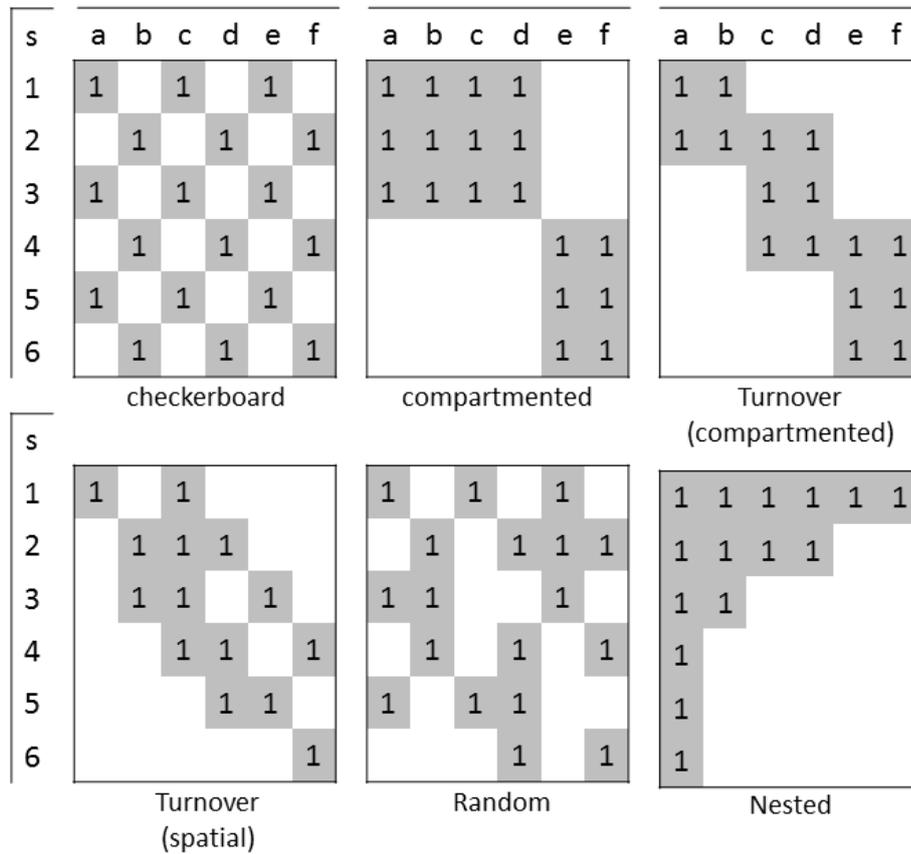


Figure 1 Six hypothetical species co-occurrence patterns, after Ulrich and Gotelli (2013). Numbers 1-6 represent species and letters a-f represent sites. Shaded cell entries (1's) represent presence of species *i* at site *j*. For alternative combinations see Ulrich and Gotelli (2013)

Null model analysis cannot identify specific processes pertaining to environment or distance as described above. However, null models and neutrality are different concepts. An important distinction between these two concepts is that they examine different spatial scales. A null model assumes that 1) each species assemblage (see Figure 1) has a uniform distance from other assemblages, 2) all species have an equal likelihood of getting to available sites, and 3) no species are forbidden from colonising each site. In contrast, the theory of neutrality predicts that stochasticity occurs at a regional scale (Hubbell 2001, Chase et al. 2005). That is, dispersal ability determines the competitive species pool at regional scales, and environmental filtering or competitive displacement determines the final arrangement of species at local scales (Hubbell 2001). Recent studies have found support for local and regional processes (Fenton and Bergeron 2013, Laliberte et al. 2013, Siefert et al. 2013), while others have shown that regional effects can mask processes operating at much finer scales (Heino and Grorocs 2013, Marquez and Kolasa 2013). However, these results do not negate the benefit of using a null model approach. Rather, a null model provides a platform for testing predictions of neutrality by including spatial distribution and connectivity between species assemblages. Adapting null models to test neutral processes in resource selection forms a major component of this thesis; I return to this concept in a later section of the introduction (see Thesis outline).

A central theme of the work discussed thus far has involved species from the same trophic level (e.g. plants). Recent effort has applied null model analysis and ecological indices to determine the connectivity between two trophic levels using ecological network analysis (Bascompte et al. 2003, Prolux et al. 2005, Vázquez

et al. 2009, Ings et al. 2009, Dormann et al. 2009, Blüthgen 2010). Analysis of ecological networks containing two trophic levels (i.e. bipartite analysis) is analytically equivalent to modelling species co-occurrence; such that columns (previously sites) are replaced by species from a higher trophic level such as birds, insects or herbivores. However, the two approaches are fundamentally different for two reasons. First, cell entries quantify observed interactions, rather than presence/absence from each site. Second, the marginal sum of observed interactions and the number of connections between other players in the network provides a quantitative metric describing the function of each species in the community.

Ecological network analysis has progressed rapidly in the last decade (Ings et al. 2009) due to conceptual development in social sciences (de Nooy et al. 2005, McFarland et al. 2010), null models (Gotelli 2000), shuffling algorithms (Ulrich and Gotelli 2007a, 2007b, Ulrich et al. 2009), ecological indices (Atmar and Patterson 1986, Stone and Roberts 1990) and freely available software (de Nooy et al. 2005, McFarland et al. 2010, Dormann et al. 2008, 2009, 2011, McFarland et al. 2010, Gotelli and Entsminger 2012). However, the idea that species could be summarised by connections in the community was well established prior to the twenty first century (Lindeman 1942, Odum 1968, Pimm 1979, Fretwell 1987). Even so, many original questions remain unanswered. For example, Pimm (1979) asked: “should model systems be organized into *compartments* of species characterized by strong interactions within compartments, but weak interactions among the compartments?” Three decades later, Stuart Pimm and colleagues concluded in the journal *Nature* that “Our knowledge of the structure of

ecological networks is still incomplete in important areas that include compartments and reciprocal specialization” (Montoya et al. 2006).

Compartmentalization has since been identified in many ecological networks (Fortuna et al. 2010, Guimerá et al. 2010, Stouffer and Bascompte 2011, González et al. 2012). Even though a mechanistic explanation remains largely unresolved, it is predicted that compartmentalisation occurs when different taxa are included in the analysis and affected by the spatial scale of the research (Montoya et al. 2006).

Despite its value to ecology, network analysis is not an analytical tool restricted to biological interactions (Girvan and Newman 2002). Networks have been examined in molecular sciences (Jeong et al. 2000, Bray 2003), social groups (de Nooy et al. 2005, Kossinets 2006), and information technology (Albert et al. 1999). Importantly, networks share four properties, including (1) the “small world effect” (also called “scale free”), meaning that the average distance between nodes is short (Albert et al. 1999), (2) right skewed degree distributions, meaning that few nodes are highly connected, (3) network transitivity, meaning that two nodes have a higher probability of being connected if they share an interaction with a third party (de Nooy et al. 2005), and (4) compartmentalisation, meaning that a network contains sub-groups with dense connections and weak between-compartment interactions (described above; Girvan and Newman 2002). For example, Jeong et al. (2000) showed that metabolic networks of 43 organisms share the same topological scaling properties and organization as non-biological systems. Thus, network analysis has been rigorously tested across multiple disciplines and provides a quantitative method for evaluating complex organisational principles of different ecosystems.

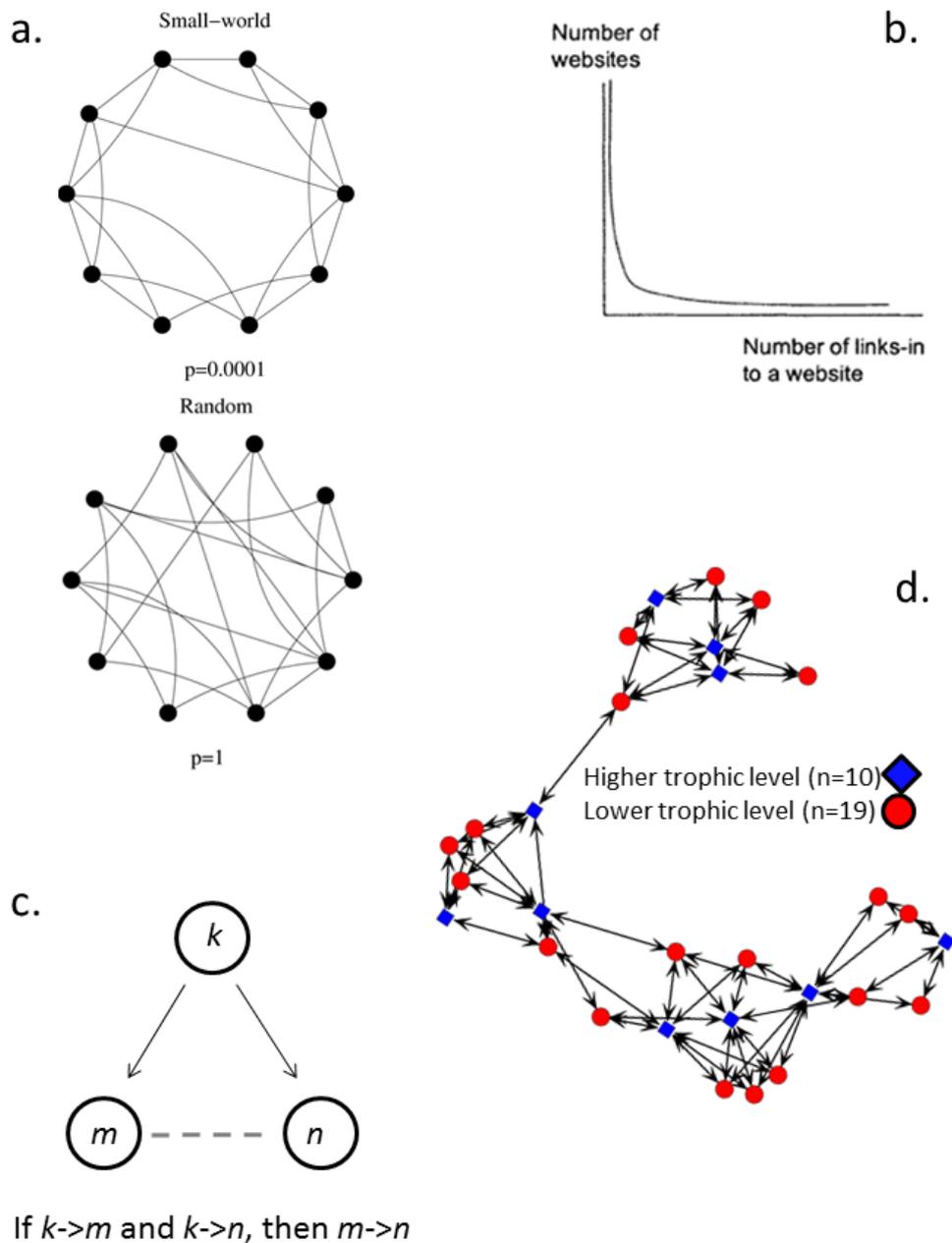


Figure 2 Four common network properties, including (a) small world network after the Institute for Complex Systems Simulation, (b) right skewed degree distribution, (c) network transitivity and (d) compartmentalisation. Circles represent nodes, lines and arrows represent connectivity between nodes.

Many ecological networks are nested, including plant-pollinator (Bascompte et al. 2003, Stang et al. 2006), epiphyte-tree (Blick and Burns 2009, Piazzon et al. 2011, Silva et al. 2011), clownfish-anemone (Ollerton et al., 2007), ant-plant (Guimarães et al. 2006) and fruit-frugivore mutualisms (Lázaro et al., 2005). A network is said to be nested when a core set of generalist species interact together, while maintaining interactions with many rare species (singletons). Species with few connections in the network form a perfect subset of highly connected species (Blüthgen 2010); similar to a Russian doll; where a smaller replica is found nested within the larger doll, and so on. Ecological networks have similar properties, such as skewed interactions and small world connections (Sole and Montoya 2001). However, not all ecological networks are the same, and their differences can describe ecological and evolutionary patterns (Proulx et al. 2005). For example, topological structure may vary according to (1) the number of forbidden links (Stang et al. 2009, Olesen et al. 2011) or (2) interaction intimacy (Pires and Guimarães 2013). In addition, a wide range of ecological indices may occur simultaneously, such as nested subsets within compartmentalised interactions (Lewinsohn et al. 2006, Fortuna et al. 2010).

Network analysis has several limitations that are well known and tested. First, pattern detection using a null model approach (i.e. the difference between observed and randomised interactions) does not identify specific mechanisms. Grounding network theory in biological and theoretical hypotheses, in association with appropriate indices, are required to explain community structure (Ulrich and Gotelli 2013). Second, ecological networks are considered a ‘snap-shot’ in space and time. That is, interactions between higher and lower trophic levels are pooled

in an interaction network, while temporal and spatial scales occurring on daily, seasonal and annual timescales are sometimes ignored (but see Baldock et al. 2011). Third, ecological networks are derived from observed interactions while alternative resources are overlooked (e.g. fruit availability). One solution is to ‘weight’ the likelihood that two species would encounter each other using abundance (Morales and Vazquez 2008). However resource-mediated selection between individuals has until recently received little attention (but see Pires et al. 2010). In addition, network analysis is susceptible to other limitations common in ecology, such as sampling effort (Martinez et al. 1999, Nielsen and Bascompte 2007). Despite these limitations network analysis is the prevailing analytical tool that describes multi-species connectivity in a community.

The development and application of network analysis has required many advances over the last century. Initial questions pertaining to the organisation of communities, ecosystems and food webs have been developed and improved with more complex and quantitative methods (reviewed by Ings et al. 2009).

Ecological network analysis has grown in popularity with literature expanding on an exponential scale each year (see Figure 1 of Ings et al. 2009). Individual interactions, described by Jeong et al. (2000) as ‘network motifs’, are now explored with increasing importance (Ings et al. 2009, Pires et al. 2010, Pires and Guimarães 2013). Ecological network analysis is still in its infancy; however it is primed for answering long standing questions in ecology and provides new avenues for assisting conservation and monitoring programs in the future.

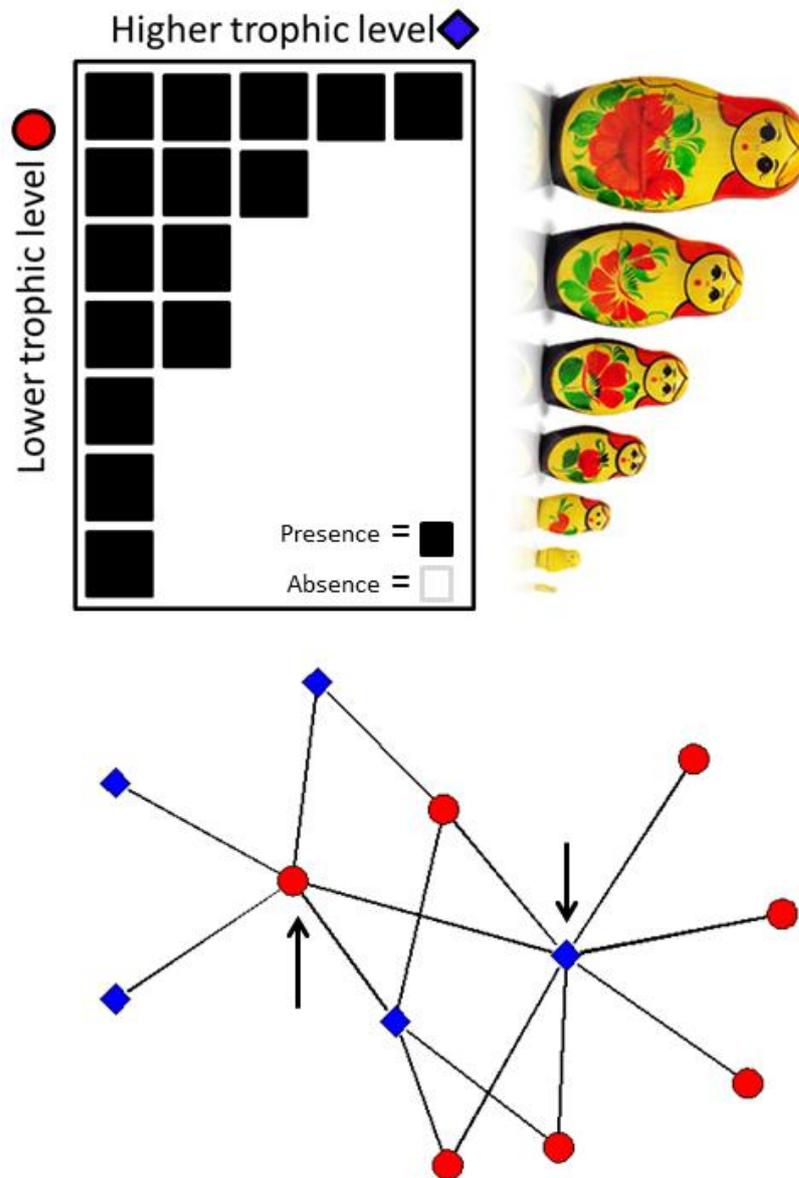


Figure 3 Hypothetical data depicting nestedness. Squares (closed fill) represent interactions between two species. A re-ordered matrix contains a core set of interacting generalist species that supports interactions with rare species. A nested matrix can be depicted as Russian dolls and examined as a social network. Interactions between the higher trophic level (blue diamond) and lower trophic level (red circles) are represented by lines. Arrows indicate core species (generated using social network analysis package ‘SNA’ in R).

From a conservation perspective, ecological networks are of high importance because they provide a framework for predicting what might happen to an ecological community during habitat fragmentation or species extinction events (Dunne et al. 2002, Thébault and Fontaine 2010, Piazzon et al. 2011, Burkle and Knight 2012, James et al. 2012). In addition, patterns of connectivity may change in response to invasive flora or fauna. Empirical evidence supporting these hypotheses is limited because long-term studies are needed and repeated sampling techniques need to be trialled. A major concern here is that short survey times or infrequent visits to a study site could generate an incomplete (sub-) network providing incorrect results (Type I error). Determining what effect sampling effort, repeated sampling, and temporal scaling has on network connectivity is important for the application of network analysis to monitoring multi-species assemblages.

Plant-plant interactions

In recent years, ecological networks have been applied to plant-plant interactions, including mistletoes, epiphytes and vines (Burns 2007, Blick and Burns 2009, Silva et al. 2010, Blick et al. 2011, Sfair et al. 2011, Piazzon et al. 2011, Genini et al. 2012, Martos et al. 2012). Current research suggests that different plant guilds have different network properties (as displayed by the mistletoes, lianas and epiphytes described in Blick and Burns, 2009). However, results have varied among researchers and ecosystems. For example, vines growing in neotropical vegetation are considered nested (Sfair et al. 2011), while temperate rainforest lianas of New Zealand are not (Blick and Burns 2009, 2011). Mistletoes and epiphytes are more consistent, showing modularity and nestedness respectively

(Piazzon et al. 2011, Silva et al. 2011, Genini et al. 2012). It is hypothesised that plant-plant interactions have different network properties (topology) because each guild has evolved different growth strategies (i.e. parasitism and commensalism). For example, Burns (2007) hypothesised that nested topology of epiphyte-host interactions were determined by the colonisation of clump forming species and the successional accumulation of species less tolerant to water and nutrient limitations in an unoccupied tree canopy. Similarly, Blick and Burns (2011) hypothesised that a checkerboard distribution (negative co-occurrence) was determined by differences in environmental conditions and coincidental pairwise associations in two areas of a single wildlife reserve. Both hypotheses remain untested; however network analysis has revealed differences in organisational complexity requiring further investigation.

Plant-plant interaction networks are tested using a standard null model procedure (i.e., are interactions different from random?). However, several assumptions limit the interpretation of plant-plant interaction networks. First, plant-plant networks are generated using data that are often focussed on the arboreal plant species (mistletoes, lianes or epiphytes), potentially under-sampling the host (tree) components of the network. Second, plant-plant networks are often generated with inaccurate or biased abundance measures (i.e. only trees carrying mistletoes or vines are included). Third, all plant-plant networks analysed to-date are generated without spatially explicit data (i.e. all interactions are assumed possible). These three points are important because they lead to an unreliable measure of dominance in the landscape, and ignore a neutral explanation for network structure (i.e. stochastic regional processes generate mismatched species-pairs).

Individual trees can be considered ‘network motifs’ (see Jeong et al. 2000) of arboreal plant networks and the most important factor uniting plant-plant interactions. Surprisingly tree size, availability, and dominance are given little consideration even though arboreal plants have different growth strategies and host tree requirements. For example, vines and mistletoes have revealed similar network properties (checkerboard distribution; Blick and Burns 2009; but see Sfair et al. 2011). However, both functional groups have evolved different strategies of host use. Vines have evolved a wide variety of climbing strategies such as hooks that grapple branches (Putz 1984, Balfour & Bond 1993), while mistletoes have evolved a parasitic growth strategy restricting infection prevalence to a range of host species. Preferential selection of vines for functional attributes (e.g. bark type or stem diameter; Campbell & Newbery 1993, DeWalt et al. 2006, Burnham 2002, Malizia et al. 2010) could generate similar properties to mistletoe–host interactions (Blick and Burns 2009). However, an equally plausible hypothesis follows that vines have opportunistic host selection within spatially explicit habitats (i.e., interactions between vines and their hosts are random; DeWalt et al. 2006). Under this hypothesis, opportunistic interactions may appear deterministic as an artefact of the method. Conflicting hypotheses raise additional questions surrounding a single trophic level in plant-plant networks (i.e., host trees); i) does tree diversity increase the number of network connections? ii) Does tree availability regulate interaction frequency? iii) Does species turnover in one trophic level influence network connectivity? These questions are the focus of this thesis.

Plant-plant interactions: data collection

Mistletoe-host interactions were recorded at Fowlers Gap Research Station (FGRS) in the semi-arid zone of New South Wales, Australia (Figure 4). A detailed description of the climate can be found in chapters two and three. FGRS lies on the border of New South Wales and South Australia approximately 1100 km from Sydney (Figure 4). Historically, FGRS was operated as farmland and extensive clear-cuts were made to large stands of mulga (*Acacia anuera*). In this area the pale leaf mistletoe (*Amyema maidenii*) is only found on branches of mulga growing in remnant patches scattered across the landscape. A major concern here is that the wild goat population has increased in size and now restricting the regrowth of mulga (pers. comm., K. Leggett). Understanding the role of different tree species in the landscape is important for the long-term persistence of mistletoe occurring at FGRS and semi-arid Australia. FGRS was considered appropriate for testing the relationship between mistletoes and host trees because (1) four mistletoe species have been identified in a small area (25km²) in which all trees could be enumerated on foot, and (2) the number of mistletoes growing in each tree could be accurately counted as the number of historia were visible from the ground.

The data set containing vine-host interactions was kindly provided by Dr Rachael Gallagher. The data consist of interactions between vines and host trees from nine littoral rainforests along the east coast of New South Wales, Australia. These data do not provide a continuous network of interactions (e.g. data were collected in plots) as was collected at FGRS. Rather, the data provide complementary information regarding geographic turnover and host availability.



Figure 4 Map of Australia showing the location of Fowlers Gap Research Station.
Image generated ArcMap 10.1.

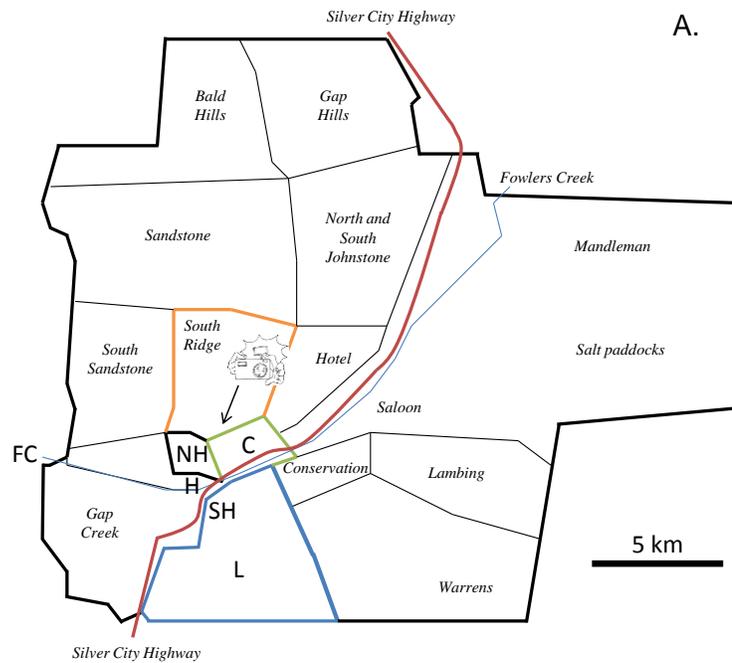


Figure 5 Fowlers Gap Research Station (FGRS) ~100 kilometres north of Broken Hill, NSW Australia. FGRS covers approximately 400 square kilometres and is subdivided by paddocks. ‘Connors’ (C) and ‘Lake’ (L) paddocks are visible in this photo. The Silver City Highway (SH) traverses the landscape via the homestead lodging (H) and is surrounded by dense vegetation growing along the bank of ephemeral creeks.

Plant-animal interactions

In the final section of this thesis I turn to plant-animal interactions. Plant-animal interactions are commonly investigated using ecological networks around the world. Many significant advances in methodology and analysis have been achieved over the past decade using plant-pollinator interaction networks (Bascompte et al. 2003). While fewer studies have focused on plant-frugivore networks, they too have been successfully depicted as ecological networks (Reid and Armesto 2011). A key advantage of switching taxonomic groups (from plants to animals) is that it allowed an explicit test of temporal scaling and sampling efficiency.

Currently it is poorly known how network interactions change with time. For example, do generalists remain common, or might we consider interactions rare in some seasons of the year? In this thesis, I use network analysis to examine long-term trends (six years) of plant-frugivore interactions in a wildlife reserve (data collected by Dr K.C. Burns) approximately 2 km south of Wellington's Central Business District, New Zealand. I test the role of sampling efficiency, temporal scaling and the application of network analysis in monitoring introduced avian fauna.

Thesis questions

The aim of this thesis was to evaluate ecological networks, discuss their limitations, and advance the understanding of plant-plant interactions. To achieve this goal, I evaluated four areas of research under the network paradigm. The fourth theme of research is comprised of three primary questions which address the stability of network interactions.

1) Biological traits:

- Can biological traits predict species interactions in ecological networks?

2) Trophic level asymmetry:

- Can one trophic level (guild) influence network topology? Or more specifically, does the distribution of individuals affect null model outcomes (prevalence in parasite interactions)?

3) Stochastic processes:

- Are opportunistic interactions considered deterministic at some, but not all spatial scales?

4) Temporal scaling and sample design:

- Is the stability of an ecological network transient on annual scales, owing to a rise and loss of species interactions?
- Are common species eventually observed and recorded visiting rare plant species on an annual basis?
- Is the overall stability of an ecological network driven by large interannual variation among rarely observed species?

I investigated three different ecological networks: mistletoe-host interactions, vine-host interactions and plant-frugivore interactions. Plant-plant and plant-animal interactions are considered independently in each chapter. However, they do share theoretical application across three broad areas, including (i) resource availability, and (ii) temporal or spatial scaling.

Thesis structure

This thesis is compiled in a format where each chapter is an individual research project and written following journal guidelines – including word and page limitations. All research followed a unifying theme (ecological networks) except chapter four which considers the spatial distribution of mistletoe-host interactions. There is some overlap in each introduction; however, each study has an independent set of references. All chapters are prepared for journal submission and the contribution of co-authors and publication status is stipulated at the beginning of each chapter. I have followed a collaborative format by using ‘we’ instead of ‘I’ throughout this thesis, except for the introduction and summary chapters. Data for chapters four and five were collected by Dr Kevin Burns and Dr Rachael Gallagher (respectively); their contribution is acknowledged at the start of each chapter and inserted where relevant in the text.

References

- Alcantara, J.M. and Rey, P.J. 2012. Linking Topological Structure and Dynamics in Ecological Networks. *American Naturalist* 180: 186-199.
- Baldock, K.C.R., Memmott, J., Ruiz-Guajardo, J.C., Roze, D. and Stone, G.N. 2011. Daily temporal structure in African savanna flower visitation networks and consequences for network sampling. *Ecology* 92: 687-698.
- Bascompte, J., Jordano, P., Melián, C.J. and Olesen, J.M. 2003. The nested assembly of plant-animal mutualistic networks. *Proceedings of the National Academy of Sciences USA* 312: 431-433.
- Blüthgen, N. 2010. Why network analysis is often disconnected from community ecology: A critique and an ecologist's guide. *Basic and Applied Ecology* 11: 185-195.
- Blick, R. and Burns, K.C. 2009. Network properties of arboreal plants: Are epiphytes, mistletoes and lianas structured similarly? *Perspectives in Plant Ecology, Evolution and Systematics* 11: 41-52.
- Blick, R.A.J. and Burns, K.C. 2011. Liana co-occurrence patterns in a temperate rainforest. *Journal of Vegetation Science* 22: 868-877.
- Bray, D. 2003. Molecular Networks: The Top-Down View. *Science* 301:1864-1865.
- Burns, K.C. 2007. Network properties of an epiphyte meta-community. *Journal of Ecology* 95: 1142-1151.
- Burkle, L.A. and Knight, T.M. In press. Shifts in pollinator composition and behavior cause slow interaction accumulation with area in plant-pollinator networks. *Ecology* [<http://dx.doi.org/10.1890/12-0367.1>]

- Clements, F.E. 1909. Darwin's influence upon plant geography and ecology. *American Naturalist* 43: 143-151.
- Clements, F.E. 1916. Plant succession. Carnegie Institution, Washington, DC, USA (Publication number 242).
- Clements, F.E. 1936. Nature and structure of the climax. *Journal of Ecology* 24: 253-284.
- Connor, E.F. and Simberloff, D.S. 1979. The assembly of species communities: chance or competition? *Ecology* 60: 1132-1140.
- de Nooy, W., Mrvar, A. and Batagelj, V. 2005, Exploratory social network analysis with Pajek. Cambridge University Press. USA.
- DeWalt, S.J., Ickes, K., Nilus, R. Harms, K.E., and Burslem, D.F.R.P. 2006. Liana habitat associations and community structure in a Bornean lowland tropical forest. *Plant Ecology* 186: 203-216.
- Diamond, J.M. 1975. Assembly of species communities. In: Cody, M.L. & Diamond, J.M. (ed) *Ecology and evolution of communities*, Harvard University Press. Cambridge, MA, US, pp 342-444.
- Dormann, C.F. 2011. How to be a specialist? Quantifying specialisation in pollination networks. *Network Biology* 1: 1 - 20.
- Dormann, C.F., Fruend, J., Blüthgen, N. and Gruber B. 2009. Indices, Graphs and Null models: Analyzing Bipartite Ecological Networks. *The Open Ecology Journal* 2: 7-24.
- Dormann, C.F., Gruber B. and Fruend, J. 2008. Introducing the bipartite Package: Analysing Ecological Networks. *R news* 8: 8-11.

- Dunne, J.A., Williams, R.J. and Martinez, N.D. 2002. Network structure and biodiversity loss in food webs: robustness increases with connectance. *Ecology Letters* 5: 558-567.
- Florent, M., Francois, M. and Thierry, P. 2012. The role of epiphytism in architecture and evolutionary constraint within mycorrhizal networks of tropical orchids. *Molecular Ecology* 21: 5098-5109.
- Fortuna, M.A., Stouffer, D.B., Olesen, J.M., Jordano, P., Mouillot, D., Krasnov, B.R., Poulin, R. and Bascompte, J. 2010. Nestedness versus modularity in ecological networks: two sides of the same coin? *Journal of Animal Ecology* 79: 811–817.
- Fretwell, S.D. 1987. Food chain dynamics: The central theory of ecology? *Oikos* 50: 291–301.
- Genini, J., Côrtes, M.C., Guimarães Jr, P.R. and Galetti, M. 2012. Mistletoes Play Different Roles in a Modular Host–Parasite Network. *Biotropica* 44: 171-178.
- Gleason, H.A. 1917. The structure and development of the plant association. *Bulletin of the Torrey Botanical Club* 44, 463–481.
- Gleason, H.A. 1926. The Individualistic Concept of the Plant Association. *Bulletin of the Torrey Botanical Club* 53: 7-26.
- Godwin, H. 1929. The Sub-Climax and Deflected Succession. *Journal of Ecology* 17: 144-147.
- González, A.M.M., Allesina, S., Rodrigo, A. and Bosch, J. 2012. Drivers of compartmentalization in a Mediterranean pollination network. *Oikos* 121: 2001–2013.
- Gotelli, N.J. 2000. Null model analysis of species co-occurrence patterns. *Ecology*

81: 2606-2621.

Gotelli, N.J. and Graves, G.R. 1996. Null models in ecology. Smithsonian Institution Press, Washington, D.C.

Gotelli, N. and McGill, B. 2006. Null versus neutral models: what's the difference? *Oikos* 29: 793-800.

Gotelli N. J. and Ulrich W. 2012. Statistical challenges in null model analysis. *Oikos* 121: 171-180.

Guimarães, P.R., Rico-Gray, V., dos Reis, S.F. and Thompson, J. 2006. Asymmetries in specialization in ant-plant mutualistic networks. *Proceedings of the Royal Society B-Biological Sciences* 273: 2041–2047.

Guimerà, R., Stouffer, D.B., Sales-Pardo, M., Leicht, E.A., Newman, M.E.J. and Amaral, L.A.N. 2010. Origin of compartmentalization in food webs. *Ecology* 91: 2941-2951.

Hausdorf, B. & Hennig, C. 2007. Null model tests of clustering of species, negative co-occurrence patterns and nestedness in metacommunities. *Oikos* 116: 818-828.

Helen, R.E. and Handley, L.L. 2012. Networking: a community approach to invaders and their parasites. *Functional Ecology* 26: 1238-1248.

Hubbell, S.P. 2001. The Unified Neutral Theory of Biodiversity and Biogeography. Princeton University Press, Princeton, USA.

Ings, T.C., Montoya, M., Bascompte, J., Blüthgen, N., Brown, L., Dormann, C.F., Edwards, F., Figueroa, D., Jacob, U., Jones, J.I., Lauridsen, R.B., Ledger, M.E., Lewis, H.M., Olesen, J.M., van Veen, F.J.F., Warren, P.H. and Woodward, G.

2009. Ecological networks – beyond food webs. *Journal of Animal Ecology* 78: 253-269.
- James, A., Pitchford, J.W. and Plank, M.J. In press. Disentangling nestedness from models of ecological complexity. *Nature* [doi:10.1038/nature11214].
- Keddy, P. and Weiher, E. 1999. Assembly rules as general constraints on community composition. In: Weiher, E., Keddy, P. (Eds.), *Ecological Assembly Rules*. Cambridge University Press, Cambridge, UK.
- Kossinets, G. (2006) Effects of missing data in social networks. *Social Networks* 28: 247–268.
- Lázaro, A., Mark, S. and Olesen, J.M. 2005, Bird-made fruit orchards in northern Europe: nestedness and network properties. *Oikos*, 110: 321–329.
- Leibold, M.A., Holyoak, M., Mouquet, N., Amarasekare, P., Chase, J.M., Hoopes, M.F., Holt, R.D., Shurin, J.B., Law, R., Tilman, D., Loreau, M., and Gonzalez, A. 2004. The metacommunity concept: a framework for multi-scale community ecology. *Ecology Letters* 7: 601–613.
- Lewinsohn, T.M., Prado, P.I., Jordano, P., Bascompte, J. and Olesen, J.M. 2006. Structure in plant-animal interaction assemblages, *Oikos*, 113: 174–184.
- Lindeman, R. L. 1942. The trophic-dynamic aspect of ecology. *Ecology* 23: 399–417.
- Martinez, N.D., Hawkins, B.A., Alidawah, H. and Feikarek, B.P. 1999. Effects of sampling effort on characterization of food-web structure. *Ecology* 80: 1044-1055.
- McFarland, D. Messing, S., Nowak, M. and Westwood, S.J. 2010. Social Network Analysis Labs in R. Stanford University.

- Montoya, J.M., Pimm, S.L. and Sole, R.V. 2006. Ecological networks and their fragility. *Nature* 442: 259-264.
- Nielsen, A. and Bascompte, J. 2007. Ecological networks, nestedness and sampling effort. *Journal of Ecology* 95: 1134-1141.
- Nuismer, S.L., Pedro, P. and Bascompte, J. 2013. Copevolution and the architecture of mutualistic networks. *Evolution* 67 338-354.
- Olesen, J.M., Bascompte, J., Dupont, Y.L., Elberling, H., Rasmussen, C. and Jordano, P. 2011. Missing and forbidden links in mutualistic networks. *Proceedings of the Royal Society B-Biological Sciences* 278: 725-732.
- Ollerton, J., McCollin, D., Fautin, D.G. and Allen, G.R. 2007. Finding NEMO: nestedness engendered by mutualistic organization in anemone fish and their hosts. *Proceedings of the Royal Society B-Biological Sciences* 274: 591-598.
- Phillips, J. 1934. Succession, Development, the Climax, and the Complex Organism: An Analysis of Concepts. Part I. *Journal of Ecology* 22: 554-571.
- Piazzon, M., Larrinaga, A.R., & Santamaría, L. 2011. Are nested networks more robust to disturbance? A test using epiphyte-tree, comensalistic networks. *PLoS ONE* 6: e19637.
- Pimm S.L. 1979. The structure of food webs. *Theoretical population biology* 16: 144-158.
- Pires, M.M., Guimarães Jr, P.R., Araújo, M.S., Giaretta, A.A., Costa, J.C.L. and dos Reis, S.F. 2011. The nested assembly of individual-resource networks 80: 896-903.
- Pires, M.M. and Guimarães Jr, P.R. 2013. Interaction intimacy organizes networks of

- antagonistic interactions in different ways. *Journal of the Royal Society Interface* 10: 7.
- Proulx, S.R., Promislow, D.E.L., and Phillips, P.C. 2005. Network thinking in ecology and evolution. *Trends in Ecology and Evolution* 20: 345–353.
- Reid, S. and Armesto, J.J. 2011. Interaction dynamics of avian frugivores and plants in a Chilean Mediterranean shrubland. – *Journal of Arid Environments* 75: 221-230.
- Sfair, J.C., Rochelle, A.L.C., Rezende, A.A., van Melis, J., de Lara Weiser, V. and Martins, F.R. (2010) Nested liana-tree network in three distinct neotropical vegetation formations. *Perspectives in Plant Ecology, Evolution and Systematics* 12: 277-281.
- Silva, I.A., Ferreira, A.W.C., Lima, M.I.S. and Soares, J.J. 2011. Networks of epiphytic orchids and host trees in Brazilian gallery forests. *Journal of Tropical Ecology* 26: 127-137.
- Solé, R.V. and Montoya, M. 2001. Complexity and fragility in ecological networks. *Proceedings of the Royal Society B-Biological Sciences* 268: 2039-2045
- Stang, M., Klinkhamer, P.G.L. and van der Meijden, E. 2006. Size constrains and flower abundance determine the number of interactions in a plant-flower visitor web. *Oikos*, 112: 111–121.
- Stang, M., Klinkhamer, P.G.L., Waser, N.M., Stang, I. and van der Meijden, E. 2009. Size-specific interaction patterns and size matching in a plant–pollinator interaction web. *Annals of Botany* 103: 1459–1469.
- Strong, D. 1980. Null hypotheses in ecology. *Synthese* 43: 271-285.

- Stone, L. and Roberts, A. 1990. The checkerboard score and species distributions. *Oecologia* 85: 74-79.
- Stouffer, D.B. and Bascompte, J. 2011. Compartmentalization increases food-web persistence. *Proceedings of the National Academy of Sciences of the United States of America* 108: 3648-3652.
- Tansley, A.G. 1904. The problems with ecology. *New Phytologist* 3: 191-200.
- Tansley, A.G. 1913. A universal classification of plant communities. *Journal of Ecology* 1: 27-42.
- Tansley, A.G. 1920. The classification of vegetation and the concept of development. *Journal of Ecology* 8: 118-149.
- Tansley, A.G. 1935. The use and abuse of vegetational concepts and terms. *Ecology* 16: 284-307.
- Thébault, E. and Fontaine, C. 2010. Stability of Ecological Communities and the Architecture of Mutualistic and Trophic Networks. *Science* 329: 853-856.
- Vázquez D.P., Blüthgen, N., Cagnolo, L. and Chacoff, N.P. 2009. Uniting pattern and process in plant-animal mutualistic networks: a review. *Annals of Botany* 103: 1445-1457.
- van der Maarel, E. 2009. Pattern and process in the plant community: Fifty years after A.S. Watt. *Journal of Vegetation Science* 7: 19-28.
- Watt, A.S. 1947. Pattern and Process in the Plant Community. *Journal of Ecology* 35: 1-22.
- Watt, A.S. 1964. The community and the individual. *Journal of Animal Ecology* 33: 203-211.

Ulrich, W., M. Almeida-Neto, and N.J. Gotelli. 2009. A consumer's guide to nestedness analysis. *Oikos* 118: 3-17.

Ulrich, W. and N.J. Gotelli. 2007a. Disentangling community patterns of nestedness and species co-occurrence. *Oikos* 116:2053-2061.

Ulrich, W. and N.J. Gotelli. 2007b. Null model analysis of species nestedness patterns. *Ecology* 88:1824-1831.

Chapter Two

Predicting network topology of mistletoe-host interactions: do mistletoes really mimic their hosts?

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Abstract

Network analysis provides a unified framework for investigating different types of species interactions at the community level. Network analysis is typically based on null models that test for specific patterns in network topology. Here we use a novel predictive approach to investigate the topology of a mistletoe-host network. It has been hypothesised that Australian mistletoes mimic the phenotype of their preferred hosts to avoid herbivory. We developed a deterministic model based on phenotypic similarity to predict the topology of a quantitative network between Lauranthaceous mistletoes and their hosts. We quantified mistletoe-host interactions in a semi-arid woodland central Australia, along with the size, shape and colour of leaves produced by both players in the interaction. Traditional null model analyses showed support for negative co-occurrence patterns, web specialisation and strong links between species pairs. However, our deterministic model showed that the observed network topology could not be predicted by phenotypic similarity, suggesting that Australian mistletoes do not mimic their hosts.

Introduction

A primary goal in community ecology is to identify the processes that govern the arrangement of species assemblages. Recently, species interaction networks have provided a useful way of interpreting community-level patterns. Under this unified framework the arrangement of species interactions (topology hereafter) can describe community structure (Bascompte 2007, Bascompte and Jordano 2007, Bascompte et al. 2003, Blüthgen 2010, Blüthgen et al. 2008, Dormann et al. 2009, Guimarães et al. 2007, Jordano et al. 2003, Krishna et al. 2008, Lehsten and Harmand 2006, Ulrich and Gotelli 2007, Vázquez et al. 2009). The network approach has been applied to a wide range of biological interactions (including herbivory, pollination, seed dispersal, parasite-host relationships and predator-prey relationships) in ecosystems ranging from rainforests to coral reefs (Ings et al. 2009 and references within) and has provided further resolution to the on-going debate between assembly rules and stochastic processes in species interactions (Diamond 1975, Connor and Simberloff 1979).

Networks are generated by recording all species interactions (usually between two trophic levels) (Vázquez and Aizen 2003, Vázquez 2005, Vázquez et al. 2009) and evaluated using null model simulations that test a wide variety of network properties (Connor and Simberloff 1979, Gotelli and Graves 1995, Gotelli 2000, Dormann et al. 2009). Observed indices that deviate from those predicted by chance indicate deterministic processes (Gotelli 2000). For example, species-pairs that do not co-occur together and interact only with specialised ‘players’ in the network, or specific

geographic locations, generate checkerboard distributions and are interpreted as negative co-occurrence patterns (Stone and Roberts 1990). However, very different networks can often show similar structural properties (see Bascompte et al. 2003). For example plant-pollinator interactions and parasite-host interactions can show similar patterns of nestedness (Fortuna et al. 2010). Inferring which processes lead to the organisation of species interactions, and ultimately the convergence of network topology, presents a new set of challenges.

Here we propose an alternative method for evaluating network topology. In addition to null model evaluations, we derive a deterministic model that predicts species interactions according to phenotype (e.g. morphology or physiology). Under this new analytical approach, biologically informative traits can be used to predict observed network topology. This approach can be used to investigate the connectivity among species interactions limited by trait matching. For example, Stang et al. (2009) recently showed the utility of using morphological information to evaluate network properties (Stang et al. 2006; 2007). However, their approach determined size class distributions that were associated with nestedness, and not specialised interactions. Here we employ a deterministic approach to evaluate a plant-plant network that displays a high degree of phenotypic similarity.

Phenotypic similarity can have substantial effects on species interactions (see Thompson 2005) by increasing the removal rate of less favorable fruit, encouraging pollen transfer to flowers without reward (i.e. nectar) or decreasing herbivore

browsing (Burns 2005, Dyer and Murphy 2009, Fadzley et al. 2009, Gaskett and Herberstein 2010). For plant-herbivore interactions, the advantages of deception are beneficial if the plant can maintain traits that lead to aversion by herbivores (Williamson 1982). In the southern hemisphere, mistletoes (*Loranthaceae*) produce leaves that have a striking resemblance to the leaves of their preferred hosts. Along with the widely held view that mistletoe leaves mimic their host leaves, some (but not all) evidence points towards an adaptive strategy for leaf concealment (Barlow and Wein 1977, Ehleringer et al. 1986, Canyon and Hill 1997). These hemi-parasitic plants obtain water and some nutrients from host trees while maintaining their own photosynthetic ability (Aukema 2003). Therefore, mistletoes stand to gain if they conceal high water and nutrient content by displaying similar traits to their host tree (Canyon and Hill 1997, Mathiasen et al. 2008). Visual deception is an important factor that may influence the evolution of leaf symmetry in parasitic plants (Brown and Lawton 1991). However, all hypotheses proposed to explain apparent similarity in mistletoe-host interactions have remained unresolved (Barlow and Wein 1977, Ehleringer et al. 1986, Canyon and Hill 1997). In this study we test the hypothesis that mistletoe leaves mimic their host leaves by quantifying community-level patterns in matching and mismatching leaf traits.

Here, we test whether phenotypic similarity in leaf traits can predict the topology of an interaction network between mistletoes and their hosts. We quantified the distribution of mistletoes among host trees in a semi-arid woodland in central Australia and used traditional null models to establish the topology of the mistletoe-

host network. Next, we quantified the size, shape and reflectance properties of the leaves of each mistletoe and host species, which we then used to derive a deterministic model to test whether the observed network topology can be predicted by phenotypic similarity. Overall results are then used to test the hypothesis that Australian mistletoes mimic their hosts at the community level. Specifically we test the following hypotheses:

- 1) Mistletoe-host interactions are structured non-randomly.
- 2) Mistletoe leaves mimic (in shape and colour) host trees in the community.
- 3) Phenotypic similarity can predict the topology of the interaction network.

Methods

Location and species

Mistletoe-host interactions were recorded in a semi-arid environment surrounding Fowlers Gap Research Station, approximately 100km north of Broken Hill in western New South Wales, Australia (31°4'13" S, 141°42'16" E). The area has a mean annual rainfall of just 222.6 mm, and is exposed to extreme temperatures in summer (mean maximum daily temperature = 31.8 °C) and relatively low temperatures in winter (mean minimum daily temperature = 4.5°C) (Broken Hill weather station, Australian Government Bureau of Meteorology, <http://reg.bom.gov.au/>, last visited on 17 March 2011). Trees and large shrubs (i.e., potential hosts for mistletoe) are primarily found at the edges of ephemeral creeks. However, some trees can be found along connecting waterways and exposed ridge tops.

The riparian woodland community is dominated by *Eucalyptus camaldulensis* Dehnh. with an understory of *Myoporum* and *Santalum* species. Connecting waterways typically consist of *Eremophilla*, *Casuarina*, and *Alectryon* species, while exposed ridge tops are primarily populated by *Acacia anuera* F. Muell. ex Benth. However, most plant species occurred across multiple regions (Appendix 1), and are therefore considered a single plant community. We recorded 127 mistletoe-host interactions for four mistletoe species occurring within six kilometers of vegetation surrounding Fowlers Gap Research Station. The mistletoes *Amyema preissii* (Miq.) Tiegh., *A. maidenii* (Blakely) Barlow, *A. linophylla* Barlow and *Lysiana exocarpi* (Behr) Tiegh. were observed colonising ten host tree species, including *Acacia loderi* A. aneura, *A. salicina* Lindl., *A. victoriae* Benth. (Fig 1A), *Alectryon oleifolium* (Desf.) S.T.Reynolds, *Casuarina pauper* F.Muell. ex L.A.S.Johnson, *Eremophila alternifolia* R.Br., *E. longifolia* (R.Br.) F.Muell., *Santalum acuminatum* R.Br. A.DC. and *S. lanceolatum* R.Br. (Fig 1B). All *Amyema* species were observed on no more than two host species, while *L. exocarpi* was found on eight host tree species. *L. exocarpi* was observed colonising *Pittosporum angustifolium* and *Senna sturtii*; however they consisted of two rare interactions and removed from analyses for consistency between models (see below). Un-parasitised trees were not included in this study.

Network topology of mistletoe-host interactions: null model

Mistletoe-host interactions were recorded as one community-level matrix following Burns (2007). Null model simulations were first used to test whether the observed

assemblage of mistletoes and their hosts were structured non-randomly. To do this we randomly generated interactions for all species in the matrix by maintaining marginal row totals (mistletoe species) and allowing the marginal totals of each column to vary (host species). All simulations generated mistletoe-host interactions by randomly drawing host trees, without replacement, from a total pool of observed host interactions. The probability of detecting a mistletoe-host interaction was proportional to host abundance. Each randomly simulated matrix consisted of quantitative cell entries containing the incidence between species pairs. All simulations were replicated 1000 times using a fixed random seed and evaluated using three community-level indices including (1) network-level specialisation (i.e. H_2'), (2) as negative co-occurrence patterns between species pairs (i.e. c-score) and (3) within each 'cell' of the interaction network (i.e. interaction strength) (Appendix 2). These network indices were selected because they assess different levels of specialization.

First the H_2' index was used to calculate network-level interactions. H_2' calculates overall network specialisation by measuring the extent to which interactions within the matrix deviate from what is expected given the abundance of each species (Blüthgen 2006). H_2' is a standardised measure of specialization that ranges between 0 (generalists) and 1 (specialists) (Blüthgen 2006). Results close to one indicate that the proportion of links between species of each trophic level is lower than expected given the abundance distribution.

Second the c-score index was used to calculate negative co-occurrence patterns among mistletoe species. C-score calculates the average checkerboard score of all species interactions (Stone and Roberts 1990, Gotelli 2000). The c-score index was obtained by calculating the number of checkerboard units (cu) for each species pair: $cu = (O_i - S)(O_j - S)$, where O_i is the total number of host species occupied by mistletoe species i, O_j is the total number of host species occupied by mistletoe species j, and S is the number of host species occupied by both species (Stone and Roberts 1990). All simulated interactions were converted from quantitative counts to binary cell entries before calculating c-score. A c-score that is greater than predicted by chance indicates that mistletoes use different host species in the community.

Third we tested for non-random patterns in interaction strength. Interaction strength is a quantitative measure of links between species pairs. Interaction strength significantly greater than predicted by chance indicates that mistletoes infect specific host tree species (i.e. host preferences). We tested interaction strength by calculating the difference between observed and expected values using Wilcoxon signed ranked tests as normality assumptions were not met. All calculations were performed in the R environment (R Development Core Team 2010). C-score and H_2' indices were calculated using functions loaded from the bipartite package in R.

Phenotypic similarity

Phenotypic similarity in leaf size, shape and colour was quantified for 20 mature leaves collected from four individuals (five leaves from each plant) of each mistletoe

and host species. Mistletoe leaves were collected randomly from different host tree species in the community. *A. linophylla* was sampled from only two individuals due to low local abundance. Three measurements were obtained. Leaf width was measured at the widest point of the lamina using digital calipers, while leaf length and area were calculated using ImageJ (ver. 1.44p; Rasband 2008). *Casuarina* species have reduced leaves surrounding the internodes of photosynthetic stems. Here we considered the stems to be the primary photosynthetic organ, and thus the appropriate unit for morphological measurements. Tree species free from mistletoe infection and rare host species observed only once in the community were excluded from analysis. All data were \log_{10} transformed before analysis.

Leaf reflectance properties (i.e. colours) were quantified for all mistletoe and host species using an Ocean Optics USB 2000+ spectrometer and a PX-2 Pulsed Xenon Light source. A diffuse white reflectance standard was used to calibrate the spectrometer and spectral curves were measured as a proportion using Spectrasuite software. A fiber optics probe was fitted with an additional matt black tube modified to include a 45° angle and a 1 cm distance between the object and light source.

Reflectance spectra were recorded between 400 and 700 nm at 5 nm intervals.

Reflectance patterns were analysed in absence of perceptual color space (for instance a bird or insect vision model), because mistletoes in Australia experience herbivory from taxa with a range of different visual systems (including insects, marsupials, ratites and placental mammals), and the degree of selection pressure from each taxon is unknown. All wavelengths were adjusted to retain only the colour (chromatic)

signal by converting percent reflectance to a proportion for each wavelength. In other words, the achromatic (brightness) component was standardized among all replicates.

We tested for differences in morphology among all species pairs using two methods. First, we assessed whether mistletoes exhibit overlapping traits with their hosts. Pair wise comparisons of length, width, area, and colour of all mistletoe-host interactions were assessed individually using TukeyHSD with adjusted p values. Leaf colour was analysed using the first three principle components that accounted for over 80 percent of the variation in Principle Components Analysis following Endler (1990).

Our second approach was two-fold. We tested whether the collective arrangement of leaf traits for each mistletoe-host species pair was more or less similar than random expectations. Next we used these results to predict network topology (see below). First we sampled twenty leaves (regardless of species identity) randomly without replacement from a pooled dataset of all host leaves sampled ($n = 200$). Twenty leaves were sampled to ensure that predicted distributional trait values were comparable with those observed. Phenotypic similarity was calculated by measuring the Euclidean distance between individual host leaves (e.g. length, width and area) and each mistletoe species (i.e the distance from the centroid of all three traits). All Euclidean distances were averaged to generate the expected similarity in leaf traits from host tree species with randomly assorted leaves and each mistletoe species. This created a benchmark to examine the degree of phenotypic similarity among observed interactions. Observed phenotypic similarity was compared to 1000 simulations and

z-scores were obtained by calculating the number of standard deviations away from the randomised distribution: $z = (X_o - R_m) / R_{sd}$, where X_o is the observed Euclidean distance, R_m is the mean, and R_{sd} is the standard deviation of all randomly generated Euclidean distances.

Predicting network topology: deterministic model

Phenotypic similarity in leaf traits was used to predict network topology and results were used to investigate whether mistletoes mimic their hosts. The analytical approach we used applied weighted probabilities during the randomization procedure to generate each simulated matrix. Therefore, all simulated interactions were selected according to phenotype (i.e. leaf traits) and abundance of individuals in the community. We weighted the likelihood that each interaction could occur in the deterministic model following two approaches that have been proposed in the literature to explain phenotypic similarity in mistletoe-host interactions (Vane-Wright 1980, Barlow and Weins 1977).

First, we tested the hypothesis that being similar to a host is advantageous (mimicry hypothesis hereafter). To test the mimicry hypothesis we adjusted the randomisation procedure by weighting each host species by their similarity in leaf traits with each mistletoe species (i.e. observed Euclidean distance). Each host tree species was weighted relative to the most dissimilar trait combination. To do this we subtracted each Euclidean distance from the largest Euclidean distance in the entire matrix. Therefore, mistletoe-host interactions that were most dissimilar in leaf traits were

least likely to occur in each simulated matrix. All Euclidean distances were weighted as a proportion between 0 and 1, where 0 represents the most dissimilar trait combination and 1 represents the most similar trait combination.

Second, although the mimicry hypothesis is the most commonly believed, and the major focus of this manuscript, there are other hypotheses about mistletoe-host resemblance, such as adaptive similarity and the cryptic mimicry hypothesis that quantify mismatched traits (Barlow and Weins 1977, Canyon and Hill 1997). For example, mistletoes with high nutrient content gain more from appearing similar to the foliage of their host trees, while mistletoes with low nutrient content gain more from appearing dissimilar to the foliage of their host trees (Barlow and Weins 1977). Assigning mismatched combinations may have important implications for predicting network topology and are therefore included in this study (i.e. trait mismatching hereafter). To test trait mismatching we adjusted the randomisation procedure by weighting each host species with the divergence from the mean similarity of all host leaf traits using a standardised distance (i.e. z-score; Appendix 3). To do this we converted each z-score to a positive value. Therefore, mistletoe-host interactions with the most dissimilar leaf traits (positive z-score) were equally likely to occur as those with very similar leaf traits (negative z-score) in each simulated matrix. All Euclidean distances were weighted as a proportion between 0 and 1, where 0 represents trait combinations that are no different to random and 1 represents very similar or dissimilar trait combinations.

Initially we tested a combined model including leaf size, shape and colour. We followed up analyses of leaf size and shape and leaf colour individually as they often show conflicting displays of mimicry. All tests followed the assumptions of the mimicry hypothesis and trait mismatching as outlined above. Observed values that deviate from the expected distribution indicate poor correspondence between leaf traits and species interactions. We did not include a ‘threshold operator’ which recognizes forbidden links following Stang et al. (2009) as we assumed all interactions were possible in the community.

Some mistletoe species colonise several hosts (Downey 1998). Of course we do not expect these generalist mistletoes to resemble all hosts equally. Under the mimicry hypothesis, the prediction is for mistletoes to most closely resemble those hosts with which they have the strongest interactions. We tested how pervasive leaf similarity is in the community by quantifying the relationship between interaction strength and pair wise similarity. In addition we tested the performance of the deterministic approach by quantifying the relationship between predicted interaction strengths and observed interaction strengths of each species pair. We compared the null model approach with the deterministic approach for each combined model defined by the mimicry hypothesis and trait mismatching hypothesis using linear regression. Results indicate the explanatory power of including leaf traits in the evaluation of mistletoe-host network topology. All other analyses were conducted using ‘bipartite’ package in the R environment (R Development Core Team 2010) and PASW v.18.

Results

Network topology of mistletoe-host interactions: null model

We found non-random structure in network topology of mistletoes and their hosts.

Mistletoe-host interactions showed network-level specialisation ($H_2' = 0.913$; $z =$

33.318 , $p < 0.001$), negative co-occurrence patterns ($cu = 0.361$; $z = 2.749$, $p =$

0.003), and strong interaction strengths between species pairs ($z = 2.771$, $p < 0.001$).

Results did not differ when we included rare interactions ($H_2' = 0.913$; $z = 33.777$, $p <$

0.001 : $cu = 0.307$; $z = 2.725$, $p = 0.003$: $z = -2.981$, $p < 0.001$).

Phenotypic similarity

Initially we predicted that mistletoes would have overlapping leaf traits with

preferred host trees; however our analysis showed that single traits were often very

different between species pairs. In leaf size and shape *A. maidenii* was significantly

different to its only host, *A. aneura*, while *L. exocarpi* was significantly different to

A. victoriae and *E. alternifolia* for all traits. All remaining mistletoe-host pairs were

similar to their hosts in only one of three leaf dimensions (i.e. length, width, or area)

(Fig 1a). In leaf colour, *A. linophylla* was significantly different to its only host, *C.*

pauper, while *A. priessii* was significantly different to its only hosts, *A. victoriae* and

A. loderi. *L. exocarpi* was significantly different to three hosts including *A. salicina*,

A. oleifolium and *A. victoriae* (Fig 1b).

Next we asked whether the collective arrangement of leaf traits were more or less

similar than random expectations in the community. For leaf size and shape, eight out

of twelve mistletoe-host interactions were more similar than predicted by chance (Fig 2) indicating that comparisons of single leaf traits can underestimate similarity between mistletoes and their hosts. However, leaf size and shape was also shared with alternative trees (non host trees) in the community (Fig 2). For leaf colour, only three interactions collectively for *L. exocarpi* and *A. maidenii* were more similar than predicted by chance (Fig 3).

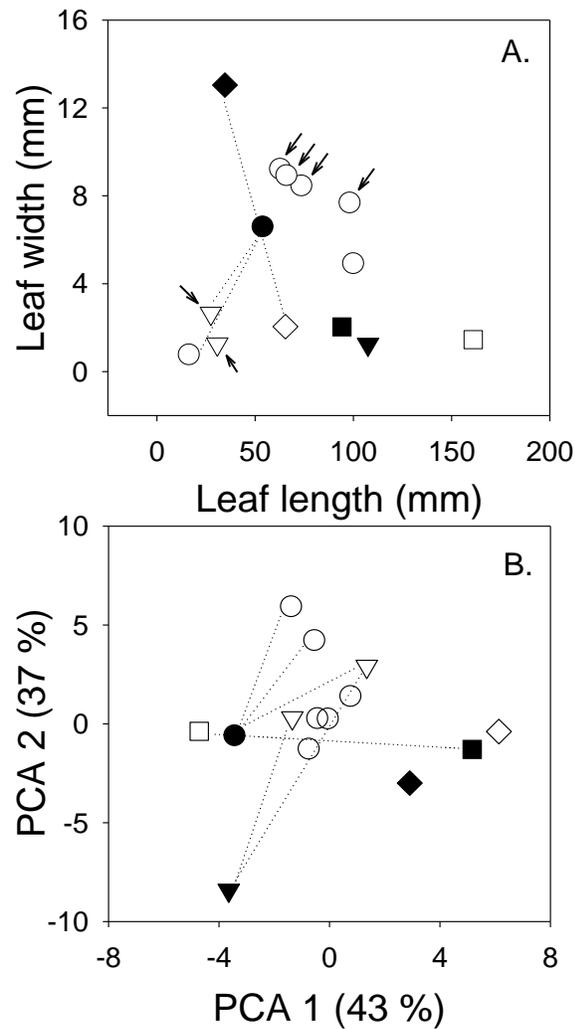


Figure 1 Pair wise comparisons of all mistletoes and their hosts according to (A) leaf size and shape and (B) leaf colour (PCA 1 and 2). Symbols represent species pairs; circles = *L. exocarpi*, triangles = *A. preissii*, diamonds = *A. maidenii*, and squares = *A. linophylla*. Filled symbols represent mistletoe species and open symbols represent host species. Lines indicate species pairs with significantly different traits across all dimensions. All remaining species pairs were similar in only one dimension. Leaf dimensions displayed as length and width only. Arrows indicate mistletoe species that were significantly different in leaf area to their preferred host tree species. All data was \log_{10} transformed before statistical analysis.

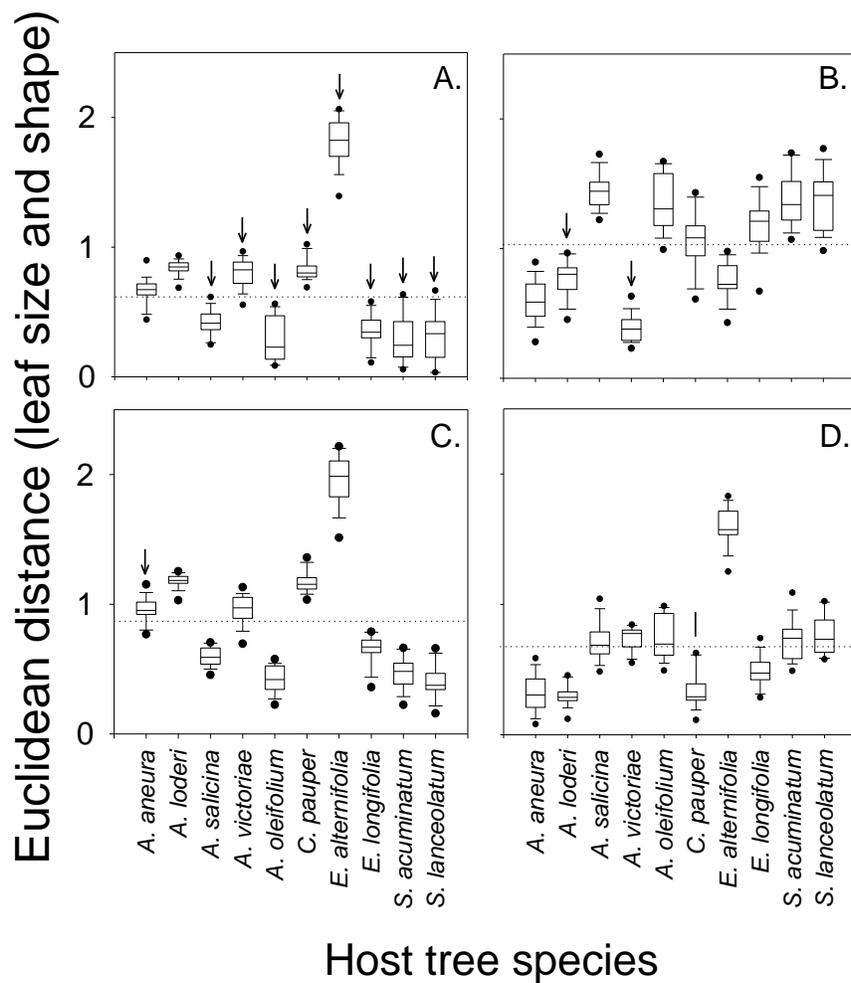


Figure 2 Similarities in leaf dimensions for all species pairs. Arrows indicate observed mistletoe-host interactions for (A) *L. exocarpi*, (B) *A. preissii*, (C) *A. maidenii* and (D) *A. linophylla*. The dashed line indicates expected similarity if leaf shape was distributed randomly among all species interactions. Error bars are 95 percent confidence intervals.

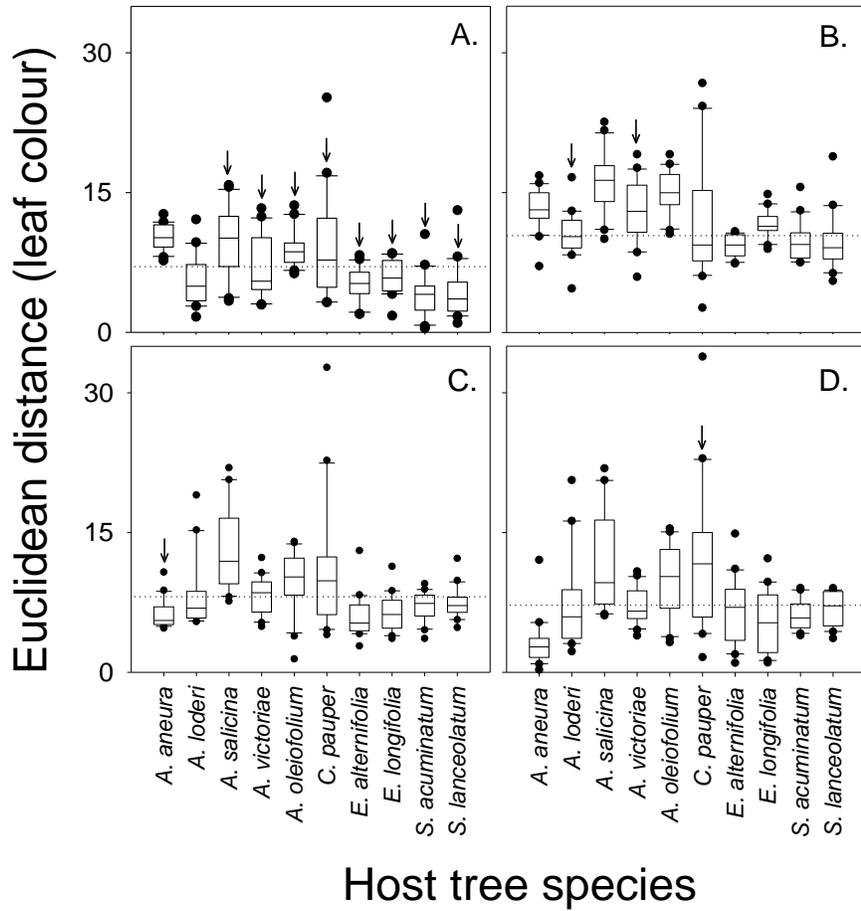


Figure 3 Similarities in leaf colour for all species pairs. Arrows indicate observed mistletoe-host interactions for (A) *L. exocarpi*, (B) *A. preissii*, (C) *A. maidenii* and (D) *A. linophylla*. The dashed line indicates expected similarity if leaf colour was distributed randomly among all species interactions. Error bars are 95 percent confidence intervals.

Predicting network topology: deterministic model

Phenotypic similarity did not predict observed network topology (Fig 4). A combined model including leaf size, shape and colour did not predict network level specialization ($H_2' = 0.086$; $z = 3.815$, $p < 0.001$), negative co-occurrence patterns ($cu = 0.128$: $z = 2.624$, $p = 0.004$) or interaction strengths ($z = -2.981$, $p < 0.001$).

Similarly, leaf size and shape, and leaf colour when analysed separately did not predict network-level specialisation ($H_2' = 0.095$; $z = 27.741$, $p < 0.001$; $H_2' = 0.108$, $z = 29.958$, $p < 0.001$, respectively), negative co-occurrence patterns ($cu = 0.128$: $z = 2.499$, $p = 0.006$; $cu = 0.144$, $z = 2.271$, $p = 0.012$; respectively) or interaction strengths ($z = -2.903$, $p < 0.001$; $z = -2.306$, $p < 0.001$; respectively) according to the mimicry hypothesis.

Similar results were found when we considered trait mismatches between mistletoes and their hosts. A combined model including leaf size, shape and colour did not predict network level specialization ($H_2' = 0.127$; $z = 23.740$, $p < 0.001$), negative co-occurrence patterns ($cu = 0.1865$: $z = 1.809$, $p = 0.035$) or interaction strength ($z = -2.668$, $p < 0.001$). Similarly, leaf size and shape, and leaf colour when analysed separately did not predicted network level specialization ($H_2' = 0.145$; $z = 21.936$, $p < 0.001$; $H_2' = 0.122$, $z = 26.37$, $p < 0.001$; respectively), negative co-occurrence patterns ($cu = 0.1918$: $z = 1.922$, $p = 0.027$; $cu = 0.194$, $z = 1.594$, $p = 0.055$; respectively) or interaction strengths ($z = -2.667$, $p = 0.002$; $z = -3.059$, $p < 0.001$; respectively) according to the trait mismatching hypothesis.

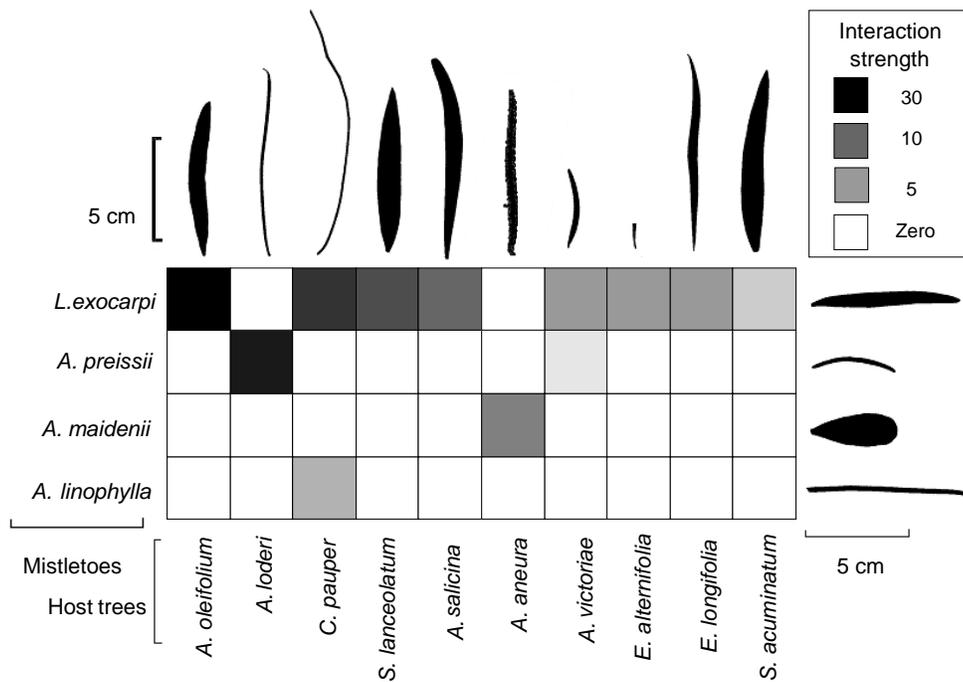


Figure 4 Network topology and leaf dimensions of all mistletoes and host trees (rows and columns, respectively) found at Fowlers Gap Research Station. Cell shade indicates interaction strength. Leaf size and shape depict the average dimensions of each plant species. The network image was developed using the “visweb” function in the bipartite library loaded in the R environment.

Neither the similarity in leaf size and shape, nor similarity in leaf colour, between mistletoe-host species pairs (measured as Euclidean distances) was correlated with interaction strength ($r^2 = 0.053$, $F_{1, 38} = 2.125$, $p = 0.153$; $r^2 = 0.013$, $F_{1, 38} = 0.483$, $p = 0.491$; respectively) indicating that apparent similarities in leaf traits are not a pervasive feature in the community. Predicted interaction strength from the null model (ie based on abundances alone) explained 47 percent of the variation in observed interaction strength ($r^2 = 0.472$, $F_{1, 38} = 33.962$, $p < 0.001$). Predicted interaction strength from the deterministic model for the mimicry hypothesis (which

included phenotypic similarity in addition to abundance), explained 43 percent of the variation in observed interaction strength ($r^2 = 0.431$, $F_{1, 38} = 28.731$, $p < 0.001$).

However, predicted interaction strength from the deterministic model for the trait mismatching hypothesis (which included phenotypic similarity in addition to abundance), explained just 23 percent of the variation in observed interaction strengths ($r^2 = 0.23$, $F_{1, 38} = 11.338$, $p = 0.002$).

Discussion

The topology of the mistletoe-host network could not be predicted by similarity in leaf traits. Mistletoe leaves are often suggested to match their hosts (Williamson 1982, Barlow and Weins 1977, Ehleringer et al. 1986, Canyon and Hill 1997).

However, size, shape and colour of leaves are also similar to non host trees. As such, the deterministic model did not predict co-occurrence patterns, interaction strengths or network specialisation. However, mistletoe-host interactions did show negative co-occurrence patterns, interaction strength and network specialisation indicating strong, exclusive host preferences.

The deterministic model did not predict host specificity when the deterministic model was defined by mimicry or trait mismatching in leaf size, shape and colour. In fact a model that incorporated biological traits performed worse than null model evaluations. This indicates that leaves of mistletoes in this study are not unique among their preferred host trees and appear to be shared traits in the community. Poor correspondence between c-score and interaction strength indicated that the

deterministic model generated alternative patterns of mistletoe-host interactions to that observed at Fowlers Gap. Shared leaf traits throughout the community may also indicate that mistletoes and trees use similar strategies for dealing with harsh environmental conditions. Traits such as leaf toughness, trichome density or internode length may provide further resolution to evolution of leaf size and shape in mistletoes. Predicting network topology of mistletoe-host interactions according to leaf traits is the first attempt that we are aware of at identifying mimicry in a plant community.

Surprisingly, few network evaluations have previously incorporated morphological and physiological information, despite many interaction types being highly suited to this sort of approach. For example, plant-pollinator interactions are the most widely investigated networks (Bascompte 2003, Bascompte et al. 2003, Jordano et al. 2003, Vázquez 2005, Bascompte and Jordano 2007 and references within) which are generally composed of complementary traits (e.g. proboscis length and corolla depth) (Stang et al. 2006, Rezende et al. 2007). However only one study to-date has applied phenotype under a unified framework that interprets community-level patterns (Stang et al. 2009). This approach may have important implications to many ecological food webs including predator-prey, plant-herbivore and symbiotic relationships and the evaluation of single or multiple processes that are underlying network topology.

Traditional null model analysis showed support for network-level specialisation, negative co-occurrence patterns and interaction strength. These results indicate that

mistletoes infect specific host trees exclusively in this plant community. The mechanisms underlying host specificity in parasitic plants are poorly known. One explanation for host specific interactions is that mistletoes have undergone local adaptation to host availability (Barlow and Weins 1977, Norton and Carpenter 1998). Although seed transplant experiments also show that some trees can resist mistletoe infection (Rodl and Ward 2002) indicating that host provenance is a combination of host availability and host quality (see Watson 2010). At present it remains unclear to what extent host resistance influences mistletoe communities. Disentangling the spatial aggregation of individuals and network topology may reveal exciting insights into mistletoe communities. Work is currently underway to address the role of host exploitation underlying network topology.

Plant-plant interaction networks have received much recent attention (Blick and Burns 2009, Blick and Burns 2011, Burns and Zotz 2010, Sfair et al. 2010, Silva et al. 2010). The fast expanding literature on plant-plant interaction networks indicate that different arboreal plant groups show different network topologies (Blick and Burns 2009). Mistletoes are primarily composed of host specific interactions and share similar network topology to lianas (Blick and Burns 2009, Blick and Burns 2011, but see Sfair et al. 2010). In contrast, epiphytes interact with hosts and other epiphytic species to form positive co-occurrence patterns, or more specifically, ‘nested’ patterns (Burns 2007, Burns and Zotz 2010, Silva et al. 2010). Differences in network topology likely reflect differences in deterministic processes. For example lianas found in a temperate New Zealand rainforest indicate that habitat partitioning is an

important determinant of their associations with host trees (Blick and Burns 2011). Interestingly, the network structure observed for mistletoes and their hosts in semi-arid Australia was similar to that previously observed for mistletoe-host interactions growing under very different conditions in New Zealand (Blick and Burns 2009). Further network evaluations of plant-plant interactions will reveal how consistent these trends are across different environments.

It remains an open question whether host specificity influences the evolution of leaf size and shape. In this study, leaf size and shape differed between mistletoes that produce broad leaves (*Amyema maidenii* and *Lysiana exocarpi*) and mistletoes that produce terete leaves (*A. linophylla* and *A. preissii*). Host specificity is known to vary drastically among these species. *L. exocarpi* has been recorded on 109 host species, *A. preissii* has been recorded on 73 host species, *A. maidenii* has been recorded on 37 host species, and *A. linophyllum* has been recorded on 8 host species (Downey 1998). Despite having a wide host range, it is suggested that mistletoes show phenotypic similarity to preferred host trees. Barlow and Weins (1977) qualitatively assessed these mistletoes as preferentially mimicking *Heterodendrum oleifolium* (now *Alectryon oleifolium*), *Acacia spp.*, *A. aneura* and *C. cristata*, respectively. However, our results only partially support this assessment. Currently, it remains unclear how spatial and temporal variations in host use influence leaf size and shape of different mistletoe species.

All current hypotheses that explain phenotypic similarity between mistletoes and their hosts suggest that herbivore damage will generate convergence in plant traits between species. However, plant-herbivore interactions form complex food webs consisting of both specialist and generalist herbivores (Novotny et al. 2010) that influence reproductive traits and leaf characteristics differently in spatially separated populations (Parra-Tabla and Herrera 2010, Muola et al. 2010). Recent studies have revealed the extent of damage caused by herbivory on mistletoe populations (Bach and Kelly 2004, Sweetapple 2008) suggesting that population level selection of plant traits is plausible. However, the only studies to examine this showed that mistletoe selection by herbivores was unrelated to leaf size and shape (Atsatt 1983, Canyon and Hill 1997). Furthermore, as our results showed considerable variation in phenotypic similarity in leaf size, shape and colour, we suggest that a universal hypothesis for mimicry in mistletoes is unlikely.

The hypothesis discussed so far has considered visual cues from herbivores, such as nocturnal marsupials. However, it is likely that other herbivores such as insects use olfactory cues for plant selection. In this study we did not explicitly test the range of herbivores currently feeding on mistletoe growing at Fowlers Gap, and previous morphological adaptations in evolutionary past can only be speculated. It is therefore, worthwhile considering alternative hypotheses void of herbivore-mediated selection. One alternative explanation follows that mimicry of mistletoe leaves is derived from selection pressure of fruit-eating birds (Calder 1983 and restated in Watson 2011). That is, mistletoe foliage that is cryptic within the tree canopy obtains a selection advantage

because fruit-eating birds must spend more time searching in new trees thereby facilitating seed dispersal. In contrast, mistletoe foliage that is conspicuous within the tree canopy obtains a disadvantage because fruit-eating birds spend more time in already infected trees, thereby facilitating seed dispersal within infected trees. We can then predict that trees containing ‘conspicuous’ mistletoe would be more heavily infested than cryptic associations. We did not observe this pattern at Fowlers Gap. Instead, the most cryptic association (*L. exocarpi* on its host *Alectryon oleifolium*) had the highest infection rate per tree, while the most conspicuous association (*A. maidenii* on its host *Acacia aneura*) had the lowest infection rate per tree (data not provided here; see chapter four). These observations are however, collected in a modified landscape of semi-arid Australia and we suggest that the results obtained in this study may differ at regional and continental scales.

Overall, plant mimicry has received much less attention than has animal mimicry (Schaefer and Ruxton 2009). As a result, understanding the evolution of deception in plants has lagged behind our understanding of deception in animals (Williamson 1982, Schaefer and Ruxton 2009). There have been few studies on cryptic leaf displays and even fewer on leaf mimicry (Brown and Lawton 1991). Leaf size and shape are known to be influenced by a wide range of factors including environmental and biotic interactions (Brown and Lawton 1991) that current mimicry hypotheses do not account for. Because mistletoes differ in host specificity (Downey 1998) and geographic range (Grenfell and Burns 2009), trait matching between mistletoe and host leaves may be better explained under the Geographic Mosaic of Coevolution

(Thompson 2005). Under this framework we suggest that (1) local adaptation in mistletoe leaves will not sweep through the entire population because herbivores differ among communities, (2) there will be common mismatches between leaf traits when multiple hosts are exploited in a community and (3) trait mixing between plants from neighboring communities will decrease selection for cryptic displays.

Overall we show that mistletoes do not mimic preferred host trees and we suggest that a unified framework consisting of geographic isolation (local environmental constraints), host specificity and herbivore interactions is required to explain the patterns of trait matches and mismatches at the community level.

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References

- Aukema, J.E. 2003. Vectors, viscin and Viscaceae: mistletoes as parasites, mutualists, and resources. - *Frontiers in Ecology* 1: 212-219.
- Bach, C.E. & Kelly, D. 2004. Effects of forest edges on herbivory in New Zealand mistletoe, *Alepis flavida*. - *New Zealand Journal of Ecology* 28: 195-205.

- Bascompte, J. 2003. Networks in Ecology. - *Basic and Applied Ecology* 8: 485-490.
- Bascompte, J. & Jordano, P. 2007. The structure of plant-animal mutualistic networks: the architecture of biodiversity. - *Annual Review of Ecology, Evolution, and Systematics* 38: 567-593.
- Bascompte, J. et al. 2003. The nested assembly of plant-animal mutualistic networks. - *Proceedings of the National Academy of Sciences USA* 312: 431-433.
- Barlow, B.A. & Weins, D. 1977. Host-parasite resemblance in Australian mistletoes: The case for cryptic mimicry. - *Evolution* 31:69-84.
- Blick, R. & Burns, K.C. 2009. Network properties of arboreal plants: Are epiphytes, mistletoes and lianas structured similarly? - *Perspectives in Plant Ecology, Evolution and Systematics* 11: 41-52.
- Blick, R.A.J. & Burns, K.C. 2011. Liana co-occurrence patterns in a temperate rainforest. - *Journal of Vegetation Science*.
- Brown, V.K. et al. 1991. Herbivory and the evolution of leaf size and shape. - *Philosophical Transactions of the Royal Society of London B* 333: 265-272.
- Burns, K.C. 2005. Evidence for mimicry in fleshy-fruits. - *Evolutionary Ecology Research* 7: 1067-1076.
- Burns, K.C. 2007. Network properties of an epiphyte meta-community. - *Journal of Ecology* 95: 1142-1151.
- Burns, K.C. & Zotz, G. 2010. A hierarchical framework for investigating epiphyte assemblages: networks, metacommunities and scale. - *Ecology* 91: 377-385.
- Blüthgen, N. et al. 2006. Measuring specialisation in species interaction networks. - *BMC Ecology* 6: 9.

- Blüthgen, N. et al. 2008. What do interaction network metrics tell us about specialisation and biological traits? - *Ecology* 89: 3387-3399.
- Blüthgen, N. 2010. Why network analysis is often disconnected from community ecology: A critique and an ecologist's guide. - *Basic and Applied Ecology* 11: 185-195.
- Calder, M. 1993. *The Biology of mistletoes*. Academic Press. USA.
- Canyon, D.V. & Hill, C.J. 1997. Mistletoe host resemblance: A study of herbivory, nitrogen and moisture in two Australian mistletoes and their host trees. - *Australian Journal of Ecology* 22: 395-403.
- Connor, E.F. & Simberloff, D.S. 1979. The assembly of species communities: chance or competition? - *Ecology* 60: 1132-1140.
- Diamond, J.M. (1975) Assembly of species communities. In: Cody, M.L., Diamond, J.M. (Eds.), *Ecology and Evolution of Communities*. Harvard University Press, Cambridge, USA, pp. 342-444.
- Dormann, C.F. et al. 2009. Indices, Graphs and Null models: Analyzing Bipartite Ecological Networks. - *The Open Ecology Journal* 2: 7-24.
- Downey, P.O., 1998. An inventory of host species for each aerial mistletoe species (Loranthaceae and Viscaceae) in Australia. - *Cunninghamia* 5: 685-720.
- Dyer, A.G. & Murphy, A.H. 2009. Honeybees choose "incorrect" colors that are similar to target flowers in preference to novel colors. - *Israel Journal of Plant Sciences* 57: 203-210.
- Ehleringer, J.R. et al. 1986. Mistletoes: a hypothesis concerning morphological and chemical avoidance of herbivory. - *Oecologia* 70: 234-237.

- Endler, J.A. 1990. On the measurement and classification of color in studies of animal color patterns. - *Biological Journal of the Linnean Society* 41: 315-352.
- Fadzly N. et al. 2009. Ontogenetic colour changes in an insular tree species: Signalling to extinct browsing birds? - *New Phytologist* 184: 495-501.
- Fortuna, M.A. 2010. Nestedness versus modularity in ecological networks: two sides of the same coin? - *Journal of Animal Ecology* 79: 811–817.
- García, D. et al. 2009. Seed dispersal by frugivorous marsupial shapes the spatial scale of a mistletoe population. - *Journal of Ecology* 97: 217-229.
- Gaskett, A.C. & Herberstein, M.E. 2010. Colour mimicry and sexual deception by Tongue orchids (*Cryptostylis*). - *Naturwissenschaften* 97: 97-102.
- Gotelli, N.J. 2000. Null model analysis of species co-occurrence patterns. - *Ecology* 81: 2606-2621.
- Gotelli, N.J. & Graves, G.R. 1995. *Null models in ecology*. Smithsonian Institution Press, Washington, D.C.
- Grenfell, M. & Burns, K.C. 2009. Sampling effects and host ranges in Australian mistletoes. - *Biotropica* 41: 656-658.
- Guimarães, P.R. 2007. Interaction intimacy affects structure and coevolutionary dynamics in mutualistic networks. - *Current Biology* 17: 1797-1803.
- Ings, T.C. 2009. Ecological networks – beyond food webs. - *Journal of Animal Ecology* 78: 253-269.
- Jordano, P. et al. 2003. Invariant properties in coevolutionary networks of plant-animal interactions. - *Ecology Letters* 6: 69-81.

- Ladley, J.J. & Kelly, D. 1996. Dispersal, germination and survival of New Zealand mistletoes (Loranthaceae): dependence on birds. - *New Zealand Journal of Ecology* 20: 69-79.
- Lehsten, V. & Harmand, P. 2006. Null models for species co-occurrence patterns: assessing bias and minimum iteration number for the sequential swap. - *Ecography* 29: 786-792.
- Krishna, A. et al. 2008. A neutral-niche theory of nestedness in mutualistic networks. - *Oikos* 117: 1609-1618.
- Mathiasen, R.L. 2008. Mistletoes: Pathology, systematics, ecology, and management. - *Plant Disease* 92: 998-1006.
- Muola, A. 2010. Associations of plant fitness, leaf chemistry, and damage suggest selection mosaic in plant-herbivore interactions. - *Ecology* 91: 2650-2659.
- Norton, D.A. & Carpenter, M.A. 1998. Mistletoes as parasites: host specificity and speciation. - *Trends in Ecology and Evolution* 13: 101-105.
- Novotny, V. 2010. Guild-specific patterns of species richness and host specialization in plant-herbivore food webs from tropical forest. - *Journal of Animal Ecology* 79: 1193-1203.
- Parra-Tabla, V. & Herrera, C.M. 2010. Spatially inconsistent direct and indirect effects of herbivory on floral traits and pollination success in tropical shrub. - *Oikos* 119: 1344-1354.
- R Development Core Team (2010) R: a language and environment for statistical computing. Vienna, Austria. <http://www.R-project.org>

- Rasband W. S. 1997/2009. ImageJ. – US Natl Inst. of Health
<<http://rsb.info.nih.gov/ij/>>
- Rezende, E.L. 2007. Non-random coextinctions in phylogenetically structured mutualistic networks. - *Nature* 448: 925-928.
- Rodl, T. & Ward, D. 2002. Host recognition in desert mistletoe: early stages of development are influenced by substrate and host origin. - *Functional ecology* 16: 128-134.
- Sfair, J.C. 2010. Nested liana-tree network in three distinct neotropical vegetation formations. - *Perspectives in Plant Ecology, Evolution and Systematics* 12: 277-281.
- Schaefer, H.M. & Ruxton, G.D. 2009. Deception in plants: mimicry or perceptual exploitation? - *Trends in Ecology and Evolution* 24: 676-683.
- Stang, M. et al. 2006. Size constraints and flower abundance determine the number of interactions in a plant-flower-visitor web. - *Oikos* 112: 111-121.
- Stang, M. et al. 2007. Asymmetric specialisation and extinction risk in plant-flower-visitor webs: a matter of morphology or abundance? - *Oecologia* 151: 442-453.
- Stang, M. 2009. Size specific interaction patterns and size matching in a plant-pollinator interaction web. - *Annals of Botany* 103: 1459-1469.
- Silva, I.A. 2010. Networks of epiphytic orchids and host trees in Brazilian gallery forests. - *Journal of Tropical Ecology* 26: 127-137.
- PASW Statistics 18 (formerly SPSS Statistics) for windows. SPSS Inc., *Chicago, USA*
- Stone, L. & Roberts, A. 1990. The checkerboard score and species distributions. -

Oecologia 85: 74-79.

- Sweetapple, P.J. 2008. Spatial variation in impacts of brushtail possums on two Loranthaceous mistletoe species. - *New Zealand Journal of Ecology* 32: 177-185.
- Thompson, J.N. 2005. *The Geographic Mosaic of Coevolution*. University of Chicago Press, Chicago, USA.
- Ulrich, W. & Gotelli, N.J. 2007. Disentangling community patterns of nestedness and species co-occurrence. - *Oikos* 116: 2053-2061.
- Vázquez, D.P. 2005. Degree distribution in plant-animal mutualistic networks: forbidden links or random interactions? - *Oikos* 108: 421-426.
- Vázquez, D.P. & Aizen, M.A. 2003. Null model analyses of specialisation: a pervasive feature of plant-pollinator interactions. - *Ecology* 84: 2493-2501.
- Vázquez, D.P. et al. 2009. Evaluating multiple determinants of the structure of plant-animal mutualistic networks. - *Ecology* 90: 2039-2046.
- Vanewright, R.I. 1980. On the definition of mimicry. - *Journal of the Linnean Society* 13: 1-6.
- Watson, D.M. 2009. Determinants of parasitic plant distribution: the role of host quality. - *Botany* 87: 16-21.
- Watson, D.M. 2011. *Mistletoe of southern Australia*. Csiro Publishing. Australia.
- Williamson, G.B. 1982. Plant mimicry – Evolutionary constraints. - *Biological Journal of the Linnean Society* 18: 49-58.

Appendix 1

Mistletoe-host interactions in different habitats.

	<u>Host</u>	<u>Mistletoes</u>
Exposed ridge	<i>Acacia aneura</i>	<i>Amyema maidenii</i>
Connecting waterways	<i>Acacia salicina</i> <i>Senna sturtii</i> <i>Pitosporum angustifolium</i> <i>Eremophila alternifolia</i> <i>Acacia victoriae</i>	<i>Lysiana exocarpi</i>
Creekline	<i>Acacia loderi</i> <i>Acacia salicina</i> <i>Acacia victoriae</i> <i>Eremophila longifolia</i> <i>Casuarina pauper</i> <i>Alecrtyon oleiofolium</i>	<i>Amyema preissii</i> <i>Amyema linophyllum</i> <i>Lysiana exocarpi</i>

Appendix 2

R script used to simulate network properties. Network indices calculated using functions loaded in the bipartite package. C.score is given here as an example.

Null model

```
Data<-read.table("F:\\file.txt",header=T)
attach(Data)
names(Data)
"host"
library(bipartite)
set.seed(34263)
for(i in 1:1000){result<-matrix(c(
(as.vector(table(sample(host,9)))),(as.vector(table(sample(host,26))))),
(as.vector(table(sample(host,87)))),(as.vector(table(sample(host,3))))),
nrow=10,ncol=4)
res<-ifelse(result>=1,1,0)
R<-matrix(res,byrow=T,nrow=4)
CS<-C.score(R)
write((CS),file="file.txt",append=TRUE)}
```

Deterministic model

```
set.seed(34263)
for(i in 1:1000){
LE<-c(rep(0.3,5),rep(0.15,2),rep(0.01,4),
AM<-c(rep(0.2,5),rep(0.345,2),rep(0.780,4),
AP<-c(rep(0.1,5),rep(0.5,2),rep(0.874,4),
AL<-c(rep(0.4,5),rep(0.91,2),rep(0.411,4),

#example probabilities for 3 host
species of 4 mistletoes. These are
manually calculated and
imported into this script.

result<-matrix(c(
(as.vector(table(sample(host,9,prob=AM)))),(as.vector(table(sample(host,26,prob=A
P))))),
(as.vector(table(sample(host,87,prob=LE))))),
(as.vector(table(sample(host,3,prob
=AL))))),nrow=3,ncol=4)

# Random sample from list AND
Probabilities. Row total must
match the number of host species
– eg 3 in this example

res<-ifelse(result>=1,1,0)
R<-matrix(res,byrow=T,nrow=4)
CS<-C.score(R)

#command to execute c-score – function
loaded in bipartite package

write((CS),file="file.txt",append=TRUE)}
```

Appendix 3

Z-scores show the similarity (Euclidean distances) between mistletoe leaves and host leaves and measured as the deviation away from a random distribution. Negative values represent interactions that are more similar than predicted by chance.

<i>Amyema maidenii</i>	<i>Amyema preissii</i>	<i>Lysiana exocarpi</i>	<i>Amyema linophylla</i>	Leaf size and shape
0.81	-2.66	-0.01	-2.49	<i>Acacia aneura</i>
5.70	-1.87	3.10	-5.01	<i>Acacia loderi</i>
-3.70	3.20	-2.36	0.27	<i>Acacia salicina</i>
0.78	-6.17	1.32	0.81	<i>Acacia victoriae</i>
-4.36	1.51	-2.16	0.43	<i>Alectryon oleiofolium</i>
3.56	0.13	1.95	-2.53	<i>Casuarina pauper</i>
5.85	-1.93	6.56	6.06	<i>Eremophila alternifolia</i>
-2.06	0.81	-2.42	-1.59	<i>Eremophila longifolia</i>
-3.33	1.54	-1.82	0.41	<i>Sanatlum acuminatum</i>
-3.90	1.76	-2.09	0.63	<i>Santatlum lanceolatum</i>

<i>Amyema maidenii</i>	<i>Amyema preissii</i>	<i>Lysiana exocarpi</i>	<i>Amyema linophylla</i>	Leaf colour
-1.46	0.59	2.48	-1.76	<i>Acacia aneura</i>
-0.03	-0.64	-0.55	-0.05	<i>Acacia loderi</i>
1.07	1.24	0.76	0.74	<i>Acacia salicina</i>
-0.02	0.30	-0.01	-0.24	<i>Acacia victoriae</i>
0.48	1.36	0.99	0.58	<i>Alectryon oleiofolium</i>
0.40	-0.03	0.36	0.58	<i>Casuarina pauper</i>
-1.13	-2.28	-1.05	-0.33	<i>Eremophila alternifolia</i>
-1.04	-0.22	-0.59	-0.65	<i>Eremophila longifolia</i>
-0.86	-1.15	-1.28	-0.90	<i>Sanatlum acuminatum</i>
-0.62	-0.76	-1.01	-0.43	<i>Santatlum lanceolatum</i>

Chapter Three

Dominant network interactions are not correlated with resource availability: a case study using mistletoe-host interactions

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This paper was conceived by RB, who also carried out all fieldwork, meta-analyses, formulated a new r script for null model analysis and did most of the writing. Input on statistical methods and manuscript revisions were contributed by KB and AM.

Abstract

Network theory in ecology has been central to understanding species co-occurrence patterns, specialization and community stability. However, network theory has traditionally focused on the ‘higher’ trophic level where exploitation of network ‘partners’ (i.e., individual interactions in response to resource availability) have remained underappreciated. In this study we tested how clumping and host availability influenced mistletoe-host interactions in a semi-arid woodland, central Australia. We used a hierarchical approach that evaluated individual interactions by modifying the traditional randomization technique to simulate clumping and host exploitation. Using published literature we then compared our results with mistletoes from other genera. We found that mistletoes clump on fewer trees than predicted, even though interaction strength was no different from random expectations, and we found no evidence that common trees were heavily infected as predicted by the host availability hypothesis. The rate of host exploitation (measured as the proportion of trees infected) in semi-arid Australia is similar to that for mistletoe genera in other parts of the world. We hypothesize that specific host trees act as a focal point for infection that facilitates the spread and overall population size of mistletoes. Overall our results indicate that resources, such as the number of trees in a mistletoe network, are less important than clumping of individual plants. We suggest that exploitation of available resources may play a similar role in other networks that extend beyond antagonistic relationships such as parasite or herbivore interactions.

Introduction

Ecological networks are used to evaluate interactions between species from one trophic level (such as birds) with species from a different trophic level (such as plants) (Bascompte et al. 2003). Over the last decade, both theoretical and empirical research has vastly improved our understanding of species interactions (Jordano 1987, Stone and Roberts 1990, Gotelli 2000, Bascompte et al. 2003, Guimarães et al. 2007, Ulrich and Gotelli 2007, Blüthgen et al. 2008, Dormann et al. 2009, Ings et al. 2009, Vázquez et al. 2009a, Vázquez et al. 2009b, Blüthgen 2010). Subsequently, ecological networks are evaluated using quantitative data that weights the frequency of interactions between individuals of each species pair (Burns 2007, Blick and Burns 2009, Krishna et al. 2010). However, a major gap in network thinking is the link between individuals and patterns of distribution (e.g. aggregation or clumping). In this study we bridge the gap between individual plants, clumping, and network topology.

Recently, individual-resource networks have been used to test how a single species occupies different resources (Pires et al. 2011). This is important because individual-resource networks show that there is hierarchical structure underpinning all species-based interaction networks; where the dispersion of individuals across multiple resources generates the topological signature of the community (Burns and Zotz 2010, Blick and Burns 2011). We use dispersion here to describe a wide range of distributions including optimum foraging strategies, ideal free distributions and negative binomial distributions. Integrating patterns of dispersion into network

topology is needed to scale up from species pairs to ecological networks – which are critically important to all species interactions. Specifically, in this study we investigate the role of clumping (i.e., the accumulation of multiple mistletoes in a single host tree) in structuring a mistletoe-host interaction network.

Mistletoes are hemi-parasitic plants recognized for their important role in stability and functioning of tropical, temperate, and arid systems (Watson 2002, Aukema 2003, Mathiasen et al. 2008). Within these ecosystems, mistletoes often exhibit patterns of clumping (also called aggregation or clustering of multiple mistletoe on a single tree); where compatible host trees (see Rödl and Ward 2002) carry multiple infections and neighboring trees harbor significantly fewer numbers of mistletoe (Hoffman et al. 1986, Yan 1990, Lavorel et al. 1999, Reid and Smith 2000, Watson 2009a, Watson 2009b, Carnegie et al. 2009, Rist et al. 2010). Similar patterns are found in marine copepods (*Gastrodelphys clausii*; Nash and Keegan 2006), human parasitic roundworms (*Ascaris lumbricoides*; Walker et al. 2010) and galls produced by wasps (*Diplolepis rosae*; László and Tóthmérész 2011) indicating that clumping may be a generally important mechanism structuring other ecological networks.

Dispersion of individuals may be inextricably linked with resource availability in the community. Resource availability is critically important for mistletoes because each species is dependent on a specific set of host tree species in the community (Blick and Burns 2009, Blick et al. 2012, Genini et al. 2011). Thus, the number of interactions observed in a community may be determined by the relative number of

mistletoe individuals and the number of trees in the ecosystem. Despite its obvious importance for network structure, the reciprocal effect between species interactions has only recently been investigated (Vázquez et al. in press). Here we establish a new method for assessing host tree exploitation (the proportional number of trees infected with mistletoe) and test its relationship with other commonly employed network properties.

The host availability hypothesis states that mistletoes infect the most common trees in a community (Norton and Carpenter 1998, Kavanagh and Burns in press). If the host availability hypothesis is a valid explanation for the structure of mistletoe-host interactions, then the number of trees available in the community should predict the number of infected trees. In this study we used host tree exploitation between species pairs to test the host availability hypothesis. To ensure that we were not characterizing a unique property of one mistletoe community we extended our analysis to published literature. This study is important because we provide a method for integrating easily obtainable biological information into network analysis and establish a deterministic approach to evaluating the structure of an ecosystem.

We quantified the dispersion of individual mistletoes in semi-arid woodland of Australia in order to test the role of host exploitation among different mistletoe species, and compared host use to other common network properties. We then used a modified randomization procedure that simulated multiple infections on individual

trees to quantify the role of dispersion in structuring a quantitative species-based network.

Specifically, we asked the following questions:

1. Are mistletoes evenly dispersed in the community?
2. Is tree availability correlated with network properties?
3. Does the host-availability hypothesis explain the structure of a mistletoe community?

Methods

Location and study species

Mistletoe-host interactions were recorded in a semi-arid environment surrounding Fowlers Gap Research Station, 112 km north of Broken Hill in western New South Wales, Australia (31°4'13" S, 141°42'16" E). Mean annual rainfall reaches 222.6 mm, and is exposed to extreme temperatures in summer (maximum temperature = 45.0 °C) and winter (minimum temperature = -3.0 °C) (Data from November 2004 to December 2011; weather station 46128, Fowlers Gap Research Station). Trees and large shrubs (i.e., potential hosts for mistletoe) are primarily found at the edges of ephemeral creeks. However, trees are also found along connecting waterways and exposed ridge tops. The riparian woodland community is dominated by *Eucalyptus camaldulensis* and connecting waterways typically consist of *Myoporum*, *Eremophilla*, *Casuarina*, and *Alectryon* species. Exposed ridge tops are primarily populated by *Acacia aneura*.

We surveyed a set number of woody trees ($n = 1800$) rather than establishing transect lines that would likely misrepresent the often sporadic vegetation at Fowlers Gap. All trees were inspected in three locations including a creek bed, isolated trees, and an exposed ridge. In each location, six hundred woody trees greater than two meters tall were visually inspected for mistletoe. Trees less than 2 meters tall were not recorded during data collection. We considered tree height of two meters to be a conservative measure of the potential host community. During the course of this study we did not see any mistletoe growing on trees less than two meters tall. Individual mistletoes were recorded as the number of haustoria per tree because all mistletoe species found at Fowlers Gap form a single connection to their host (i.e., absence of external runners that generate multiple connections).

Four mistletoe species have been identified at Fowlers Gap (Blick et al. 2012). Fowlers Gap is located at the northern and southern geographic range of *Amyema linophylla* and *A. maidenii* (respectively), and the eastern geographic range of *A. priessii*. *Lysiana exocarpi* has the widest geographic range and common in areas surrounding Fowlers Gap (Appendix 1). Mistletoes were recorded growing on 12 host tree species including, *Acacia loderi*, *A. aneura*, *A. salicina*, *A. victoriae*, *Alectryon oleifolium*, *Casuarina pauper*, *Eremophila longifolia*, *E. alternifolia*, *Pittosporum angustifolium*, *Santalum acuminatum*, *S. lanceolatum* and *Cassia sturtii*. During the time of this study *Santalum* species were primarily without fruit and not confidently

identified to species level. All *Santalum* species were only infected by *L. exocarpi* and later joined together as *Santalum* spp..

We evaluated the dispersion of individual mistletoes by implementing a range of analytical techniques. First we used a null model to test the overall rate of host use regardless of species identity (total number of trees infected). Second, we used the variance-to-mean ratio (VMR) and a chi-squared goodness-of-fit test following a Poisson distribution to calculate the dispersion of mistletoes. Trees with greater than 14 mistletoes were combined during the chi-squared test to remove zero entries and evaluated with 13 degrees of freedom. Botanical nomenclature follows Cunningham et al. (1981) and Cayzer et al. (2000).

We recorded the number of individual mistletoes occurring on all trees as a single quantitative interaction matrix. Previous analyses of network structure at Fowlers Gap indicated that each mistletoe species occurs on a specific set of host tree species (conducted November 2009; Blick et al. 2012). However, after surveying the same vegetation in August of 2011 we found *A. maidenii* growing on an additional host species (*Acacia tetragonophylla*) and *A. preissii* growing on two additional host species (*Acacia aneura* and *A. tetragonophylla*) indicating that a greater number of interactions in this plant community are possible, albeit rare. To ensure our dataset was consistent with the previous network we conducted null model simulations following Blick et al. (2012). Therefore, we could be confident that any differences in results were due to changes in the randomization procedure (below) and not due to

new interactions in the network. The new network with additional links supported previous results by maintaining a higher number of interaction absences than predicted by chance (checkerboard units: $cu = 0.5$, $z = -17.315$ $p < 0.001$; web specialization: $H^2 = 0.658$, $z = -50.074$, $p < 0.001$; interaction strength (Wilcoxon signed rank test): $z = -1.851$, $p = 0.067$). Spatial and temporal changes in mistletoe interactions may indicate transitivity in network structure that is influenced by changing weather patterns (e.g. increased annual rainfall) or sampling intensity; however further research is required to validate these hypotheses.

Null model analysis

In this study we employed a randomization technique that accounted for clumping in mistletoes. Null model simulations are typically evaluated in absence of biological information (Gotelli and Graves 1996, Gotelli 2000, Lehsten and Harmond 2006). However, results from null model simulations are ultimately determined by which constraints are imposed. For example, the simulation may include the randomization of individuals or impose column or row constraints (Gotelli and Ulrich 2011). Our randomization procedure included the abundance of all ‘available’ trees and all mistletoe individuals (i.e. proportional probability of tree species abundances). Available trees are defined as the total number of individual trees (regardless of mistletoe infection), from each species that had at least one mistletoe present. All tree species with no mistletoe (column or row absences) were removed from the model because it would generate null communities of unrealistic species interactions. Marginal row totals representing the number of individuals of each mistletoe species

was fixed, and marginal column totals representing the number of individuals of each host species were allowed to vary (equiprobable). Therefore individual mistletoes were capable of infecting any tree while maintaining the observed rank abundance of mistletoes. In order to simulate clumping, each randomized matrix was generated using a with-replacement method of cell fill; where each tree is equally likely to exclusively harbor all mistletoes from the model. To do this we generated a dataset containing individual trees with unique identities. During each simulation, a new matrix was generated that consisted of individual trees (represented by 1298 rows) harboring the sum of all mistletoes of four species (represented by 4 columns). All matrices were amalgamated before analysis and transposed so that mistletoe species represent rows and host tree species represent columns. Network indices typically evaluate a higher trophic level as columns; however we transposed matrices in this study to ensure that we were testing the number of tree species that were shared between mistletoe species. Simulations were replicated 1000 times.

Interaction frequency and interaction strength were used to investigate the underlying properties of the mistletoe-host network from both perspectives. Interaction frequency is typically applied to the mobile ‘players’ in the network (e.g. bees, flies, birds). Therefore, we retain this method and specifically refer to interaction frequency as the number of mistletoe occurring on each host species within the spatially defined boundary of this study (1800 trees). To avoid confusion, interaction frequency from the hosts perspective is referred to as ‘the number of infected trees’; meaning the total number of trees in the community that have at least one mistletoe present.

Interaction strength was calculated by dividing the frequency of observed interactions by the total number of interactions possible in the network for each mistletoe species. Therefore, interaction strength ranged between zero and one, where zero represents no interaction and one represents complete dependency on one host species. In this study we refer to interaction strength as the proportion of interactions from the mistletoes' perspective. To avoid confusion, interaction strength from the hosts' perspective is referred to as 'host exploitation'. Host exploitation was calculated by dividing the number of infected trees by the sum of all trees that were observed carrying one or more mistletoes. In addition we included another index from the hosts' perspective that would indicate the overall use of trees that were available in the community (Resource exploitation hereafter). Resource exploitation was calculated for each host species by dividing the number of infected trees by the total number of available trees of that species. All indices measuring interaction strength ranged between zero and one; where zero represents no interaction and one represents maximum exploitation in the community. Both resource exploitation and host exploitation are referred to in other fields, such as parasitological studies as 'prevalence' (see Alizer et al. 2007). Here we restrict these terms to follow the ecological literature.

Interaction strength and host exploitation indicate the relative impact between species pairs, while interaction frequency and the number of infected trees indicate the intensity of each interaction (Vázquez et al. 2005). We used least squares regression

to investigate the relationship between network indices and null model simulations to test whether each index was different to random expectations at the community-level. Observed values were compared to the mean value derived from 1000 simulations. All analyses were conducted using students paired t-tests or Wilcoxon signed rank test and data were transformed where necessary for normality.

Host availability hypothesis

We tested whether mistletoes formed dominant interactions with certain host species because they were more prevalent in the community. We did this by quantifying the relationship between each index (described above) and tree availability. Least squares linear regression was used to assess the impact of tree availability and a relationship close to 1 would indicate that tree availability is an important component in promoting favored species interactions.

We compiled data from published literature to test the rate of resource exploitation for different mistletoes around the world. To do this we collated information regarding (i) the number of infected trees, (ii) the number of available trees and (iii) resource exploitation (% of trees infected in the community) of 110 mistletoe-host interactions from 25 publications (including this study; Appendix 2). We analyzed these data using a linear mixed effects model. The model included resource exploitation or the number of infected trees as the dependent variable, tree availability as a covariate and mistletoe species as a random effect. The random effects model was conducted using a Markov Chain Monte Carlo simulation with

15000 simulations after a burnin period of 5000 simulations following an R script published in Moles et al. (2011). Results indicate the effect of tree availability on host exploitation across and within mistletoe species. All data were analyzed using SigmaPlot 11.0 (Systat software, Inc 2008), SPSS 19 and the lme4 package loaded in the R environment 2.11.1 (R Development Core Team 2010).

Results

There were almost twice as many trees in the community ($n = 1800$) as mistletoes ($n = 757$). After removing all tree species devoid of mistletoe, 1298 trees were considered susceptible to infection. Among the trees removed were two dominant taxa, *Eucalyptus camaldulensis* Dehnh. ($n = 231$) and *Myoporum spp.* ($n = 243$). From all available trees, just 14.6 % ($n = 189$) were infected with mistletoe. This was significantly less than predicted from a model that randomly distributed individual mistletoes regardless of species identification (predicted mean = 573.3 ± 9.067 , $z = 42.4$, $p < 0.001$). The dispersion of mistletoes followed a negative binomial distribution (VMR = 11.405) which was significantly different to a Poisson distribution ($X^2 = 289.947$, d.f. = 13, $p < 0.001$). These results show that mistletoes clump together leaving a large number of trees free from infection.

Null model analysis

Results from our null model analysis showed that each index was similar to random expectations at the community level (interaction frequency, $t = 0.544$, d.f. = 17, $p = 0.594$; the number of infected trees, $t = -1.637$, d.f. = 17, $p = 0.12$; interaction

strength, $z = -0.806$, $p = 0.442$; host exploitation, $z = -0.893$, $p = 0.393$; resource exploitation, $z = 0.893$, $p = 0.393$).

Host availability hypothesis

Host exploitation was strongly positively correlated with interaction strength ($r^2 = 0.943$, $F_{1,16} = 266.677$, $p < 0.001$; Figure 1a) and the number of infected trees was strongly positively correlated with interaction frequency ($r^2 = 0.886$, $F_{1,16} = 124.559$, $p < 0.001$; Figure 1b). However, resource exploitation was not correlated with interaction strength ($r^2 = 0.005$, $F_{1,16} = 0.0739$, $p = 0.789$) indicating that network properties were independent of resource availability.

Tree availability was not correlated with interaction strength ($r^2 = 0.0127$, $F_{1,16} = 0.206$, $p = 0.656$), host exploitation ($r^2 = 0.066$, $F_{1,16} = 1.136$, $p = 0.302$), interaction frequency ($r^2 = 0.051$, $F_{1,16} = 0.852$, $p = 0.370$) or the number of infected trees ($r^2 = 0.016$, $F_{1,16} = 0.262$, $p = 0.616$; Figure 2a). However, tree availability was negatively correlated with resource exploitation (Second order polynomial regression; $r^2 = 0.692$, $F_{1,16} = 16.842$, $p < 0.001$; Figure 2b).

Analysis of data from 92 mistletoe-host interactions from the global literature gave similar results to our local study. Tree availability was not correlated with the number of trees infected with mistletoe ($r^2 = 0.253$, $F_{23,85} = 1.192$, $p = 0.275$; Figure 3a). However, tree availability was negatively correlated with resource exploitation (linear regression; $r^2 = 0.635$, $F_{23,85} = 2.565$, $p = 0.001$; Figure 3b). Therefore the patterns we

observed were not simply a unique characteristic of a single community, but a general pattern for mistletoes.

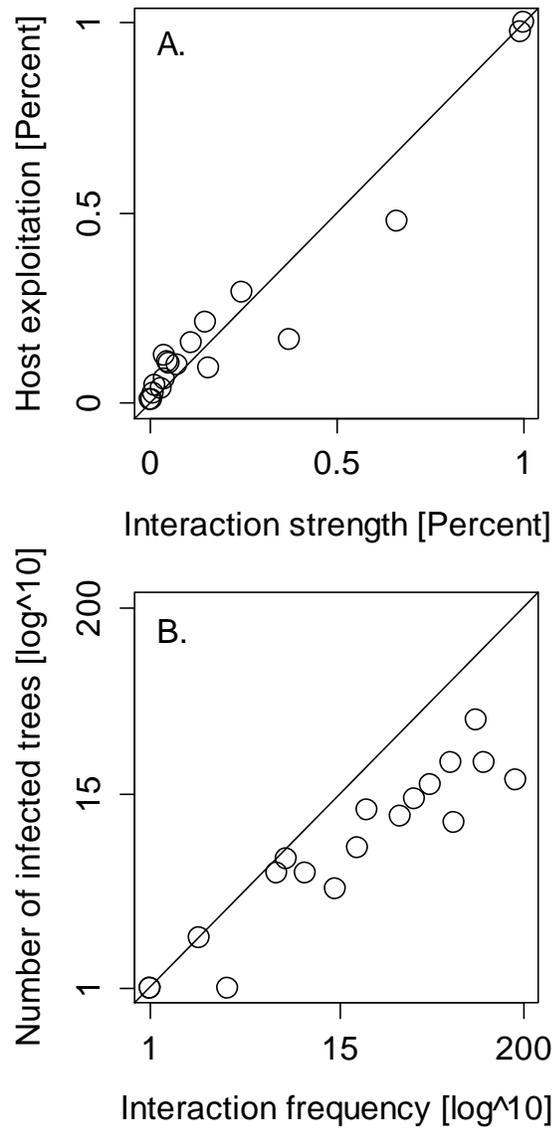


Figure 1 The intensity that each mistletoe species interacted with a host species was proportional to the number of trees that were used (A). In addition, the magnitude of change in interaction frequency was reciprocal to the change in the number of infected trees (B). Note that figure 1b is log transformed on both axis and therefore a power law distribution.

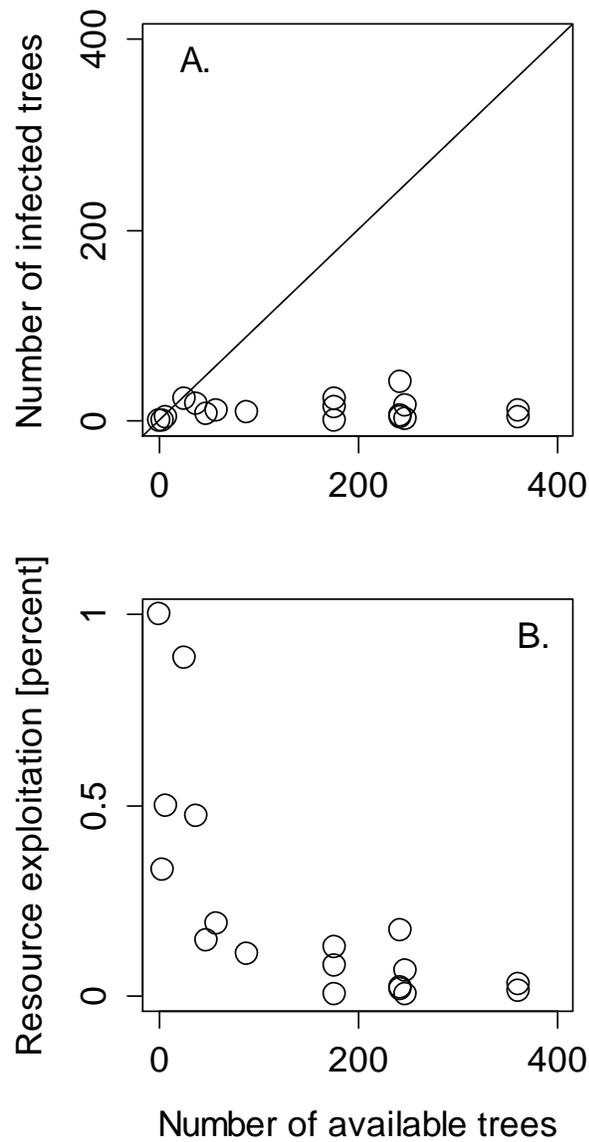


Figure 2 Tree availability does not influence network properties. The number of infected trees did not increase with tree availability (A), while resource exploitation (total number of available trees exploited in the community) declined rapidly with more available trees in the community (B).

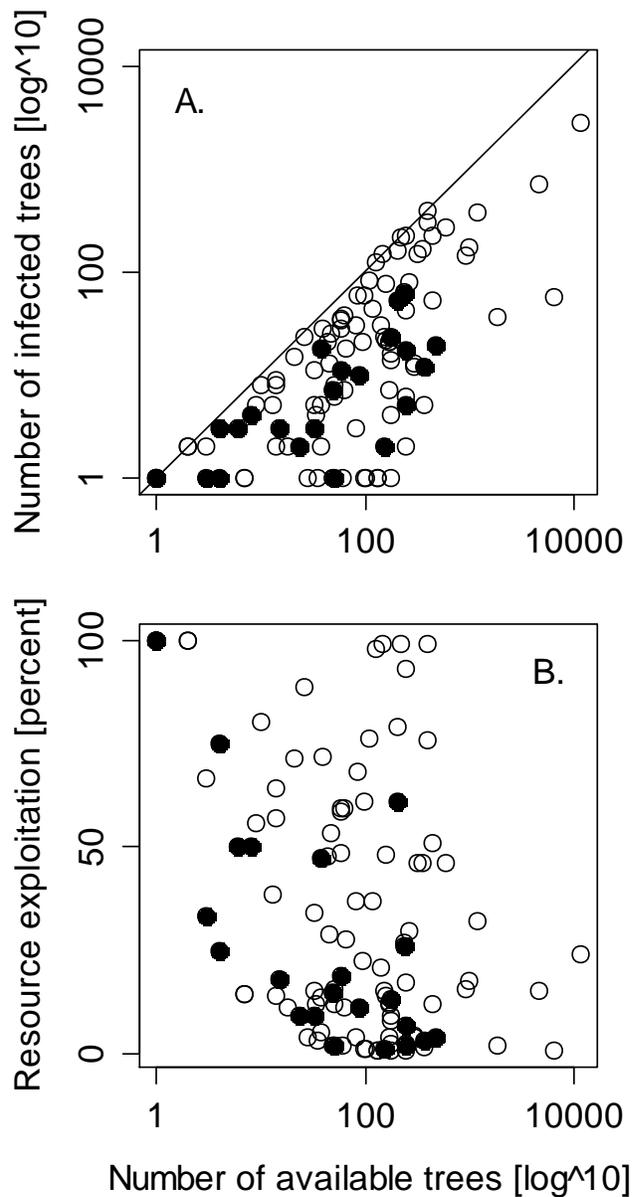


Figure 3 Mistletoe from other genera around the world supported our findings from Fowlers Gap Research Station ($n = 110$ including this study, see appendix 2). The number of infected trees did not increase with tree availability (A), while resource exploitation decreased significantly with tree availability (B). For example, the filled circles represent *Lysiana exocarpi*. Open circles represent all remaining species. The one to one line represents the maximum number of host trees that can be infected. Note the x-axis is a logged and the graph is not isometric.

Discussion

Interaction strength was positively correlated with host exploitation indicating that there was symmetry in species interactions underpinning an ecological network (i.e. negative co-occurrence patterns in a mistletoe-host network). However, interaction strength was not dependent on the number of trees that were available in the community. This is important because it shows for the first time that negative co-occurrence patterns in a mistletoe community – a common structure found in many other ecological communities – are not determined by resource limitation and network properties are maintained through clumping of individual plants. Evaluating resource exploitation between species pairs, will allow other researchers to test the importance of both trophic levels in structuring communities with very different ecological patterns (such as nestedness).

Our null model analyses indicated that interactions were not deterministic when we considered the likelihood of multiple infections. However, this occurred because the community-wise test statistic under represents differences between mistletoes.

Dominant interactions were stronger than predicted by chance, while rare interactions were weaker than predicted by chance, suggesting that the community may be structured by both deterministic and opportunistic interactions. In support of this argument, Morales and Vázquez (2008) showed that spatially restricted movements can change the probability (i.e., an increased number of absent interactions) of interactions occurring in a plant-animal mutualism. That is, dominant interactions between mistletoes and their hosts may represent spatially explicit movements of

fauna (such as frugivorous birds), while rare interactions may be formed through opportunistic associations between frugivorous birds and irregularly visited trees (Carlo and Aukema 2005).

We found strong evidence that mistletoes clump on few trees in the community (Overton 1996, Rist et al. 2010), refuting the host availability hypothesis. However, the observed distribution of mistletoe indicates a strong mechanistic explanation for the structure of this community. Other studies have found that larger trees house higher densities of mistletoes (Reid and Smith 2000, Aukema and Martinez del Rio 2002, Roxburgh and Nicolson 2008), and that spatial movements of frugivores influence dispersal of mistletoe (Murphy et al. 1993, Ladley and Kelly 1996, García et al. 2009, Rawsthorne et al. 2011). It is likely that specific trees are preferentially selected by territorial or breeding birds (Roxburgh and Nicolson 2008, Rawsthorne et al. 2011), allowing neighboring trees to become highly susceptible to infection (Donohue 1995). This mechanistic explanation for clumping is directly relevant to the structure of the community, and subsequently network analysis. We hypothesize that few specific individual trees maintain strong interactions that act as a focal point for infection, facilitating the spread and overall population size of mistletoes.

Understanding the mechanisms that determine network topology is a current focus for many community ecologists and conservation biologists. In order to achieve this goal we argue that resource availability and exploitation needs to be addressed at a population level for each species pair. Temporal changes in resources (Diaz-castelazo

et al 2010, Kaiser et al. 2010, Zahng et al. 2011) and spatially isolated interactions (Burns and Zotz 2010, Blick and Burns 2011) are a pervasive feature of many networks. In other ecosystems, fluctuations in resource availability can occur rapidly over shorter time scales which could determine connectivity and compartmentalization in network structure (Baldock et al. 2011, Diaz-castelazo et al. 2011, Encinas-Viso et al. 2012). This is important because it has wide implications for the processes that determine network structure (e.g. increased probability of intercepting a new network partner). We recommend that resource exploitation and interaction frequency are equally important in determining community organization. However, further studies evaluating resource exploitation from a network perspective are highly desirable to test the generality of these results with other antagonistic interactions.

Overall, in this study we found the surprising result that interaction strength between mistletoes and their hosts were not dependent on the number of trees in the community (resource availability). However, network properties did show strong reciprocal interaction properties indicating that clumping is an important factor in structuring the mistletoe community. We hypothesize that clumping could maintain network interactions if resource availability decreases.

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References

- Altizer, S., Nunn, C.L., and Lindenfors, P. 2007. Do threatened hosts have fewer parasites? A comparative study in primates. *Journal of Animal Ecology* 76: 304-314.
- Aukema, J.E. 2003. Vectors, viscin and Viscaceae: mistletoes as parasites, mutualists, and resources. - *Frontiers in Ecology and the Environment* 1: 212-219.
- Aukema, J.E. & Martinez del Rio, C. 2002. Variation in mistletoe seed deposition: effects of intra- and interspecific host characteristics. - *Ecography* 25: 139-144.
- Baldock, K.C.R. et al. 2011. Daily temporal structure in African savanna flower visitation networks and consequences for network sampling. - *Ecology* 92: 687-698.
- Bascompte, J. et al. 2003. The nested assembly of plant-animal mutualistic networks. - *Proceedings of the National Academy of Sciences USA* 100: 9383-9387.
- Berlow, E.L. et al. 2004. Interaction strengths in food webs: issues and opportunities. - *Journal of Animal Ecology* 73: 585-598.
- Blick, R. & Burns, K.C. 2009. Network properties of arboreal plants: Are epiphytes, mistletoes and lianas structured similarly? - *Perspectives in Plant Ecology, Evolution and Systematics* 11: 41-52.

- Blick, R.A.J. & Burns, K.C. 2011. Liana co-occurrence patterns in a temperate rainforest. – *Journal of Vegetation Science* 22: 868-877.
- Blick, R.A.J. et al. 2012. Predicting network topology of mistletoe-host interactions: do mistletoes really mimic their hosts? – *Oikos* 121: 761-771.
- Burns, K.C. 2007. Network properties of an epiphyte meta-community. - *Journal of Ecology* 95: 1142-1151.
- Burns, K.C. & Zotz, G. 2010. A hierarchical framework for investigating epiphyte assemblages: networks, meta-communities, and scale. – *Ecology* 91: 377-385.
- Blüthgen, N. et al. 2006. Measuring specialization in species interaction networks. - *BMC Ecology* 6: 9.
- Blüthgen, N. et al. 2008. What do interaction network metrics tell us about specialization and biological traits? - *Ecology* 89: 3387-3399.
- Blüthgen, N. 2010. Why network analysis is often disconnected from community ecology: A critique and an ecologist's guide. - *Basic and Applied Ecology* 11: 185-195.
- Carnegie, A.J. et al. 2009. Distribution, host preference, and impact of parasitic mistletoes (Loranthaceae) in young eucalypt plantations in New South Wales, Australia. – *Botany* 87: 49-63.
- Carlo, T.A. & Aukema, J.E. 2005. Female directed dispersal and facilitation between a tropical mistletoe and its dioecious host. - *Ecology* 86: 3245-3251.
- Cayzer, L.W. et al. 2000. Revision of *Pittosporum* (Pittosporaceae) in Australia. - *Australian Systematic Botany* 13: 845-902.

- Cunningham, G.M. et al. 1981. Plants of Western New South Wales. – CSIRO publishing. Press.
- Dormann, C.F. et al. 2009. Indices, Graphs and Null models: Analyzing Bipartite Ecological Networks. - *The Open Ecology Journal* 2: 7-24.
- Donohue, K. 1995. The spatial demography of mistletoe parasitism on a Yemeni Acacia. - *International Journal of Plant Science* 156: 816–823.
- Downey, P.O. 1998. An inventory of host species for each aerial mistletoe species (Loranthaceae and Viscaceae) in Australia. - *Cunninghamia* 5: 685–720.
- Diaz-castelazo, C. et al. 2011. Changes of a mutualistic network over time: reanalysis over a 10-year period. - *Ecology* 91: 793-801.
- Encinas-Viso, F. et al. 2012. Phenology drives mutualistic network structure and diversity. – *Ecology Letters* 15: 198-208.
- García, D. et al. 2009. Seed dispersal by frugivorous marsupial shapes the spatial scale of a mistletoe population. - *Journal of Ecology* 97: 217-229.
- Genini, J. et al. 2011. Mistletoes play different functional roles in a modular host-parasite network. – *Biotropica* 44: 171-178.
- Gotelli, N.J. 2000. Null model analysis of species co-occurrence patterns. - *Ecology* 81: 2606-2621.
- Gotelli, N.J. & Graves, G.R. 1996. Null models in ecology. Smithsonian Institution Press, Washington, D.C.
- Gotelli, N.J. & Ulrich, W. 2011. Statistical challenges in null model analysis. – *Oikos* 121: 171-180.
- Guimarães, P.R. et al. 2007. Interaction intimacy affects structure and coevolutionary

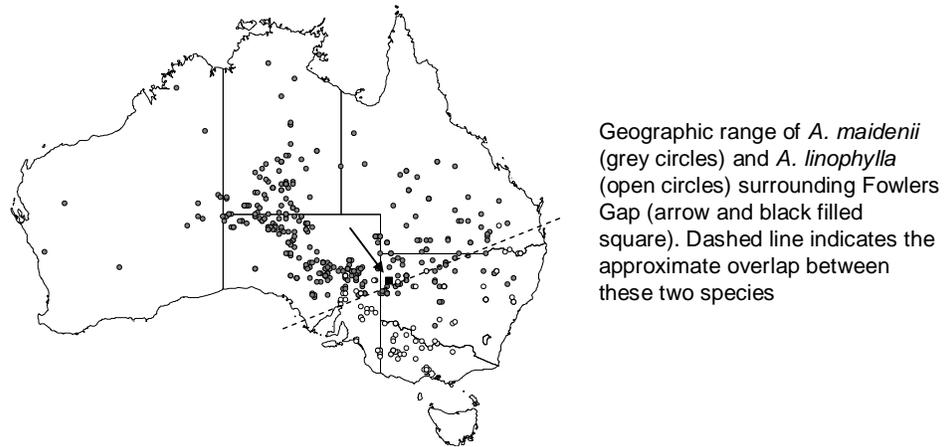
- dynamics in mutualistic networks. - *Current Biology* 17: 1797-1803.
- Hoffman, A.J. et al. 1986. *Tristerix tetrandrus* (Loranthaceae) and its host-plants in the Chilean matorral: patterns and mechanisms. – *Oecologia* 2: 202-206.
- Ings, T.C. et al. 2009. Ecological networks – beyond food webs. *Journal of Animal Ecology* 78: 253-269.
- Jordano, P. 1987. Patterns of mutualistic interactions in pollination and seed dispersal: connectance, dependence asymmetries, and coevolution. - *American Naturalist* 129: 657-677.
- Kaiser, C.N. et al. 2010. The robustness of pollination networks to the loss of species and interactions: a quantitative approach incorporating pollinator behaviour. - *Ecology Letters* 13: 442-452.
- Kavanagh, P. & Burns, K.C. In press. Mistletoe macroecology: spatial patterns in species diversity and host use across Australia. - *Biological Journal of the Linnean Society*.
- Krishna, A. et al. 2008. A neutral-niche theory of nestedness in mutualistic networks. - *Oikos* 117: 1609-1618.
- Ladley, J.J. & Kelly, D. 1996. Dispersal, germination and survival of New Zealand mistletoes (Loranthaceae): dependence on birds. - *New Zealand Journal of Ecology* 20: 69-79.
- Lavorel, S. et al. 1999. Spread of mistletoes (*Amyema preissii*) in fragmented Australian woodlands: a simulation study. - *Landscape Ecology* 14: 147-160.

- László, Z. & Tóthmérész, B. 2011. High host plant aggregation involves uniform gall distribution and high prevalence: the case of wild roses and Bedeguar gall wasps (*Diplolepis rosae*). – *North-Western Journal of Zoology* 7: 112-117.
- Lehsten, V. & Harmand, P. 2006. Null models for species co-occurrence patterns: assessing bias and minimum iteration number for the sequential swap. - *Ecography* 29: 786-792.
- Nash, R. & Keegan, B.F. 2006. Characteristics of the association between the marine copepod, *Gastrodelpys clausii*, and its fanworm host *Bispira volutacornis*. - *Helgoland Marine Research* 60: 32-38.
- Mathiasen, R.L. 2008. Mistletoes: Pathology, systematics, ecology, and management. - *Plant Disease* 92: 988-1006.
- Morales, J.M. and Vázquez, D.P. 2008. The effect of space in plant-animal mutualistic networks: insights from a simulation study. – *Oikos* 117: 1362-1370.
- Moles, A.T. et al. 2011. Putting plant resistance traits on the map: a test of the idea that plants are better defended at lower latitudes. - *New Phytologist* 191: 777-788.
- Murphy, S.R. et al. 1993. Differential passage time of mistletoe fruits through the gut of honeyeaters and flowerpeckers: effects of seedling establishment. - *Oecologia* 93: 171-176.
- Norton, D.A. & Carpenter, M.A. 1998. Mistletoes as parasites: host specificity and speciation. - *Trends in Ecology and Evolution* 13: 101-105.
- Overton, J. 1996. Spatial autocorrelation and dispersal in mistletoes: field and simulation results. - *Plant Ecology* 125: 83-98.

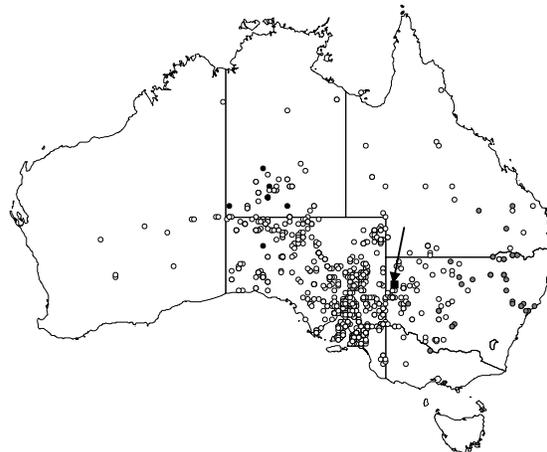
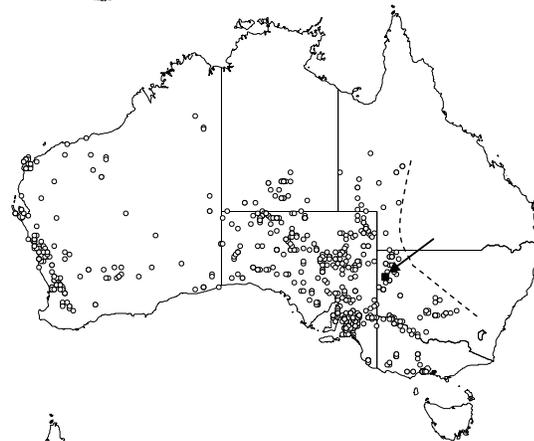
- Pires, M.M. et al. 2011. The nestedness assembly of individual-resource networks. – *Journal of Animal Ecology* 80: 896-903.
- R Development Core Team (2010) R: a language and environment for statistical computing. Vienna, Austria. <http://www.R-project.org>
- Rawsthorne, J. et al. 2011. Implications of movement patterns of a dietary generalist for mistletoe seed dispersal. - *Austral Ecology* 36: 650-655.
- Rist, L. et al. 2011. The spatial distribution of mistletoe in a southern Indian tropical forest at multiple scales. – *Biotropica* 43:50-57.
- Reid, N. & Smith, S.S. 2000. Population dynamics of an arid zone mistletoe (*Amyema preissii*, Loranthaceae) and its host *Acacia victoriae* (Mimosaceae). - *Australian Journal of Botany* 48: 45-58.
- Rodl, T. & Ward, D. 2002. Host recognition in desert mistletoe: early stages of development are influenced by substrate and host origin. - *Functional ecology* 16: 128-134.
- Roxburgh, L. & Nicolson, S.W. 2008. Differential dispersal and survival of an African mistletoe: does host size matter? - *Plant Ecology* 195: 21-31.
- Stone, L. & Roberts, A. 1990. The checkerboard score and species distributions. - *Oecologia* 85: 74-79. IBM SPSS Statistics 20.0.0 for windows. IBM corp.
- Thompson, J.N. 2005. The Geographic Mosaic of Coevolution. - University of Chicago. Press.
- Ulrich, W. & Gotelli, N.J. 2007. Disentangling community patterns of nestedness and species co-occurrence. - *Oikos* 116: 2053-2061.
- Vázquez, D.P. et al. 2005. Interaction frequency as a surrogate for the total effect of

- animal mutualists on plants. – *Ecology Letters* 8: 1088-1094.
- Vázquez, D.P. et al. 2009a. Uniting pattern and process in plant-animal mutualistic networks: a review. - *Annals of Botany* 103: 1445-1457.
- Vázquez, D.P. et al. 2009b. Evaluating multiple determinants of the structure of plant-animal mutualistic networks. - *Ecology* 90: 2039-2046.
- Vázquez, D.P. et al. In press. The strength of plant-pollinator interactions. – *Ecology* [doi:<http://dx.doi.org/10.1890/11-1356.1>].
- Walker, M.A. et al. 2010. Trickle or clumped infection process? A stochastic model for the infection process of the parasitic roundworm of humans, *Ascaris lumbricoides*. – *International Journal of Parasitology* 40: 1381-1388.
- Watson, D.M. 2002. Effects of mistletoe on diversity: a case-study from southern New South Wales. - *Emu* 102: 275-281.
- Watson, D.M. 2009a. Parasitic plants as facilitators: more dryad than Dracula. – *Journal of Ecology* 97: 1151-1159.
- Watson, D.M. 2009b. Determinants of parasitic plant distribution: the role of host quality. - *Botany* 87: 16-21.
- Yan, Z. 1990. Host specificity of *Lysiana exocarpi* subsp. *Exocarpi* and other mistletoes in southern South Australia. – *Australian Journal of Botany* 38: 475-486.
- Zahng, F. et al. 2011. An interaction switch predicts the nested architecture of mutualistic networks. – *Ecology Letters* 14: 797-803.

Appendix 1 Geographic range of mistletoes found growing at Fowlers Gap. All data points are from Australian Virtual Herbarium (AVH) and filtered to only include coordinates with an accuracy of less than 10 km. Interpretations of geographic range are gleaned from the available herbarium records only. Geographic range is influenced by sample size. As we did not account for this, we caution extensive interpretations. Three maps were used for clarity.



Geographic range of *A. preissii* (open circles) surrounding Fowlers Gap (arrow and black filled square). Dashed line indicates the approximate boundary of this species.



Geographic range of *Lysiana exocarpi* subsp. *exocarpi* (open circles), *L. exocarpi* subsp. *tenuis* (grey circles) and *L. exocarpi* subsp. *diamantinensis* (black circles) surrounding Fowlers Gap (arrow and black filled square).

Appendix 2 Collated data for all known mistletoe-host interactions in the literature. Data also includes this study. Resource exploitation and the number of infected trees were calculated when necessary to complete the table. Area of each study is included or though not utilized in this study. Some studies did not investigate a defined area; rather they have surveyed a set number of trees, as we did in this study. Table arranged alphabetically for mistletoe species.

Mistletoe species	Location	Area (ha)	Number of trees infected with mistletoe	Number of available trees	Resource Exploitation (%)	Host tree species	Source
<i>Amyema linophylla</i>	Australia	0.43	2	247	0.8	<i>Casuarina pauper</i>	This study
			6	51	12.0	<i>Casuarina cristata</i>	Yan (1990)
<i>Amyema maidenii</i>	Australia		42	242	17.4	<i>Acacia aneura</i>	This study
			1	176	0.6	<i>Acacia tetragonophylla</i>	
<i>Amyema miquelii</i>	Australia	0.43	23	151	15.0	<i>Eucalyptus oleosa</i>	Yan (1990)
		0.43	9	14	64.0	<i>Eucalyptus gracilis</i>	
		11.20	383	1200	31.9	<i>Eucalyptus fasciculosa</i>	Ward (2005)
		1.40	57	6427	0.9	<i>Eucalyptus microcarpa</i>	MacRaid et al. (2009)
<i>Amyema miraculosum</i>	Australia	0.43	64	238	27.0	<i>Myoporum platycarpum</i>	Yan (1990)
<i>Amyema preissii</i>	Australia		5	362	1.4	<i>Acacia victoriae</i>	This study
			6	242	2.5	<i>Acacia aneura</i>	
			14	176	8.0	<i>Acacia tetragonophylla</i>	

			23	26	88.5	<i>Acacia loderi</i>	
		14.60	148	321	46.1	<i>Acacia victoriae</i>	Reid and Smith (2000)
		0.43	2	18	11.0	<i>Acacia collectoides</i>	Yan (1990)
		0.43	1	98	1.0	<i>Acacia papyrocarpa</i>	
		0.43	2	14	14.0	<i>Cassia nemophylla</i>	
<i>Amyema preissii</i>	Australia	0.43	11	32	34.0	<i>Acacia nyssophylla</i>	Yan (1990)
<i>Amyema quandang</i>	Australia	0.43	60	98	61.0	<i>Acacia papyrocarpa</i>	Yan (1990)
		2.50	58	85	68.0	<i>Acacia papyrocarpa</i>	Reid and Lange (1988)
<i>Amyema quandang</i>	Australia	2.50	1	61	2.0	<i>Lysiana exocarpis</i>	Reid and Lange (1988)
<i>Arceuthobium abietinum</i>	USA	1.68	159	201	79	<i>Picea breweriana</i>	Mathiasen (2011)
<i>subsp. wiensii</i>		1.68	147	148	99	<i>Tsuga mertensiana</i>	
		1.68	30	81	37	<i>Pinus monticola</i>	
<i>Arceuthobium monticola</i>	USA	1.68	388	392	99	<i>Picea breweriana</i>	Mathiasen (2011)
		1.68	225	242	93	<i>Abies magnifica</i>	
		1.68	166	361	46	<i>Abies concolor</i>	
		1.68	1.02	102	1	<i>Pinus monticola</i>	
<i>Arceuthobium tsugense</i>	USA	1.68	218	220	99	<i>Picea breweriana</i>	Mathiasen (2011)
<i>subsp. mertensianne</i>		1.68	126	129	98	<i>Pinus monticola</i>	
		1.68	1	35	3	<i>Pinus lambertiana</i>	
<i>Lysiana exocarpis</i>	Australia		12	362	3.3	<i>Acacia victoriae</i>	This study
			17	247	6.9	<i>Casuarina pauper</i>	
			5	242	2.1	<i>Acacia aneura</i>	
			23	176	13.1	<i>Acacia tetragonophylla</i>	
			10	89	11.2	<i>Acacia salacina</i>	
			11	58	19.0	<i>Santalum sp.</i>	
			7	48	14.6	<i>Eremophila longifolia</i>	
			18	38	47.4	<i>Alectryon oleiofolium</i>	

			4	8	50.0	<i>Eremophila alternifolia</i>	
			1	3	33.3	<i>Pittosporum angustifolia</i>	
			1	1	100.0	<i>Cassia sturtii</i>	
		2.70	2	23	9.0	<i>Heterodendrum oleifolium</i>	Reid and Lange (1988)
		2.80	3	6	50.0	<i>Myoporum platycarpum</i>	
		2.90	1	1	100.0	<i>Eremophila oppositifolia</i>	
<i>Lysiana exocarpi</i>	Australia	3.00	19	473	4.0	<i>Amyema quandang</i>	
		0.43	1	4	25.0	<i>Acacia anuera</i>	Yan (1990)
		0.43	3	32	9.0	<i>Acacia nyssophylla</i>	
		0.43	1	51	2.0	<i>Casaurina cristata</i>	
		0.43	2	151	1.0	<i>Eucalyptus oleosa</i>	Yan (1990)
		0.43	3	15	18.0	<i>Exocarpus aphyllus</i>	
		0.43	52	207	61.0	<i>Heterodendrum oleifolium</i>	
		0.43	62	238	26.0	<i>Myoporum platycarpum</i>	
		0.43	3	4	75.0	<i>Pittosporum phillyeoides</i>	
<i>Phcosepalus kalachariensis</i>	Zambia	20.00	16	177	9.0	<i>Acacia polycantha</i>	Roxburgh and Nicolson (2005)
		20.00	7	172	4.1	<i>Acacia sieberana</i>	
		20.00	12	299	4.0	<i>Dichrostachys cinerea</i>	
		20.00	1	132	0.8	<i>Ziziphus abyssinica</i>	
<i>Phoradendron californicum</i>	Mexico	1.55	83	111	76.0	<i>Ceridium microphyllum</i>	Overton (1996)
	USA	0.92	21	93	22.6	<i>Ceridium microphyllum</i>	Aukema and Matinez del Rio (2002)
		0.92	5	37	13.5	<i>Acacia constricta</i>	
		0.92	15	21	71.4	<i>Olneya tesota</i>	
			2834	11808	24.0	<i>Prosopis velutina</i>	Aukema (2004)
<i>Phragmanthera dschallensis</i>	Zambia	20.00	21	172	12.0	<i>Acacia sieberana</i>	Roxburgh and Nicolson (2008)
		20.00	4	177	2.3	<i>Acacia polycantha</i>	Roxburgh and Nicolson (2005)

		20.00	21	172	12.2	<i>Acacia sieberana</i>	
		20.00	4	34	11.8	<i>Albizia harveyi</i>	
		20.00	13	299	4.3	<i>Dichrostachys cinerea</i>	
		20.00	5	13	38.5	<i>Faidherbia albida</i>	
		20.00	5	9	55.6	<i>Lonchocarpus capassa</i>	
		20.00	1	7	14.3	<i>Combretum fragans</i>	
		20.00	1	48	2.1	<i>Combretum molle</i>	
		20.00	1	7	14.3	<i>Ficus sycamorus</i>	
		20.00	1	132	0.8	<i>Ziziphus abyssinica</i>	
<i>Phrygilanthus sonorae</i>	Mexico	1.55	30	144	21.0	<i>Bursera microphylla</i>	Overton (1996)
<i>Plicosephalus curviflorus</i>	Yemen	1.50	76	159	48.0	<i>Acacia tortilis</i>	Donohue (1995)
<i>Psittacanthus plagiophyllus</i>	Brazil	4.50	44	118	37	<i>Anacardium occidentale</i>	Fadini et al. (2010)
<i>Psittacanthus robustus</i>	Brazil	8.00	143	923	15.5	<i>Qualea grandiflora</i>	Monteiro et al. (1992)
		8.00	5	33	15.2	<i>Qualea multiflora</i>	
		8.00	13	45	28.9	<i>Vochysia cinnamomea</i>	
		8.00	36	1872	1.9	<i>Miconia albicans</i>	
		2.82	79	267	29.6	<i>Vochysia thyrsoidea</i>	Teodoro et al. (2010)
<i>Psittacanthus schiedeanus</i>	Mexico	1.00	28	39	71.8	<i>Liquidambar styraciflua</i>	de Buen et al. (2002)
		1.00	21	44	47.7	<i>Liquidambar styraciflua</i>	
		1.00	34	58	58.6	<i>Liquidambar styraciflua</i>	
		1.00	35	59	59.3	<i>Liquidambar styraciflua</i>	
<i>Struthanthus aff. polyanthus</i>	Brazil	1.30	1	1	100.0	<i>Bauthinia sp.</i>	Arruda et al. (2006)
		1.30	2	2	100.0	<i>Cabralea canjerana</i>	
		1.30	2	38	5.3	<i>Caryocar brasiliense</i>	
		1.30	2	3	66.7	<i>Dalbergia violacea</i>	
		1.30	8	51	15.7	<i>Dimorphandra mollis</i>	

		1.30	2	2	100.0	<i>Hymenaea courbaril</i>	
		1.30	25	47	53.2	<i>Kielmeyera coriacea</i>	
		1.30	8	10	80.0	<i>Plathymenia reticulata</i>	
		1.30	28	58	48.3	<i>Pouteria ramiflora</i>	
		1.30	38	64	59.4	<i>Styrax ferrugineus</i>	
		1.30	1	1	100.0	<i>Leguminosa indet 1</i>	
		1.30	1	1	100.0	<i>Leguminosa indet 2</i>	
<i>Taxillus tomentosus</i>	India	3.00	227	445	51.0	<i>Phyllanthus emblica</i> & <i>P. indofischeri</i>	Rist et al. (2010)
<i>Tristerix aphyllus</i>	Chile	3.00	270	588	46.0	<i>Echinopsis chilensis</i>	Medel et al. (2004)
<i>Tristerix aphyllus</i>	Chile		707	4711	15.0	<i>Echinopsis acida</i> & <i>E. skottsbergii</i>	Martinez del Rio et al. (1996)
<i>Tristerix corymbosus</i>	Argentina	3.00	174	993	17.6	<i>Aristotelia chilensis</i>	García et al. (2009)
		3.00	53	446	11.9	<i>Azara microphylla</i>	
		3.00	7	62	11.2	<i>Maytenus boaria</i>	
<i>Tristerix tetrandrus</i>	Chile	1.50	22	160	14.0	<i>Kageneckia oblonga</i>	Hoffman et al. (1986)
		1.50	3	83	4.0	<i>Colliguaya odorifera</i>	
		1.50	1	28	4.0	<i>Talguenea quinquinervia</i>	
<i>Viscum album</i>	Iran	3.00	306	403	75.9	<i>Parrotia persica</i>	Kartoolinejad et al. (2007)
		3.00	18	65	27.7	<i>Carpinus betulus</i>	
		3.00	8	14	57.1	<i>Populus caspica</i>	

Supplementary references (Appendix 2)

- Arruda, R. et al. 2006. Host specificity of a Brazilian mistletoe, *Struthanthus* aff. *polyanthus* (Loranthaceae), in cerrado tropical savanna. – *Flora* 201: 127–134.
- Aukema, J. E. 2004. Distribution and dispersal of desert mistletoe is scale-dependent, hierarchically nested. - *Ecography* 27: 137–44.
- Aukema, J. E. & Martinez del Rio C. 2002. Where does a fruit-eating bird deposit mistletoe seeds? Seed deposition patterns and an experiment. - *Ecology* 83: 3489–96.
- de Buen, L.L. et al. 2002. Mistletoe infection of trees located at fragmented forest edges in the cloud forests of Central Veracruz, Mexico. – *Forest Ecology and Management* 164: 293-302.
- Donohue, K. 1995. The spatial demography of mistletoe parasitism on a Yemeni *Acacia*. – *International Journal of Plant Sciences* 156: 816–23.
- Fadini, R.F. et al. 2010. Consistency in seed-deposition patterns and the distribution of mistletoes among its host trees in an Amazonian savanna. - *Australian Journal of Botany* 57: 640-646.
- García, D. et al. 2009. Seed dispersal by frugivorous marsupial shapes the spatial scale of a mistletoe population. - *Journal of Ecology* 97: 217-229.
- Hoffman, A.J. et al. 1986. *Tristerix tetrandrus* (Loranthaceae) and its host-plants in the Chilean matorral: patterns and mechanisms. – *Oecologia* 69: 202-206.
- Kartoolinejad, D. et al. 2007. The Relationship among Infection Intensity of *Viscum album* with some Ecological Parameters of Host Trees. *International Journal of Environmental Research* 1: 143-149.
- Overton, J. 1996. Spatial autocorrelation and dispersal in mistletoes: field and simulation results. - *Plant Ecology* 125: 83-98.

- Mathiasen, R. 2011. Susceptibility of Conifers to Three Dwarf Mistletoes in the Klamath-Siskiyou Mountains. – *Western Journal of Applied Forestry* 26: 13-18.
- MacRaid, L.M. et al. 2009. Box Mistletoe (*Amyema miquelii*) parasitism is not detrimental to the health of Grey Box (*Eucalyptus microcarpa*) trees at a regional scale. – *Ecological Management and Restoration* 10: 148-150.
- Martínez del Rio, C. et al. 1996. Seed dispersers as disease vectors: bird transmission of mistletoe seeds to plant hosts. - *Ecology* 77: 912–921.
- Medel, R. et al. 2004. Effects of vector behavior and host resistance on mistletoe aggregation. - *Ecology* 85: 120–6.
- Monteiro, R. F. et al. 1992. Host specificity and seed dispersal of *Psittacanthus robustus* (Loranthaceae) in south-east Brazil. – *Journal of Tropical Ecology* 8: 307–14.
- Reid, N. and Smith, S.S. 2000. Population dynamics of an arid zone mistletoe (*Amyema preissii*, Loranthaceae) and its host *Acacia victoriae* (Mimosaceae). - *Australian Journal of Botany* 48: 45-58.
- Reid, N. & Lange, R.T. 1988. Host Specificity, Dispersion and Persistence Through Drought of Two Arid Zone Mistletoes. – *Australian Journal of Botany* 36: 299-313.
- Rist, L. et al. 2011. The spatial distribution of mistletoe in a southern Indian tropical forest at multiple scales. – *Biotropica* 43:50-57.
- Roxburgh, L. & Nicolson, S.W. 2005. Patterns of host use in two African mistletoes: the importance of mistletoe-host compatibility and avian disperser behaviour. - *Functional Ecology* 19: 865–873.
- Roxburgh, L. & Nicolson, S.W. 2008. Differential dispersal and survival of an African mistletoe: does host size matter? - *Plant Ecology* 195: 21-31.
- Teodoro, G.S. et al. 2010. How does a *Psittacanthus robustus* Mart. population

structure relate to a *Vochysia thyrsoidea* Pohl. host population? – *Flora* 205: 797-801.

Ward, M. J. 2005. Patterns of box mistletoe *Amyema miquelii* infection and pink gum *Eucalyptus fasciculosa* condition in the Mount Lofty Ranges, South Australia. – *Forest Ecology and Management* 213: 1–14.

Yan, Z. 1990. Host specificity of *Lysiana exocarpi* subsp. *Exocarpi* and other mistletoes in southern South Australia. – *Australian Journal of Botany* 38: 475-486.

Chapter Four

Does abundance, species turnover or climbing strategy
predict connectivity in a vine-host network?

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“We habitually underestimate the effects of randomness... In the political world, the economic world, the business world – chance events are often conspicuously misinterpreted as accomplishments or failures.”

Quote from Leonard Mlodinow (2008), *The Drunkards Walk*, pp 11.

This paper was conceived, analysed and written by RB. Vine-host data were contributed by RG. Comments on an early draft were provided by all co-authors.

Abstract

Ecological networks are now widely adopted to evaluate species interactions and organisational complexity of communities in different ecosystems across different continents. In this study we focus on two published articles in *Perspectives in Ecology, Evolution and Systematics* which show contrasting network properties of vine-host interactions (i.e., nestedness and negative co-occurrence patterns). In an attempt to resolve these differences we ask whether vine-host interactions follow a third, and otherwise untested hypothesis; that connectivity is determined by opportunistic pairwise associations in the landscape. We evaluated abundance, geographic area, species turnover and climbing strategy as ecological processes underpinning seemingly disparate network patterns. In addition we used a modified null model to account for spatial constraints (i.e., only neighbouring trees were available in the model) and we evaluated host specialisation in light of stand level diversity. Even after we accounted for tree availability, our vine-host network containing 48 vine species and 79 host tree species, observed in nine tropical rainforests along eastern Australian coastline, had a higher number of checkerboard pairs, was more specialised with fewer interactions than predicted by chance (i.e., negative co-occurrence patterns). Overall results indicate that vine-host interactions can form both positive and negative co-occurrence patterns found previously. Network connectivity was largely determined by abundance and geographic distribution, or though scrambling vines did have a higher rate of connectivity over other growth strategies when they were low in abundance.

Introduction

Identifying structure in biological communities has remained a hot topic in ecology for over a century (Clements 1916, Gleason 1917, Diamond 1975, Connor and Simberloff 1979, Stone and Roberts 1990, Gotelli and Graves 1995, Keddy and Weiher 1999, Gotelli 2000, Hubbell 2001, Leibold et al. 2004, Hausdorf and Hennig 2007, Ulrich and Gotelli 2011). Over the last decade, network theory has been applied to ecological systems to understand species interactions in a community (Jordano 1987, Bascompte et al. 2003, Proulx et al. 2005, Guimarães et al. 2007, Vázquez 2009a, Vázquez et al. 2009b, Ings et al. 2009, Blüthgen 2010, Dormann 2009). A primary advantage of using the network approach is the ability to track the connectivity between organisms by mapping interactions; thereby establishing the role of specialization, vulnerability to disturbance and the introduction of foreign species. Importantly, many species interactions share organizational properties depicted by network analysis, such as nested patterns of plant-pollinator (Bascompte et al. 2003, Stang et al. 2006), clownfish–anemone (Ollerton et al., 2007), ant–plant (Guimarães et al. 2006) and fruit–frugivore mutualisms (Lázaro et al., 2005).

Recently, ecological networks have been applied to plant-plant interactions, including mistletoes, epiphytes and vines (Burns 2007, Blick and Burns 2009, Silva et al. 2010, Blick et al. 2011, Sfair et al. 2011, Piazzon et al. 2011, Genini et al. 2012, Martos et al. 2012). Current research suggests that different plant guilds have different network properties (as displayed by the mistletoes, lianas and epiphytes described in Blick and Burns, 2009). It is hypothesised that plant-plant interactions have different network properties (topology) because each guild has evolved different growth strategies (i.e. parasitism and commensalism). For example, Burns (2007) hypothesised that nested

topology of epiphyte-host interactions were determined by the colonisation of clump forming species and the successional accumulation of species less tolerant to water and nutrient limitations in an unoccupied tree canopy. Similarly, Blick and Burns (2011) hypothesised that a checkerboard distribution (negative co-occurrence) was determined by differences in environmental conditions across two areas of a single wildlife reserve. Both hypotheses remain untested; however network analysis has revealed differences in organisational complexity requiring further investigation.

The focus of this study is to identify what mechanisms generate disparate network properties of vine-host interactions revealed in two published articles from the seminal journal *Perspectives in Ecology, Evolution and Systematics* (Blick and Burns 2009, Sfair et al. 2011). Both studies used the same method for analysing network interactions and yet revealed contrasting results. Blick and Burns (2009) found negative co-occurrence patterns (i.e., some trees are only inhabited by certain vines), while Sfair et al. (2011) found positive nested patterns (i.e., trees accumulate vine species). Further evaluation of vine-host networks is necessary to establish how organisational complexity varies across communities, ecosystems and continents. In this study we empirically test the role of stochastic co-occurrence at broad spatial scales.

Vines are recognised globally as important components of forest structure (Schnitzer 2005, Schnitzer and Bongers 2002). In order to ascend the forest interior, vines have evolved a wide variety of climbing strategies including hooks that grapple branches, adventitious roots that attach to bark, and twining or coiling around trunks (Putz 1984, Balfour & Bond 1993, Gallagher & Leishman 2012). In addition, vines grow in a multitude of ecosystems, including coastal dune systems, low growing heath

communities and tropical rainforests (Schnitzer and Bongers 2002), and include a wide range of growth forms, such as scandent shrubs capable of surviving without host structural support, twinning petioles and knot forming stems.

The habit of vines (Campbell & Newbery 1993, DeWalt et al. 2006, Burnham 2002, Malizia et al. 2010) leads to two plausible hypotheses: (1) that vines have preferential selection for host species based on their functional attributes such as bark type or stem diameter (i.e., interactions between vines and their hosts will be deterministic), or (2) that vines have opportunistic host selection within spatially explicit habitats (i.e., interactions between vines and host trees are random). A reasonable extension of the second hypothesis follows that spatial constraints influence null model outcomes (Roxburgh et al. 1998, Roxburgh et al. 1999, Peres-Neto et al. 2001) because the probability of interacting with different trees is greatly reduced when the number of shared locations is low (Morales and Vázquez 2008, Gotelli and Ulrich 2012). Therefore, we predict that an ecological network containing vines and trees are more connected when vines are abundant, cover a wider area and interact with a greater number of trees.

The arrangement of interactions in an ecological network is derived from spatially distinct assemblages of plants that have adapted to local environmental conditions (Dalling et al. in press). Including patch dynamics (*censu* Leibold et al. 2004) into bipartite network analysis may reveal disparate interaction properties found in previous work. In this study, we used a large vine-host network spanning nine forests along the east coast of New South Wales, Australia to evaluate whether vine-host interactions were determined by stochastic processes in the landscape.

Specifically, we tested the following hypotheses:

1. Network connectivity is correlated with tree availability
2. Connectivity is predicted by abundance, area and the number of interactions
 - (i) Abundance: due to a higher probability of interacting with rare species
 - (ii) Geographic area: by increasing the probability of intercepting new species
 - (iii) The number of interactions: due to non-specific selection of host trees.
3. Vegetation dynamics do not predict species connectivity
 - (i) Species diversity: due to a higher probability of interacting with new species
 - ii) Exploitation: by maximising interactions with individuals

Methods

Species and study location

Vine-host interactions including 48 vine species and 79 host trees were recorded in nine littoral rainforest sites in eastern Australia (28°S and 36°S) by Rachael V. Gallagher. A low canopy height (generally < 25 m) allowed species identification of canopy lianas from the ground. All species interactions were observed in 88 30 x 10 m plots (300 m²) using binoculars. We included all vines originating inside the quadrat and interacting with host trees within the perimeter of the quadrat. We excluded from analysis: 1) the number of times individual plants interacted together and 2) connections between multiple host species that may form canopy ‘bridges’. All plots were summed to generate a single quantitative interaction network covering the landscape (Figure 1). Mean annual temperature and annual precipitation vary between 19.9–15.3°C and 1813–958 mm respectively in a North to South direction. A detailed description of the study sites and species can be found in Gallagher and Leishman (2012).

Network analysis

We used five commonly employed network indices because they cover a wide range of properties from web specialisation to nestedness: (1) Nestedness was evaluated using the BINMATNEST algorithm to generate an index that ranges between zero and 100 (i.e., temperature). The BINMATNEST algorithm was because it is similar to *NESTEDNESS* (Ulrich 2006) which alleviates several problems from the *NTC* (Atmar and Patterson 1995). Temperature tests whether host trees with species poor vine communities form a subset of trees within species rich vine communities. (2) Web specialisation was evaluated using the H2 function to generate a normalised index ranging between zero and one where one is more specialised. Web specialisation tests whether the number of links between species is less than expected after considering the abundance of each species. (3) A checkerboard distribution was evaluated using a normalised c-score function that ranges between zero and one. C-score tests the number of times each host tree species carries a specific set of vines. (4) Compartmentalisation tests whether groups of vines co-occur on subsets of trees in the community. (5) Connectivity is calculated as the number of pairwise associations formed in each network and expressed as a percentage of possible links that could be formed given the number of species observed. Connectivity is evaluated from the perspective of vines and host trees to calculate the relationship between connectivity with abundance, area and interaction intensity. A full description of each metric can be found elsewhere and not repeated here in detail (Blüthgen 2010).

All metrics were analysed using a null model approach; where observed indices were compared to a distribution of expected values in a random community (Gotelli 2000).

Each observed index was analysed using a standardised z-score; such that the mean is centered at zero and the deviation of the observed value is measured as the number of standard deviations away from the mean. During each simulation all interactions were shuffled within the matrix allowing marginal row totals to vary while fixing marginal column totals. These constraints conform to a biologically intuitive randomisation process where a vine species may select a different tree species in the randomised community, but the vine cannot become a different species. All indices were calculated using the 'bipartite' function and observed values were compared to 1000 simulations generated using the 'permatfull' function from the vegan package loaded in the R environment v. 2.15.1 (R code provided in Appendix 1).

Scaling species interactions with tree availability

Landscape-level connectivity is defined by the number of species interactions divided by the number of potential species interactions. That is, all spatial constraints in the landscape are omitted during null model analysis. To calculate patch-level connectivity we included spatial constraints such that species could only interact if they occurred in the same plot. Importantly, this approach retained information on the abundance of each species in the focal region. All simulations at the landscape and patch level were carried out using a with replacement method of randomisation to include the possibility that individual trees carried multiple vines.

Finally, landscape and patch-level connectivity were compared using a single sample t-test to evaluate whether landscape-level connectivity minus patch-level connectivity was different to zero. We used least squares linear regression to test parameters predicted to (i) increase connectivity across the landscape including abundance, area

and the number of interactions, and (ii) increase connectivity within patches including diversity (Shannon-Weiner index) and the proportion of individuals that were exploited.

Species accumulation and species turnover

Species accumulation and the acquisition of novel interactions were evaluated using rarefaction curves (vegan package). Each rarefaction curve was generated using 1000 simulations and 95 % confidence intervals. We used a fitted spline curve and the 'predict' function (stats package) to calculate the required number of observations needed to record an additional species. The predict function approximates the number of observations required to find a known Y value using a linear progression from the final set of data points. In this study we consider a linear progression satisfactory because we are characterising the final shape of each curve and the relative difference between the higher and lower trophic levels in each network. Typically, species accumulation curves are restricted to evaluating species richness. Here we extended this approach to evaluate novel interactions. All analyses were calculated using the R environment (R core team, 2012).

Results

The vine network contained 1126 interactions between 48 vine species and 79 host tree species (Figure 1). The final interaction matrix had 11.63 % of all possible interactions ($n = 441$ of a possible 3972). Each forest ($n = 9$) had between 45 and 250 interactions (mean = 112) and had between 14 and 38.9 % of all possible connections with host tree species. All observations included at least 9 vine species (9-21) and 10 host tree species (10-28). When we considered each forest separately, we found three forests that were significantly nested ($t = 14.512-46.246$, $p < 0.008$), eight forests that were significantly

specialised ($H2 = 0.260-0.407$, $p < 0.046$) and nine forests that maintained fewer interactions than expected by chance (observed = 0.14-0.389, $p < 0.02$).

Scaling species interactions with tree availability

Network analysis performed at the landscape scale and the patch scale showed similar results. Vines were more specialized, had a higher c-score and maintained fewer interactions than random expectations (Figure 2). However, the standardised Z scores from each set of analyses indicated that the inclusion of stand scale biological information (i.e., likelihood of interacting) reduced dissimilarity (from -370 to -3) between observed and randomly generated data. In contrast, nestedness and compartmentalisation was no different to random expectations (Table 1). Importantly, both null models predicted a higher number of species interactions than we observed (vines: $t = 11.628$, $df = 47$, $p < 0.001$, hosts: $t = 19.273$, $df = 78$, $p < 0.001$) suggesting that tree availability is overestimated when simulating network interactions.

Abundance, area and climbing strategy

Network connectivity was correlated with abundance (vines: $r^2 = 0.794$, $F_{1,46} = 177.100$, $p < 0.001$; hosts: $r^2 = 0.905$, $F_{1,77} = 735.700$, $p < 0.001$; Figure 3a, d), geographic area (vines: $r^2 = 0.919$, $F_{1,46} = 519.400$, $p < 0.001$, hosts: $r^2 = 0.857$, $F_{1,77} = 461.000$, $p < 0.001$; Figure 3b, e), and the number of interactions (vines: $r^2 = 0.957$, $F_{1,46} = 1013.000$, $p < 0.001$, hosts: $r^2 = 0.792$, $F_{1,77} = 293.300$, $p < 0.001$; Figure 3c, f). However, when we considered interactions within each stand we found no significant correlation between connectivity and tree diversity (vines: $r^2 = 0.005$, $F_{1,46} = 0.243$, $p = 0.624$; hosts: $r^2 = 0.000$, $F_{1,77} = 0.001$, $p = 0.982$; figure 4a, c). In contrast, we found a strong positive correlation between connectivity and the number of trees that were

used in each stand ($r^2 = 0.663$, $F_{1, 46} = 90.510$, $p < 0.001$; figure 4b; $r^2 = 0.473$, $F_{1, 77} = 69.190$, $p < 0.001$; figure 4d).

The 10 most connected species (> 21% connected) came from eight different families (Appendix 3) and included four different climbing strategies (Appendix 2). The most connected species (48 %), *Smilax australis* was found in fewer plots ($n = 25$) than *Geitonoplesium cymosum*, but was more abundant ($n = 159$) and attached to a greater number of trees ($n = 74$). However, vine abundance does not always permit higher connectivity in network interactions. *Smilax glyciophylla* was abundant ($n=109$) and only reached 22% connectivity; which is less than half of all interactions found for *S. australis*. The reason for limited connectivity in *S. glyciophylla* was due to geographic distribution, which was found in only three forests, by comparison to seven forests of *S. australis*. These results support the hypothesis that connectivity between vines and host trees are determined by the distribution and access to different tree species.

These results do not omit the possibility that functional traits limit species interactions. Although, climbing strategy may be less obvious than abundance and overloading of tree species. For example, scrambling plants were more likely to interact with a greater number of host species when they were less abundant (Appendix 2). In addition, singletons (i.e., having just one network interaction) were typically species from Apocynaceae and had (i) a twinning growth strategy, (ii) reached lower parts of the canopy, and (iii) were less abundant. Only one singleton (*Asparagus aethiopicus*) was abundant ($n=18$); however *Asparagus* was found in only one plot and generally forms dense thickets less dependent on host availability.

The most connected trees included Myrtle Ebony (*Diospyros pentamera*), Yellow Tulip Wood (*Drypetes deplanchei*), Guioa (*Guioa semiglauca*), Yellow Pear-fruit (*Mischocarpus pyriformis*), and Rose Tamarind (*Arytera divaricata*) which ranged from 29% to 40% connectivity. Not surprisingly, the most abundant and wide spread trees carried the most vine species. Some trees, such as Red Ash (*Alphitona excelsa*) carried more interactions when less abundant (e.g. four trees carried 23 interactions; Appendix 3). However, this occurred due to a strong interaction with Asparagus (18) in one plot. All singletons were species that occurred only once and included shrubs growing to a low maximum height of 1.5m (e.g. Bitou Bush).

Sampling effort and species accumulation

The accumulation of both vine and tree species were not exhausted in this study. We predicted that 13 plots are required to find one additional vine species and only nine plots are required to find an additional tree species. More importantly for network connectivity, we predicted that an additional species interaction would be found in the next plot (est. plots = 0.34).

Table 1 Five network indices were evaluated across the landscape and within patches. Results support deterministic patterns in both levels of investigation. Note the difference in z scores for each index between levels.

Index	Observed	Patch		Landscape	
		Z score	P value	Z score	P value
Web specialisation	0.351	8.3	< 0.001	33.7	< 0.001
C-score	0.606	2.8	0.002	27.5	< 0.001
Connectivity	0.113	-3.0	0.001	-370.0	< 0.001
Nestedness	36.757	0.2	0.439	-1.2	0.119
Compartments	1.000	-0.3	0.393	N/A	N/A

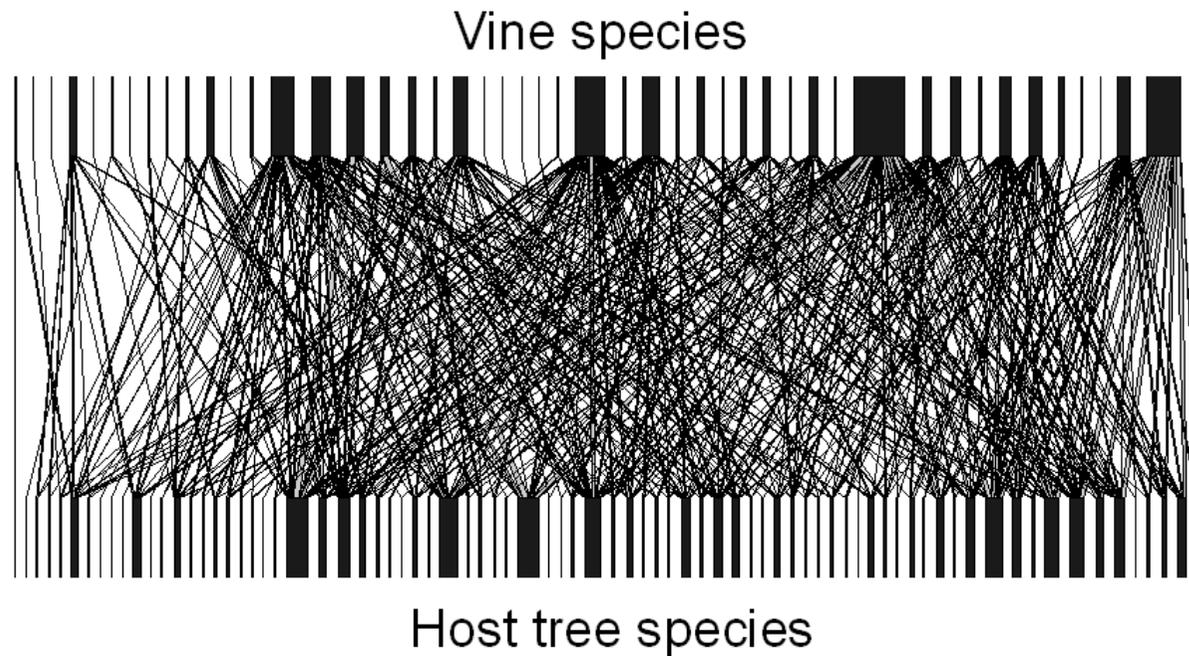


Figure 1 Forty eight vine species were observed interacting with 79 host tree species in nine forests along the east coast of New South Wales, Australia. The size of each node (filled rectangles) represents species abundance, and lines represent frequency of observed interactions. Many interactions were infrequent. The position of each species in the network was determined by minimising the number of overlapping interactions. Species names were removed for clarity.

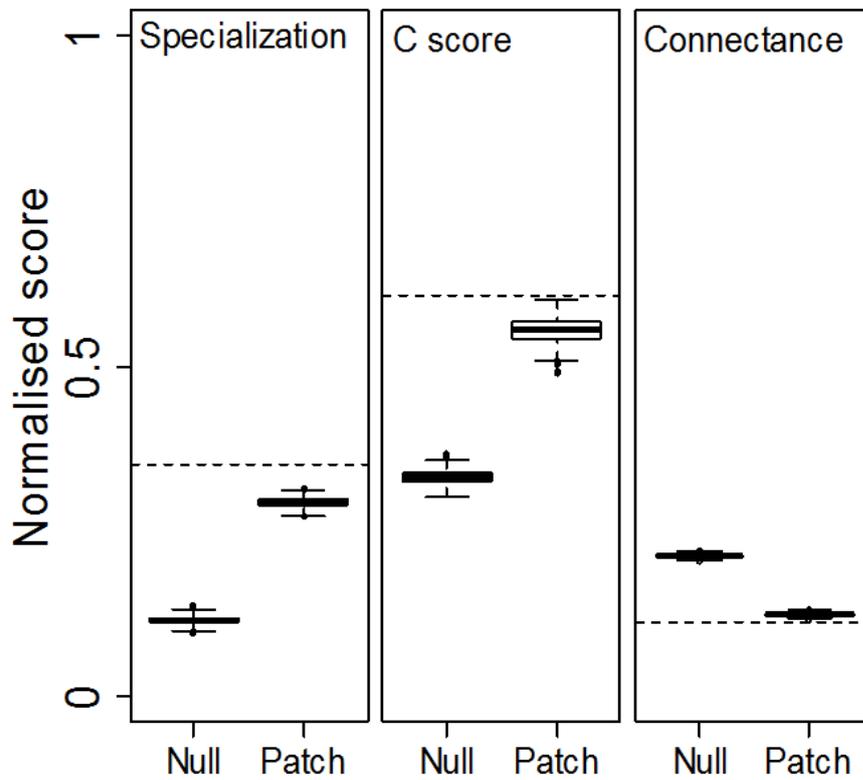


Figure 2 Null model outcomes for negative co-occurrence patterns. Each index described in each panel has been normalised to a score between 0 and 1. The landscape scale is referred to here as ‘Null’ and is a model without constraints, while patch refers to a model that is constrained by overlap in species distributions. Expected values were significantly different to observed indices of specialisation, c-score and connectivity (dashed line).

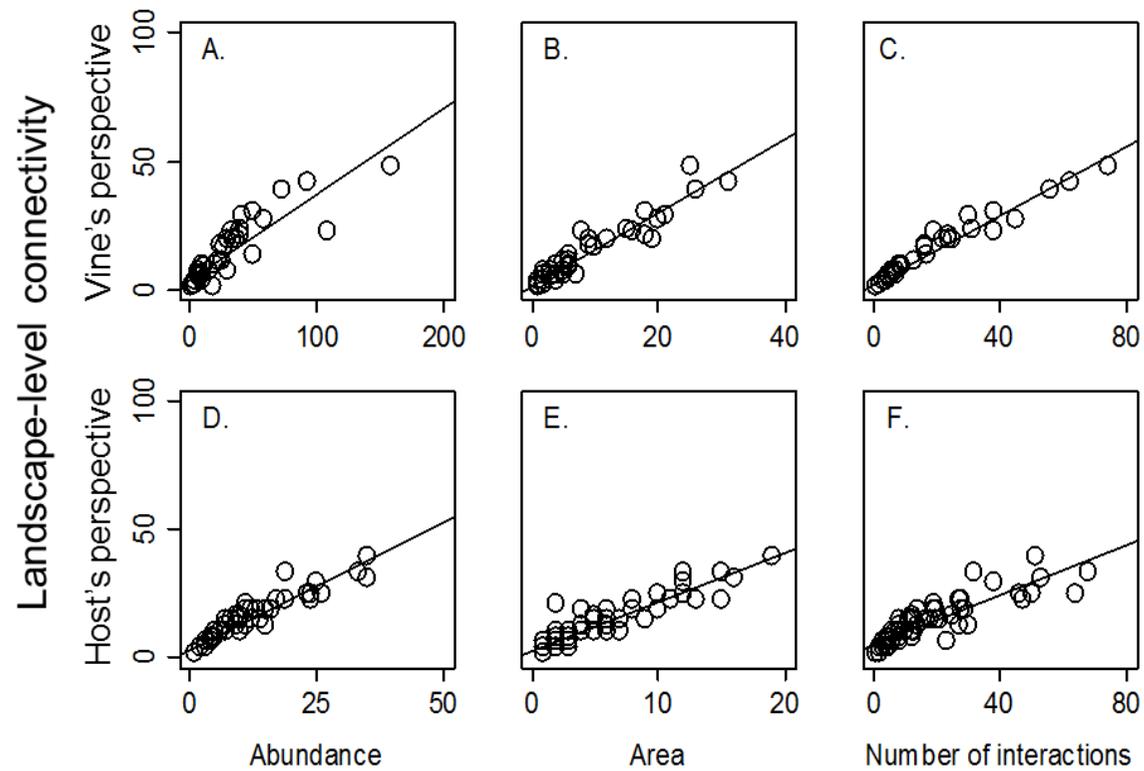


Figure 3 Network-level connectivity increased with area (A, D), abundance (B, E) and the number of interactions (C, F) for vines and host trees.

Note that figures including abundance and area have different values on the x-axis.

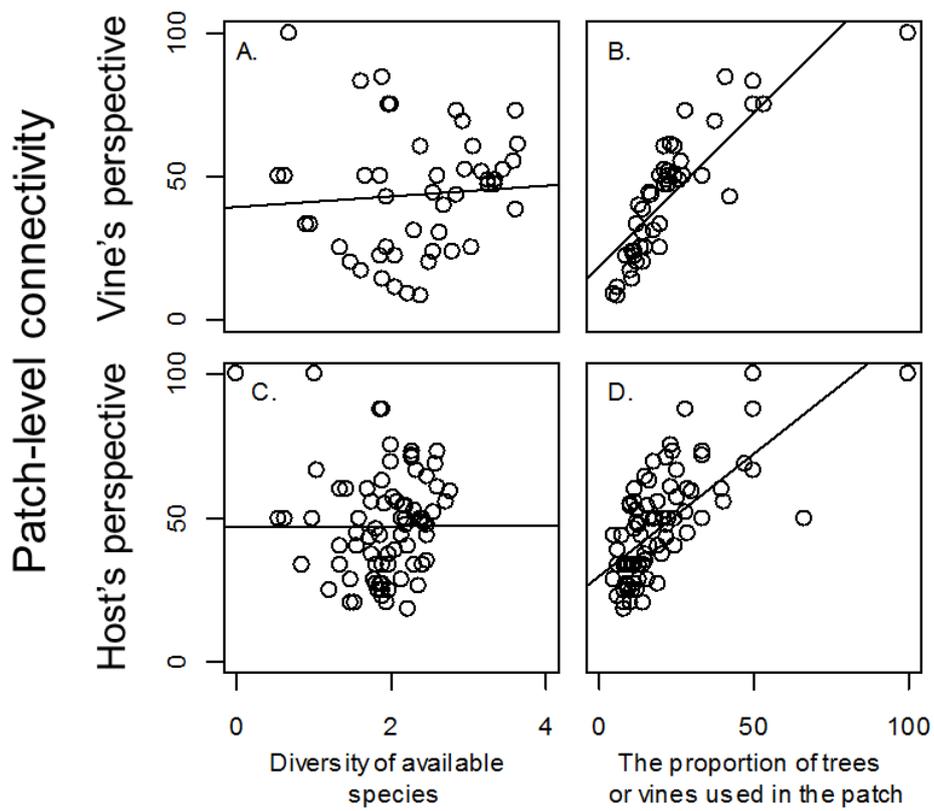


Figure 4 Patch-level connectivity did not increase with diversity (Shannon-Weiner diversity index), but did increase with the proportion of individuals that were ‘used’ in the patch. The most connected species are those interacting with all available individuals.

Discussion

Network interactions were more specialised than predicted by chance, supporting the results found in a New Zealand temperate forest (Blick and Burns 2009). Though, when we considered each forest individually, we found both positive (Sfair et al. 2011) and negative co-occurrence patterns (Blick and Burns 2009). These results indicate that specialisation in a vine-host network is determined by combining data from spatially separated forests. However, after considering tree availability and the likelihood of interacting only with neighbouring trees (adjusting null model evaluations) we still found more specialised interactions than predicted by chance. Further analyses revealed that connectivity was correlated with abundance, geographic distribution and the number of trees that were used as structural support. Surprisingly, stand diversity did not increase connectivity and novel interactions were not exhausted in this study. Together these results suggest that abundance and geographic distribution can generate coincidental associations and network properties are largely dependent on geographic extent and the study site selected for investigation.

We did not expect to find both patterns of connectivity (i.e., nestedness and negative co-occurrence patterns) in neighbouring forests. For a network to be considered nested, a core set of generalist species from both guilds need to interact, while maintaining many rare species (e.g. singletons). This was not the case in our vine-host network. From the four most connected species from each guild in the pooled network, two interactions were never made (*Geitonoplesium cymosum* on *Drypetes deplanchei* and *Trophis scandens* on *Guioa semiglauca*) and all interactions remained weak (e.g. *Morinda jasmoniodes* was considered common and only interacted with one of 38 individuals of *Drypetes deplanchei*). In addition, species that were rarely observed could form strong

interactions if they were abundant or dominant in a single plot. For example, Red Ash (*Alphitona excelsa*) while occurring only four times in three plots, maintained a high interaction frequency with Asparagus (*Asparagus aethiopicus*) even though interactions were possible with vine species (*Pandorea pandorana* and *Smilax glyciophylla*) that were considered generalists as they were connected with a greater number of tree species. We suggest that highly connected vine species interact with available trees regardless of host identity. Therefore, our observations of vines interacting with host trees could deviate from the expected association between a ‘core set of generalists’, required from a nested pattern.

Even though we found specialised interactions in the pooled dataset, we did find nested interactions in three from nine forests. Nested organisation of vines growing on host tree species occurred because there were a greater number of interactions between abundant species from each guild. For example, *Smilax glyciophylla*, *Smilax australis*, *Pandorea pandorana* and *Cissus hypoglauca* were abundant and interacted frequently with *Notolea longifolia*, *Elaeocarpus reticularis*, and *Syzygium smithii*. Each of these species in turn carried or climbed less abundant species. However, these species are not restricted to individual forests and when combined with additional data from surrounding regions the nested pattern is no longer apparent.

The structural requirements of vines and host characteristics have been hypothesised to generate patterns of connectivity. One explanation follows that the habit of different species can facilitate interactions between canopy tree species. In this study we found one tree species, Native Pomegranate (*Capparis arborea*), that has a scandant shrub habit. Interestingly, Native Pomegranate occurred seven times across five plots and

interacted with seven different species of vine. Despite finding 15% connectivity for the Native Pomegranate, the single forest where it was found was considered more specialised than predicted by chance, not nested. From the vines observed in this study, two highly connected species, including *Trophis scandens* and *Morinda jasmonoides*, are considered to have scrambling, scandent shrub forming habit. These species, included with *Geitonoplesium cymosum*, were among the most highly connected ‘scrambling’ species that interacted with less abundant tree species. It is therefore more appropriate to consider that vine-host interactions are simply more abundant in specific locations than predicted by chance, rather than adopting the notion that connectivity is more specialised than predicted by chance. Establishing the role of beta-diversity (changing host trees more rapidly than vines) on network structure is an important step worth much further consideration (Dalling et al. in press).

Disentangling spatial patterning from growth habit is difficult. Previous work indicates that the height to the first branch and bark characteristics could confer resistance to the colonisation of vine species (Balfour and Bond 1993). Results from this study are mixed. For example, *Asparagus aethiopicus* (scandent ascending shrub) interacted with *Alphitona excelsa* (small to medium sized tree) while twinning species did not (*Pandorana pandorana* and *Smilax glyciophylla*). Rather, *Pandorana pandorana* and *Smilax glyciophylla* interacted frequently with small to large trees (*Notolea*, *Elaeocarpus* and *Syzygium*). It is important to note that connectivity of vines with host trees is variable within our dataset (see Appendix 2) and the most common species included shrubs, small trees and large trees which have variable bark types (e.g. flakey or smooth). However, our data regarding plant morphology is compiled from reference

material. Disentangling the role of growth form would be an interesting direction for future research.

An alternative perspective asserts that plant-plant signalling could also generate co-occurrence patterns among different functional groups. A growing body of literature indicates that plants perceive neighbouring plants through shade-avoidance (Aphalo et al. 1999), recognising volatile compounds (Baldwin et al. 2002, Baldwin et al. 2006) and rhizosphere interactions not visible above ground (Estabrook and Yoder 1998). Under this hypothesis a vine may confer selective advantage by growing on specific trees that release volatile chemicals by avoiding herbivory. In support of this hypothesis, host volatile chemicals have been found in mistletoe-host interactions that mediate pollination events (Troncoso et al. 2010). In addition, several vine species may confer an advantage by facilitating growth towards the canopy. In order to test these hypotheses an alternative framework that looks beyond bipartite interactions is required. This would be a very interesting direction for future research. One approach would be to use a spatial association matrix implemented in the social network package, Pajek (Gomez et al. 2010), or to implement spatial covariance to evaluate organization of vine-host assemblages (Wagner 2003).

In summary, we show that nested and negative co-occurrence patterns are plausible properties of vine-host networks. We suggest that interactions are largely determined by the distribution and abundance of each species, rather than growth habit or preferential selection. We hypothesise that negative co-occurrence patterns of Blick and Burns (2009, 2011) may tend towards nested patterns of Sfair et al. (2011) given different sampling regimes and sampling effort.

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References

- Atmar, W. And Patterson, B.D. 1995. The nestedness temperature calculator. AICS Inc., University Park, USA.
- Aphalo, P.J., Ballare, C.L. and Scopel, A.L. 1999. Plant-plant signalling, the shade-avoidance response and competition. *Journal of Experimental Botany* 50: 1629-1634.
- Baldwin, I.T., Halitschke, R., Paschold, A., von Dahl, C.C. and Preston, C.A. 2006. *Science* 311: 812-815.
- Baldwin, I.T., Kessler, A. and Halitschke, R. 2002. Volatile signaling in plant-plant-herbivore interactions: what is real? *Current Opinion in Plant Biology* 5: 351-354.
- Balfour, D.A. & Bond, W.J. 1993. Factors limiting climber distribution and abundance in a southern African forest. - *Journal of Ecology* 81: 93-99.
- Bascompte, J. et al. 2003. The nested assembly of plant-animal mutualistic networks. *Proceedings of the National Academy of Sciences USA* 312: 431-433.
- Blüthgen, N. 2010. Why network analysis is often disconnected from community ecology: A critique and an ecologist's guide. - *Basic and Applied Ecology* 11: 185-195.
- Biswas, S.R. & Wagner, H.H. 2012. Landscape contrast: a solution to hidden assumptions in the metacommunity concept? - *Landscape Ecology* 27: 612-631.
- Blick, R. & Burns, K.C. 2009. Network properties of arboreal plants: Are epiphytes, mistletoes and lianas structured similarly? *Perspectives in Plant Ecology, Evolution*

- and Systematics 11: 41-52.
- Blick, R.A.J. & Burns, K.C. 2011. Liana co-occurrence patterns in a temperate rainforest. - *Journal of Vegetation Science* 22: 868-877.
- Blick, R.A.J. et al. 2012. Predicting network topology of mistletoe-host interactions: do mistletoes really mimic their hosts? - *Oikos* 121: 761-771.
- Burnham, R.J. 2002. Dominance, diversity and distribution of lianas in Yasuní, Ecuador: who is on top? - *Journal of Tropical Ecology* 18: 845-864.
- Burns, K.C. 2007. Network properties of an epiphyte meta-community. *Journal of Ecology* 95: 1142-1151.
- Burns, K.C. & Zotz, G. 2010. A hierarchical framework for investigating epiphyte assemblages: networks, metacommunities and scale. - *Ecology* 91: 377-385.
- Campbell, E.J.F. & Newbery, D.M.G. 1993. Ecological relationships between lianas and trees in lowland rain forest in Sabah, East Malaysia. - *Journal of Tropical Ecology* 9: 469-490.
- Clements, F.E. 1916. Plant succession. Carnegie Institution, Washington, DC, USA (Publication number 242).
- Connor, E.F. & Simberloff, D.S. 1979. The assembly of species communities: chance or competition? - *Ecology* 60, 1132-1140.
- Dalling, J.W. et al. In press. Resource-based habitat associations in a neotropical liana community. - *Journal of Ecology* [doi: 10.1111/j.1365-2745.2012.01989.x]
- DeWalt, S.J. et al. 2006. Liana habitat associations and community structure in a Bornean lowland tropical forest. - *Plant Ecology* 186: 203-216.
- Diamond, J.M. 1975. Assembly of species communities. In: Cody, M.L. & Diamond, J.M. (ed) *Ecology and evolution of communities*, Harvard University Press. Cambridge, MA, US, pp 342-444.

- Diamond, J.M. & Gilpin, M.E. 1982. Examination of the “null” model of Connor and Simberloff for species co-occurrence on islands. - *Oecologia* 52: 64-74.
- Dormann, C.F. et al. 2009. Indices, Graphs and Null models: Analyzing Bipartite Ecological Networks. - *The Open Ecology Journal* 2: 7-24.
- Estabrook, E.M. and Yoder, J.I. 1998. Plant-plant communications: Rhizosphere signalling between parasitic angiosperms and their hosts. *Plant Physiology* 116: 1-7.
- Gallagher, R.V. et al. 2011. Traits and ecological strategies of Australian tropical and temperate climbing plants. - *Journal of Biogeography* 38: 828-839.
- Gallagher, R.V. & Leishman, M.R. 2012. Contrasting patterns of community assembly in lianas and trees from temperate Australia - *Oikos* [doi: 10.1111/j.1600-0706.2012.20025.x]
- García, D. et al. 2011. The spatial scale of plant-animal interactions: effects of resource availability and habitat structure. - *Ecological Monographs* 81: 103-121.
- Genini, J. et al. 2012. Mistletoes Play Different Roles in a Modular Host–Parasite Network. - *Biotropica* 44: 171-178.
- Gilarranz, L.J. et al. 2012. The architecture of weighted mutualistic networks. - *Oikos* 121: 1154-1162.
- Gleason, H.A. 1917. The structure and development of the plant association. - *Bull. Torrey Bot. Soc.* 44, 463–481.
- Gomez, J.M., Verdu, M. and Perfectti, F. 2010. Ecological interactions are evolutionarily conserved across the entire tree of life. *Nature* 465: 918-921.
- Gotelli, N.J. 2000. Null model analysis of species co-occurrence patterns. - *Ecology* 81: 2606-2621.
- Gotelli, N.J. & Graves, G.R. 1995. Null models in ecology. Smithsonian Institution Press, Washington, D.C.

- Hausdorf, B. & Hennig, C. 2007. Null model tests of clustering of species, negative co-occurrence patterns and nestedness in metacommunities. - *Oikos* 116: 818-828.
- Holt, R.D. 2002. Food webs in space: on the interplay of dynamic instability and spatial processes. – *Ecological Research* 17: 261-273.
- Hubbell, S.P. 2001. The Unified Neutral Theory of Biodiversity and Biogeography. Princeton University Press, Princeton, USA.
- Ings, T.C. et al. 2009. Ecological networks – beyond food webs. *Journal of Animal Ecology* 78: 253-269.
- Jordano, P. 1987. Patterns of mutualistic interactions in pollination and seed dispersal: connectance, dependence asymmetries, and coevolution. - *American Naturalist* 129: 657-677.
- Keddy, P., Weiher, E., 1999. Assembly rules as general constraints on community composition. In: Weiher, E., Keddy, P. (Eds.), *Ecological Assembly Rules*. Cambridge University Press, Cambridge, UK.
- Kotliar, N.B. & Wiens, J.A. 1990. Multiple scales of patchiness and patch structure: a hierarchical framework for the study of heterogeneity. - *Oikos* 59: 253-260.
- Lafferty, K.D. & Dunne, J.A. 2010. Stochastic ecological network occupancy (SENO) models: a new tool for modeling ecological networks across spatial scales. - *Theoretical Ecology* 3: 123-135.
- Leibold, M.A. et al. 2004. The metacommunity concept: a framework for multi-scale community ecology. - *Ecology Letters* 7: 601–613.
- Malizia, A. et al. 2010. Soil phosphorus and disturbance influence liana communities in a subtropical montane forest. - *Journal of Vegetation Science* 21: 551-560.
- Morales J.M. and Vázquez D.P. 2008. The effect of space in plant-animal mutualistic networks: insights from a simulation study. - *Oikos* 117: 1362-1370.

- Pandit, S.N. et al. 2009. Contrasts between habitat generalists and specialists: an empirical extension to the basic metacommunity framework. - *Ecology* 90: 2253-2262.
- Peres-Neto, P.R. et al. 2001. Environmentally constrained null models: site suitability as occupancy criterion. - *Oikos* 93: 110-120.
- Pillai, P., Lorwau, M. and Gonzalez, A. 2010. A patch-dynamic framework for food web metacommunities. - *Theoretical Ecology* 3: 223-237.
- Proulx, S.R. et al. 2005. Network thinking in ecology and evolution. - *Trends in Ecology and Evolution* 20: 345–353.
- Putz, F.E. 1984. The natural history of lianas on Barro Colorado Island, Panama. - *Ecology* 65: 1713-1724.
- R Development Core Team 2006 R: A language and environment for statistical computing R Foundation for Statistical Computing, Vienna, Austria. Available at: <http://www.r-project.org>.
- Roxburgh, S.H. & Chesson, P. 1998. A new method for detecting species associations with spatially autocorrelated data. - *Ecology* 79: 2180-2192.
- Roxburgh, S.H. & Matsuki, M. 1999. The statistical validation of null models used in spatial association analyses. - *Oikos* 85: 68-78.
- Schnitzer, S.A. 2005. A mechanistic explanation for the global patterns of liana abundance and distribution. - *American Naturalist* 166: 262-276.
- Schnitzer, S.A. & Bongers, F. 2002. The ecology of lianas and their role in forests. - *Trends in Ecology and Evolution* 17: 223-230.
- Sfair, J.C. et al. 2010. Nested liana-tree network in three distinct neotropical vegetation formations. - *Perspectives in Plant Ecology, Evolution and Systematics* 12: 277-281.
- Stang, M. et al. 2006. Size constraints and flower abundance determine the number of

- interactions in a plant-flower-visitor web. - *Oikos* 112: 111-121.
- Stang, M. et al. 2009. Size specific interaction patterns and size matching in a plant-pollinator interaction web. - *Annals of Botany* 103: 1459-1469.
- Stone, L. & Roberts, A. 1990. The checkerboard score and species distributions. - *Oecologia* 85: 74-79.
- Troncoso, A.J., Cabezas, N.J., Faúndez, E.H. and Urzúa, A. 2010. Host-mediated volatile polymorphism in a parasitic plant influences its attractiveness to pollinators. *Oecologia* 162: 413-425.
- Gotelli N. J. & Ulrich W. 2012. Statistical challenges in null model analysis. - *Oikos* 121: 171-180.
- Ulrich, W., 2006. Nestedness-A FORTRAN program for calculating ecological matrix temperatures. www.uni.torun.pl/~ulrichw.
- Ulrich, W. & Gotelli, N. 2010. Null model analysis of species associations using abundance data. - *Ecology* 9: 3384-3397.
- Vázquez, D.P. et al. 2009a. Evaluating multiple determinants of the structure of plant-animal mutualistic networks. - *Ecology* 90: 2039-2046.
- Vázquez D.P. et al. 2009b. Uniting pattern and process in plant-animal mutualistic networks: a review. - *Annals of Botany* 103: 1445-1457.
- Wagner, H.H. 2003. Spatial covariance in plant communities: integrating ordination, geostatistics, and variance testing. *Ecology* 84: 1045-1057.

Appendix 1

Used to constrain interactions between vines and trees down to plot level

```
for (i in 1:1000) {  
  zero.matrix<-ifelse(vine.neutral.matrix>=1,0,0) #make a zero matrix to add later  
  zeros<-as.matrix(zero.matrix[1,1:79])  
  mat<-ifelse(vine.neutral.matrix>=1,1,0) # change matrix to 1s and 0's
```

1. Carry out this procedure for all 48 species (e.g. Aph.res is the species name of the first vine in the matrix)

```
Aph.res.prob<-as.vector(vine.neutral.matrix[1,1:79])  
# assigns probabilities based on abundance  
Aph.res1<-sample(mat[1,],15,prob= Aph.res.prob, replace=T)  
# weighted sampling using probability  
Aph.res2<-t(sapply(by(as.matrix(Aph.res1), rownames(as.matrix(Aph.res1)), colSums),  
identity))  
Aph.res3<-cbind(as.matrix(Aph.res2),t(zeros))  
sp1<-aggregate (t(Aph.res3), by= list(row.names(as.data.frame (t(Aph.res3))))), FUN =  
"sum")  
Aph.res<-sp1$V1
```

...n

bind all species together to get a single interaction matrix

```
patch.matrix<-t(rbind(Aph.res, Asp.aet, Bill.sca, Cal.mue, Cal.aus, Cay.cle, Cel.sub,  
Cis.ant, Cis.hyp, Cis.ste, Cle.ari, Cle.gly, Coe.pan, Del.odo, Dio.tra, Emb.aus, Eus.lat,  
Fla.ind, Gei.cym, Hib.sca, Hip.bar, Hoy.aus, Ipo.cai, Jas.vol, Lan.cam, Leg.moo,  
Mac.coc, Mar.fla, Mar.hem, Mar.llo, Mar.ros, Mor.jas, Muc.gig, Pan.jas, Pan.pan,  
Par.rot, Par.str, Pip.nov, Rip.alb, Sar.har, Sec.ell, Smi.aus, Smi.gly, Ste.jap, Tet.nit,  
Tro.sca, Tyl.bar, Tyl.ben))
```

calculate network statistics using indices from the *bipartite* package

```
nest<-nestedness(patch.matrix,null.models=FALSE,n.nulls=0)  
cs<-C.score(t(patch.matrix),normalise=TRUE)  
Web.spec<-as.vector(H2fun(patch.matrix))  
comp<-compart(patch.matrix)  
connectance<-(sum(ifelse(patch.matrix>=1,1,0))/(79*48))*100
```

```
indices<-c(nest$temperature,cs,Web.spec[1],comp$n.compart,connectance)  
write(indices, file="C:\\ Desktop\\results.txt",append=TRUE)  
}
```

interpret the expected distribution using *stats* package

Appendix 2 Summary of all vine data. Five climbing strategies were observed across 48 vine species. Numbers in bold represent the five highest values in each column. Data is arranged in descending order of connectivity.

Vine species	Abundance	<i>N</i> forests	<i>N</i> plots	<i>N</i> Host species	<i>N</i> trees used	Connectivity	Climbing strategy	Other strategies
<i>Smilax australis</i>	159	7	25	38	74	48.10	Hooks	Tendrils
<i>Geitonoplesium cymosum</i>	92	8	31	33	62	41.77	Scrambling	
<i>Trophis scandens</i>	73	6	26	31	56	39.24	Scrambling	Scandent shrub
<i>Morinda jasminoides</i>	50	5	18	24	38	30.38	Scrambling	Scandent shrub
<i>Cissus antarctica</i>	41	5	21	23	30	29.11	Tendrils	
<i>Dioscorea transversa</i>	58	5	20	22	45	27.85	Twinning	
<i>Cissus hypoglauca</i>	40	6	15	19	31	24.05	Tendrils	
<i>Eustrephus latifolius</i>	32	5	16	18	19	22.78	Scrambling	
<i>Smilax glycyphylla</i>	109	3	8	18	38	22.78	Tendrils	
<i>Marsdenia rostrata</i>	39	6	18	17	24	21.52	Twinning	
<i>Cissus sterculiifolia</i>	34	3	12	16	24	20.25	Tendrils	
<i>Pandorea pandorana</i>	36	3	9	16	25	20.25	Scrambling	Twinning
<i>Parsonsia straminea</i>	29	5	19	16	22	20.25	Adventitious roots	Twinning
<i>Flagellaria indica</i>	24	3	9	14	16	17.72	Tendrils	
<i>Tetrastigma nitens</i>	26	3	10	13	16	16.46	Tendrils	
<i>Ripogonum album</i>	49	3	6	11	17	13.92	Hooks	Scrambling
<i>Hippocratea barbata</i>	25	2	5	9	13	11.39	Twinning	
<i>Jasminum volubile</i>	23	1	6	9	13	11.39	Scrambling	
<i>Embelia australiana</i>	9	2	6	8	8	10.13	Scandent shrub	
<i>Hoya australis</i>	19	2	4	8	9	10.13	Twinning	Scrambling
<i>Stephania japonica</i>	11	4	6	7	9	8.86	Twinning	Hooks
<i>Aphanopetalum resinsum</i>	15	1	3	6	7	7.59	Scandent shrub	

<i>Maclura cochinchinensis</i>	6	5	5	6	6	7.59	Scandent shrub	Spines
<i>Secamone elliptica</i>	30	1	2	6	6	7.59	Scrambling	
<i>Callerya australis</i>	9	3	4	5	6	6.33	Scrambling	
<i>Celastrus subspicata</i>	7	2	2	5	5	6.33	Scrambling	
<i>Lantana camara</i>	8	2	5	5	6	6.33	Scandent shrub	hooks
<i>Sarcopetalum harveyanum</i>	9	5	7	5	7	6.33	Twinning	
<i>Clematis glycinoides</i>	7	2	3	4	5	5.06	Twinning	
<i>Cayratia clematidea</i>	3	1	1	3	3	3.80	Tendrils	
<i>Coelospermum paniculatum</i>	4	1	4	3	4	3.80	Scrambling	
<i>Delairea odorata</i>	3	1	1	3	3	3.80	Twinning	
<i>Tylophora benthamii</i>	10	2	2	3	3	3.80	Scrambling	
<i>Hibbertia scandens</i>	2	2	2	2	2	2.53	Scrambling	
<i>Legnephora moorei</i>	3	1	1	2	2	2.53	Twinning	
<i>Pandorea jasminoides</i>	2	1	1	2	2	2.53	Scrambling	
<i>Tylophora barbata</i>	2	2	2	2	2	2.53	Twinning	
<i>Asparagus aethiopicus</i>	18	1	1	1	1	1.27	Scandent shrub	Spines
<i>Billardiera scandens</i>	1	1	1	1	1	1.27	Scandent shrub	
<i>Calamus muelleri</i>	1	1	1	1	1	1.27	Spines	
<i>Clematis aristata</i>	1	1	1	1	1	1.27	Twinning	
<i>Ipomoea cairica</i>	1	1	1	1	1	1.27	Twinning	
<i>Marsdenia flavescens</i>	1	1	1	1	1	1.27	Twinning	
<i>Marsdenia hemiptera</i>	1	1	1	1	1	1.27	Twinning	
<i>Marsdenia lloydii</i>	1	1	1	1	1	1.27	Twinning	
<i>Mucuna gigantea</i>	1	1	1	1	1	1.27	Scrambling	
<i>Parsonsia rotata</i>	1	1	1	1	1	1.27	Twinning	
<i>Piper novae-hollandiae</i>	1	1	1	1	1	1.27	Adventitious roots	

Appendix 3 Summary of all host tree data. Trees varied in size across 78 species. Numbers in bold represent the five highest values in each column. Data is arranged in descending order of connectivity. Size categories and bark type according to Plantnet.com.

Host species	Abundance	<i>N</i> forests	<i>N</i> plots	<i>N</i> vine species	<i>N</i> interactions	Connectivity	Size	Branch/Bark/leaf texture
<i>Diospyros pentamera</i>	35	4	19	19	51	39.6	Small-med	
<i>Drypetes deplanchei</i>	33	4	15	16	68	33.3	Small-med	
<i>Guioa semiglauca</i>	19	7	12	16	32	33.3	6m	Smooth bark
<i>Mischocarpus pyriformis</i>	35	4	16	15	53	31.3	18m	
<i>Arytera divaricata</i>	25	4	12	14	38	29.2	30m	
<i>Cupaniopsis anacardioides</i>	23	4	12	12	64	25.0	Small-med	
<i>Notelaea longifolia</i>	26	5	12	12	46	25.0	9m	
<i>Syzygium smithii</i>	24	4	10	12	50	25.0	Shrub	Smooth to slightly flaky bark
<i>Elaeodendron australe</i>	17	5	11	11	28	22.9	8m	
<i>Euroschinus falcatus</i>	24	5	15	11	47	22.9	Small-med	Glabrous/pubescent Buttressed and slightly flaky bark
<i>Syzygium luehmannii</i>	19	2	8	11	27	22.9	Med-large	
<i>Syzygium oleosum</i>	17	4	13	11	27	22.9	Small	Flaky
<i>Diospyros fasciculosa</i>	11	1	2	10	19	20.8	Small-med	
<i>Acronychia imperforata</i>	16	4	8	9	28	18.8	9m	
<i>Celtis paniculata</i>	11	3	6	9	14	18.8	Small-med	
<i>Pittosporum undulatum</i>	13	3	8	9	29	18.8	15m	
<i>Pouteria australis</i>	15	2	10	9	19	18.8	Med-tall	
<i>Pouteria chartacea</i>	12	1	4	9	20	18.8	Small	
<i>Endiandra sieberi</i>	9	3	5	8	25	16.7	Small-med	Hard corky bark
<i>Eupomatia laurina</i>	10	2	5	8	12	16.7	Shrub-small tree	
<i>Capparis arborea</i>	7	1	5	7	8	14.6	Scandent shrub	

<i>Clerodendrum tomentosum</i>	8	4	5	7	11	14.6	10m	Velvety leaf surface
<i>Glochidion ferdinandi</i>	8	4	5	7	17	14.6	Small-med	
<i>Myrsine howittiana</i>	10	2	5	7	18	14.6	Shrub-small tree	
<i>Podocarpus elatus</i>	8	2	6	7	13	14.6	Med-large	Fissured and scaly
<i>Polyalthia nitidissima</i>	14	1	6	7	21	14.6	Shrub-small tree	
<i>Polyscias elegans</i>	10	3	7	7	11	14.6	30m	Sparingly branched
<i>Scolopia braunii</i>	12	3	9	7	20	14.6	Med	Coppice shoots
<i>Syzygium australe</i>	8	4	6	7	12	14.6	Shrub-small tree	Flaky
<i>Acronychia wilcoxiana</i>	9	2	6	6	14	12.5	9m	
<i>Elaeocarpus reticulatus</i>	15	1	6	6	30	12.5	Shrub-small tree	
<i>Livistona australis</i>	11	2	6	6	27	12.5	30m	Solitary stem
<i>Trochocarpa laurina</i>	7	1	4	6	8	12.5		
<i>Acronychia littoralis</i>	6	1	3	5	7	10.4	6m	
<i>Acronychia oblongifolia</i>	6	2	6	5	6	10.4	27m	
<i>Cyclophyllum longipetalum</i>	6	3	4	5	8	10.4	10m	
<i>Diospyros australis</i>	5	3	5	5	6	10.4	Shrub-small tree	
<i>Endiandra discolor</i>	5	2	2	5	10	10.4	Med	Buttressed, becoming rough bark
<i>Myrsine variabilis</i>	6	4	6	5	6	10.4	Shrub-small tree	
<i>Psychotria loniceroides</i>	10	2	7	5	12	10.4	5m	
<i>Synoum glandulosum</i>	7	1	4	5	13	10.4	7m	Brown scaly bark Spreading, deeply fissured
<i>Acacia maidenii</i>	5	3	3	4	12	8.3	20m	Buttressed
<i>Elaeocarpus obovatus</i>	4	3	2	4	5	8.3	Small	Buttressed
<i>Ficus fraseri</i>	4	1	3	4	8	8.3	Med	Scabrous
<i>Acacia disparrima</i>	3	1	2	3	4	6.3	12m	Bark fissured
<i>Alphitonia excelsa</i>	4	1	3	3	23	6.3	Small-med	

<i>Baloghia inophylla</i>	3	1	3	3	5	6.3	Med	
<i>Breynia oblongifolia</i>	3	2	2	3	6	6.3	3m	
<i>Dysoxylum rufum</i>	4	1	1	3	3	6.3	Small-med	wrinkled bark
<i>Eucalyptus botryoides</i>	3	1	1	3	3	6.3	40m	Bark persistent, platy and fissured
<i>Flindersia australis</i>	3	1	1	3	3	6.3	40m	Moderately buttressed
<i>Litsea australis</i>	4	1	3	3	8	6.3	20m	Scaly bark
<i>Psydrax lamprophylla</i>	3	1	2	3	4	6.3	12m	
<i>Rhodomyrtus psidioides</i>	3	1	2	3	8	6.3	12m	Scaly bark
<i>Senna pendula</i>	4	2	3	3	3	6.3	3m	Noxious weed, spreading
<i>Syzygium hemilamprum</i>	3	1	1	3	4	6.3	med-tall	flaky, often fissured
<i>Acacia irrorata</i>	2	1	1	2	3	4.2	12m	Smooth
<i>Alectryon coriaceus</i>	2	2	2	2	3	4.2	Small	
<i>Allocasuarina littoralis</i>	2	1	2	2	2	4.2	15m	
<i>Aphananthe philippinensis</i>	2	1	1	2	4	4.2	20m	
<i>Dysoxylum fraserianum</i>	2	2	2	2	2	4.2	Med-large	Scaly bark
<i>Exocarpos latifolius</i>	3	1	3	2	5	4.2	10m	
<i>Ficus coronata</i>	2	1	1	2	2	4.2	Shrub-small tree	Scabrous
<i>Helicia glabriflora</i>	2	1	1	2	2	4.2	10m	
<i>Monotoca elliptica</i>	2	1	1	2	4	4.2	4m	
<i>Pararchidendron pruinosum</i>	2	1	2	2	2	4.2	15m	
<i>Pittosporum revolutum</i>	2	1	1	2	2	4.2	3m	
<i>Syzygium francisii</i>	2	1	2	2	3	4.2	Med	Large buttress and flaky bark
<i>Syzygium moorei</i>	2	1	1	2	2	4.2	Large	Bark flaky
<i>Wilkiea huegeliana</i>	2	2	2	2	3	4.2	Shrub-small tree	

<i>Chrysanthemoides monilifera</i>	1	1	1	1	1	2.1	1.5m	
<i>Claoxylon australe</i>	1	1	1	1	1	2.1	9m	
<i>Commersonia bartramia</i>	1	1	1	1	1	2.1	Shrub-small tree	
<i>Cryptocarya triplinervis</i>	1	1	1	1	2	2.1	Small-med	
<i>Ficus obliqua</i>	1	1	1	1	2	2.1	Med-large	
<i>Macrozamia communis</i>	1	1	1	1	1	2.1	2m	
<i>Notelaea venosa</i>	1	1	1	1	1	2.1	6m	
<i>Pittosporum multiflorum</i>	1	1	1	1	1	2.1	3m	Much branched
<i>Trema tomentosa</i>	1	1	1	1	1	2.1	Shrub-small tree	

Appendix4 Common names and taxonomic authority of all vine species recorded in this study.

Vine species	Family	Common name	Authority
<i>Smilax australis</i>	Smilacaceae	Lawyer Vine	R. Br.
<i>Geitonoplesium cymosum</i>	Luzuriagaceae	Scrambling Lilly	(R.Br.) A.Cunn. ex Hook.
<i>Trophis scandens</i>	Moraceae	Burny Vine	(Lour.) Hook. & Arn.
<i>Morinda jasminoides</i>	Rubiaceae	Sweet Morinda	A.Cunn.
<i>Cissus antarctica</i>	Vitaceae	Kangaroo Vine	Vent.
<i>Dioscorea transversa</i>	Dioscoreaceae	Native Yam	R. Br.
<i>Cissus hypoglauca</i>	Vitaceae	Water Vine	A. Gray
<i>Eustrephus latifolius</i>	Luzuriagaceae	Wombat Berry	R.Br. ex Ker Gawl.
<i>Smilax glyciophylla</i>	Smilacaceae	Sweet Sarsaparilla	Sm.
<i>Marsdenia rostrata</i>	Apocynaceae	Milk Vine	R. Br.
<i>Cissus sterculiifolia</i>	Vitaceae	Yaroong	(F.Muell. ex Benth.) Planch.
<i>Pandorea pandorana</i>	Bignoniaceae	Wonga Wonga	(Andrews) Steenis
<i>Parsonsia straminea</i>	Apocynaceae	Monkey Rope	(R.Br.) F.Muell.
<i>Flagellaria indica</i>	Flagellariaceae	Whip Vine	L.
<i>Tetrastigma nitens</i>	Vitaceae		(F.Muell.) Planch.
<i>Ripogonum album</i>	Ripogonaceae	White Supplejack	R. Br.
<i>Hippocratea barbata</i>	Celastraceae	Knot Vine	F.Muell.
<i>Jasminum volubile</i>	Oleaceae	Stiff Jasmine	Jacq.
<i>Embelia australiana</i>	Myrsinaceae		(F.Muell.) F.M.Bailey
<i>Hoya australis</i>	Apocynaceae	Native Hoya	
<i>Stephania japonica</i>	Menispermaceae	Snake Vine	(Thunb.) Miers
<i>Aphanopetalum resinosum</i>	Aphanopetalaceae	Gum Vine	Endl.

<i>Maclura cochinchinensis</i>	Moraceae	Cockspur Thorn	(Lour.) Corner
<i>Secamone elliptica</i>	Apocynaceae	Corky Milk Vine	R. Br.
<i>Callerya australis</i>	Fabaceae	Native Westeria	(Endl.) Schot
<i>Celastrus subspicata</i>	Celastraceae	Large Leaf Staff Vine	Hook.
<i>Lantana camara</i>	Verbenaceae	Lantana	L.
<i>Sarcopetalum harveyanum</i>	Menispermaceae	Pearl Vine	F.Muell.
<i>Clematis glycinoides</i>	Ranunculaceae	Headache Vine	DC.
<i>Cayratia clematidea</i>	Vitaceae	Native Grape	(F.Muell.) Domin
<i>Coelospermum paniculatum</i>	Rubiaceae		F.Muell.
<i>Delairea odorata</i>	Asteraceae	Cape Ivy	Lem.
<i>Tylophora benthamii</i>	Apocynaceae	Coast Tylophora	Tsiang
<i>Hibbertia scandens</i>	Dilleniaceae	Climbing Guinea Flower	(Willd.) Gilg
<i>Legnephora moorei</i>	Menispermaceae	Round-leaf Vine	(F.Muell.) Miers
<i>Pandorea jasminoides</i>	Bignoniaceae	Bower Vine	(Lindl.) Schum.
<i>Tylophora barbata</i>	Apocynaceae	Bearded Tylophora	R. Br.
<i>Asparagus aethiopicus</i>	Asparagaceae	Asparagus 'Fern'	L.
<i>Billardiera scandens</i>	Pittosporaceae	Hairy Apple Berry	Sm.
<i>Calamus muelleri</i>	Arecaceae	Wait-a-while	H.Wendl.
<i>Clematis aristata</i>	Ranunculaceae	Old Man's Beard	Ker Gawl.
<i>Ipomoea cairica</i>	Convolvulaceae	Coastal Morning Glory	(L.) Sweet
<i>Marsdenia flavescens</i>	Apocynaceae	Hairy Milk Vine	A.Cunn. ex Hook.
<i>Marsdenia hemiptera</i>	Apocynaceae		Rchb.
<i>Marsdenia lloydii</i>	Apocynaceae	Corky Marsdenia	P.I.Forst.
<i>Mucuna gigantea</i>	Fabaceae	Burny Bean	(Willd.) DC.
<i>Parsonsia rotata</i>	Apocynaceae	Veinless Silkpod	Maiden & Betche
<i>Piper novae-hollandiae</i>	Piperaceae	Giant Pepper Vine	(Miq.) C.DC. var. <i>hederaceum</i>

Appendix 5 Common names and taxonomic authority of all tree species recorded in this study.

Tree species	Family	Common name	Authority
<i>Diospyros pentamera</i>	Ebenaceae	Mrytle Ebony	(Woolls & F.Muell.) F.Muell.
<i>Drypetes deplanchei</i>	Putranjivaceae	Yellow Tulip Wood	(Brongn. & Gris) Merr.
<i>Guioa semiglauc</i>	Sapindaceae	Guioa	(F.Muell.) Radlk.
<i>Mischocarpus pyriformis</i>	Sapindaceae	Yellow Pear-fruit	F.Muell.) Radlk.
<i>Arytera divaricata</i>	Sapindaceae	Rose Tamarind	F.Muell.
<i>Cupaniopsis anacardioides</i>	Sapindaceae	Tuckeroo	(A.Rich.) Radlk.
<i>Notelaea longifolia</i>	Oleaceae	Large Mock-olive	Vent.
<i>Syzygium smithii</i>	Myrtaceae	Lilly Pilly	(Poir.) Nied.
<i>Elaeodendron australe</i>	Celastraceae		Vent.
<i>Euroschinus falcatus</i>	Anacardiaceae	Ribbonwood	Hook.f.
<i>Syzygium luehmannii</i>	Myrtaceae	Small-leaved Lilly Pilly	(F.Muell.) L.A.S.Johnson
<i>Syzygium oleosum</i>	Myrtaceae	Blue Lilly Pilly	(F.Muell.) B.Hyland
<i>Diospyros fasciculosa</i>	Ebenaceae	Grey Ebony	(F.Muell.) F.Muell.
<i>Acronychia imperforata</i>	Rutaceae	Logan Apple	F.Muell.
<i>Celtis paniculata</i>	Ulmaceae	Native Celtis	(Endl.) Planch.
<i>Pittosporum undulatum</i>	Pittosporaceae	Native Daphne	Vent.
<i>Pouteria australis</i>	Sapotaceae	Black Apple	(R.Br.) Pierre
<i>Pouteria chartacea</i>	Sapotaceae	Thin-leaved Coondo	(F.Muell. ex Benth.) Baehni
<i>Endiandra sieberi</i>	Lauraceae	Hard Corkwood	Nees
<i>Eupomatia laurina</i>	Eupomatiaceae	Bolwara	R. Br.

<i>Capparis arborea</i>	Capparaceae	Native Pomegranate	(F.Muell.) Maiden
<i>Clerodendrum tomentosum</i>	Lamiaceae	Downy Chance Tree	R. Br.
<i>Glochidion ferdinandi</i>	Phyllanthaceae	Cheese Tree	(Muell.Arg.) F.M.Bailey
<i>Myrsine howittiana</i>	Myrsinaceae	Brush Muttonwood	(F.Muell. ex Mez) Jackes
<i>Podocarpus elatus</i>	Podocarpaceae	Plum Pine	R.Br. ex Endl.
<i>Polyalthia nitidissima</i>	Annonaceae	Shiny-leaf Tree	(Dunal) Benth.
<i>Polyscias elegans</i>	Araliaceae	Celery Wood	(C.Moore & F.Muell.) Harms
<i>Scolopia braunii</i>	Flacourtiaceae	Flintwood	(Klotzsch) Sleumer
<i>Syzygium australe</i>	Mrytaceae	Brush Cherry	(J.C.Wendl. ex Link) B.Hyland
<i>Acronychia wilcoxiana</i>	Rutaceae	Silver Aspen	(F.Muell.) T.G.Hartley
<i>Elaeocarpus reticulatus</i>	Elaeocarpaceae	Blueberry Ash	Sm.
<i>Livistona australis</i>	Arecaceae	Cabbage Fan Palm	(R.Br.) Mart.
<i>Trochocarpa laurina</i>		Tree Heath	
<i>Acronychia littoralis</i>	Rutaceae	Scented Acronychia	T.G.Hartley & J.B.Williams
<i>Acronychia oblongifolia</i>	Rutaceae	White Aspen	(A.Cunn. ex Hook.) Endl. ex Heynh.
<i>Cyclophyllum longipetalum</i>	Rubiaceae	Coast Canthium	S.T.Reynolds & R.J.F.Hend.
<i>Diospyros australis</i>	Ebenaceae	Black Plum	(R.Br.) Hiern
<i>Endiandra discolor</i>	Lauraceae	Rose Walnut	Benth.
<i>Myrsine variabilis</i>	Myrsinaceae		R.Br.
<i>Psychotria loniceroides</i>	Rubiaceae	Hairy Psychotria	Sieber ex DC.
<i>Synoum glandulosum</i>	Meliaceae	Scentless Rosewood	(Sm.) Juss.
<i>Acacia maidenii</i>	Fabaceae	Maidens Wattle	F.Muell.
<i>Elaeocarpus obovatus</i>	Elaeocarpaceae	Hard Quangdong	G.Don
<i>Ficus fraseri</i>	Moraceae	Sandpaper Fig	Miq.
<i>Acacia disparrima</i>	Fabaceae		M.McDonald & Maslin
<i>Alphitonia excelsa</i>	Rhamnaceae	Red Ash	(A.Cunn. ex Fenzl) Benth.

<i>Baloghia inophylla</i>	Euphorbiaceae	Brush Bloodwood	(G.Forst.) P.S.Green
<i>Breynia oblongifolia</i>	Phyllanthaceae	Coffee Bush	Muell.Arg.
<i>Dysoxylum rufum</i>	Meliaceae	Hairy Rosewood	(A.Rich.) Benth.
<i>Eucalyptus botryoides</i>	Mrytaceae	Bangalay	Sm.
<i>Flindersia australis</i>	Rutaceae	Crows Ash	R.Br.
<i>Litsea australis</i>	Lauraceae	Brown Bolly Gum	B.Hyland
<i>Psyrax lamprophylla</i>	Rubiaceae	Large-leaved Canthium	(F.Muell.) Bridson
<i>Rhodomyrtus psidioides</i>	Mrytaceae	Native Guava	(G.Don) Benth.
<i>Senna pendula</i>	Fabaceae		
<i>Syzygium hemilamprum</i>	Myrtaceae	Broad-leaf Lilly Pilly	(F.Muell.) Merr. & L.M.Perry
<i>Acacia irrorata</i>	Fabaceae	Green Wattle	Sieber ex Spreng.
<i>Alectryon coriaceus</i>	Sapindaceae	Beach Alectryon	(Benth.) Radlk.
<i>Allocasuarina littoralis</i>	Casuarinaceae	Black She-Oak	(Salisb.) L.A.S.Johnson
<i>Aphananthe philippinensis</i>	Ulmaceae	Rough-leaved Elm	Planch.
<i>Dysoxylum fraserianum</i>	Meliaceae	Rosewood	(Juss.) Benth.
<i>Exocarpos latifolius</i>	Santalaceae	Broad-leaved Native Cherry	R.Br.
<i>Ficus coronata</i>	Moraceae	Sandpaper Fig	Spin
<i>Helicia glabriflora</i>	Proteaceae	Smooth or Pale Helicia	F.Muell.
<i>Monotoca elliptica</i>	Ericaceae	Tree Broom-heath	(Sm.) R.Br.
<i>Pararchidendron pruinsum</i>	Fabaceae	Snow Wood	(Benth.) I.C.Nielsen
<i>Pittosporum revolutum</i>	Pittosporaceae	Wild Yellow Jasmine	Dryand. ex W.T.Aiton
<i>Syzygium francisii</i>	Myrtaceae	Giant Water Gum	(F.M.Bailey) L.A.S.Johnson
<i>Syzygium moorei</i>	Myrtaceae	Coolamon	(F.Muell.) L.A.S.Johnson
<i>Wilkiea huegeliana</i>	Monimiaceae	Veiny Wilkiea	(Tul.) A.DC.
<i>Chrysanthemoides monilifera</i>	Asteraceae	Bitou Bush	(L.) Norl.
<i>Claoxylon australe</i>	Euphorbiaceae	Brittlewood	Baill.

<i>Commersonia bartramia</i>	Malvaceae	Brown Kurrajong	(L.) Merr.
<i>Cryptocarya triplinervis</i>	Lauraceae	Three-veined Cryptocarya	R.Br.
<i>Ficus obliqua</i>	Moraceae	Small-leaved Fig	G.Forst.
<i>Macrozamia communis</i>	Zamiaceae	Burrawang	L.A.S.Johnson
<i>Notelaea venosa</i>	Oleaceae	Veined Mock-olive	F.Muell.
<i>Pittosporum multiflorum</i>	Pittosporaceae	Orange Thorn	(A.Cunn. ex Loudon) L.W.Cayzer et al.
<i>Trema tomentosa</i>	Ulmaceae	Native Peach	(Roxb.) H.Hara

Chapter five

Temporal variation in a plant-animal network

Ray A.J. Blick, Kevin C. Burns & Angela T. Moles

“Nestled among those haphazardly scattered stars are patterns. A lion here, a dipper there. The ability to detect patterns can be both a strength and a weakness... ..Among all the patterns of nature, how do we distinguish the meaningful ones?”

Quote from Leonard Mlodinow (2008), *The Drunkards Walk*, pp 21.

This paper was conceived, analysed and written by RB. Data were contributed by KB. Comments on an early draft were provided by all co-authors.

Abstract

Network analysis can depict interactions between species over short timeframes; however, it is less clear whether we can use network theory to evaluate variability among interactions throughout the course of data collection. The aim of this study is to quantify variability in a plant-animal network by detecting interactions that change annually in a frugivore network spanning six sequential years in a wildlife reserve in New Zealand. We used five commonly employed indices to quantify network properties covering positive and negative co-occurrence patterns (nestedness and web specialisation, respectively) and we use a heat map to depict species interactions that are changing frequently. Frugivore interactions changed from year-to-year, formed inconsistent patterns of nestedness, and we found new interactions forming between previously recorded species even in the final year of data collection. Overall, our results suggest that temporal variation can generate contrasting network properties. Even though we did not determine the reason these changes took place, we suggest that measuring variability under a network framework shows promise for monitoring functional attributes of ecosystems. We hypothesise that interaction turnover plays a functional role in the stability and architecture of ecological networks.

Introduction

Network theory has been applied to ecological communities to identify the frequency that species, such as pollinators, visit the available resources (Bascompte et al. 2003, Guimarães et al. 2007, Blüthgen et al. 2008, Vázquez et al. 2009a, Ings et al. 2009, Vázquez et al. in press). Network analysis is carried out by summing the total frequency of species' interactions throughout the course of the study, thereby eliminating evidence of interaction flexibility, fragility or extinction. This is potentially problematic, because previous research has shown convincingly that interactions change frequently within a regional species pool (Morales and Vázquez 2008), with size of available habitat (Burkle and Knight 2012) and some interactions are regulated by diurnal rhythm (Baldock et al. 2011). However, these concerns are magnified if interactions change over longer time periods (e.g. annually) – a limitation that could influence how researchers perceive or predict network collapse and species extinction events (Petandidou et al. 2008, Dupont et al. 2009, Vázquez et al. 2009a, Díaz-castelazo et al. 2010, Gibson et al. 2010, Chacoff et al. 2011).

Network variability is often considered at the community level. However, arguably more important to network architecture are the interactions between individuals (e.g. hierarchical interactions from trees to communities; Burns and Zotz 2011). For example, a species or individual that is competitively excluded from a primary resource will then visit a secondary preference, and so on (Pires et al. 2011). The major concern here is that an interaction loss is not accounted for. The notion that species interactions are lost (or shift among resources) is a less tangible concept under the network paradigm. This is because species are always seen interacting with certainty, and a true loss is only inferred from variation after the study is completed (Alarcon et al. 2008,

Díaz-castelazo et al. 2010, but see Petandidou et al. 2008). We suggest that disentangling interaction frequency from interaction variability (by way of species accumulation and novel interactions) may offer new insight into the topology of network interactions.

In this study we investigate a plant-animal network including frugivorous birds which have revealed nested interactions in other ecosystems (Reid and Armesto 2011, Wangworn et al. 2011). A wide range of other species interactions are also considered nested, including plant-pollinator (Bascompte et al. 2003, Stang et al. 2006), epiphyte-tree (Blick and Burns 2009, Piazzon et al. 2011, Silva et al. 2011), clownfish–anemone (Ollerton et al., 2007), ant–plant (Guimarães et al. 2006) and fruit–frugivore mutualisms (Lázaro et al., 2005). Importantly, it is suggested that community-level interactions that are nested are more stable and less likely to collapse. The reason for this prediction is that nested interactions are formed by a core set of generalist species that interact together, while maintaining interactions with many rare species (Bluthgen 2010). Under this scenario, if any species goes extinct a majority of interactions remain intact; however, species that have evolved unique traits may go extinct (Olesen et al. 2011, Encinas-Viso et al. 2012). There are however, a multitude of reasons why frugivores might change resources, including physiological and behavioural factors relating to breeding; problems which may be further inflated by sampling effort and the likelihood of detecting less common bird species (Morales and Vázquez 2008). Despite these limitations, an important step for network analysis is to include the likelihood that network interactions change and to evaluate uncertainty in community structure using this method. More specifically, with ever increasing numbers of publications revealing

nested interactions it is necessary to evaluate the possibility that ecological communities are as stable as theory predicts.

In summary, we evaluated a frugivore network over six sequential years in a New Zealand wildlife reserve. We predict that ecological networks have variable and non-variable regions with few common persistent interactions and many weak variable interactions.

Specifically we tested the following hypotheses:

1. The stability of an ecological network is transient annually owing to a rise and loss of species interactions.
2. Over multiple years, bird species that are common in the area will eventually be observed and recorded visiting rare plant species.
3. The overall stability of an ecological network will be driven by large interannual variation among rarely observed species.

Methods

All data were recorded over six years in Zealandia (previously known as Karori Wildlife Sanctuary) situated 2 km south of Wellington on the south coast of North Island, New Zealand (41°18.3' S, 174°44.8' E). Zealandia is a wildlife reserve enclosed in an 8.6 km predator-proof fence designed to exclude mammalian predators and restore native flora and fauna (visitzealandia.com; viewed 27/07/2013). Zealandia was chosen for this study because the reserve contains common and reintroduced native bird species and defined tracks within a single valley that makes species identification possible from a distance. The vegetation community consists of canopy emergent trees (e.g.

Rewarewa; *Knightia excelsa*), a dense canopy of broadleaf and conifer species (e.g. Hinau; *Elaeocarpus dentatus* and Totara; *Podocarpus totara*), and a mixed understory of climbing plants (e.g. Rata; *Metrosideros diffusa*), ferns (e.g. Silver fern; *Cyathea dealbata*) and woody shrub species (e.g. Kohekohe; *Dysoxylum spectabile*). A detailed description of this site can be found elsewhere (Blick et al. 2008). All species interactions were observed twice a week on random days between 10 am and 3 pm for six years with the data collection ending 22nd October 2011. All data were recorded by Kevin C. Burns. Observations were conducted along walking tracks and interactions were confirmed visually using binoculars. During each day, a path of approximately 4 km was followed and all bird species observed removing fruit were identified and recorded. To ensure individual birds were not double counted, no more than three individual birds were recorded during each observation. Observations were compiled each year to generate a quantitative interaction network and later pooled to form a pooled network.

Network analysis

Species interactions were evaluated both annually, and as accumulated data. All matrices were arranged with bird species representing columns and plant species representing rows. All matrices were populated with the number of times each species was observed interacting.

All networks were assessed using five common indices from the *bipartite* package in R, including nestedness, web specialisation, connectance, c-score and the number of compartments. Nestedness is a measure of order; where a core set of generalist species interact together while maintaining a wider set of specialist species (Blüthgen 2010). A

nested arrangement is depicted by strong interactions at the top left corner of an interaction matrix and evaluated in this study as departures from perfect order using the 'nest' function.

Web specialisation is a measure of network connectance; where the number of connections are either more or less than expected given the abundance of each species (i.e., row and column totals). Web specialisation is depicted by strong infrequent interactions with no specific order and is quantified using the 'H2' function.

Connectance is a supporting measure of web specialisation; where all realised interactions (1s and 0s) are determined from all possible interactions in the network (row length x column length). Connectance was used to evaluate how network fill changes in response to each network approach. C-score is a measure of negative co-occurrence between species pairs (Stone and Roberts 1990); where certain species are not observed in the same location or interacting with the same resource. C-score is depicted as a checkerboard arrangement and evaluated using the 'C.score' function.

Compartmentalisation is a measure of subsets within a network; where certain species assemblages interact frequently. Compartments are also depicted as a checkerboard arrangement (with multiple species) and evaluated using the 'compart' function.

Therefore these network indices cover a wide range of properties that either support the prediction that many species share resources (e.g. nestedness), or species that only ever visit some, but not all, plant species in the community. We used five commonly employed network indices because they cover a wide range of properties from web specialisation to nestedness

A null model was employed to compare observed interactions with that expected by chance (Gotelli 2000). All calculations were evaluated so that the mean simulated index (e.g. mean c-score) is centred at zero and an observed index is a measurable distance away from the mean (Z score). Null model evaluations are susceptible to changes in network size. Therefore, we constrained all simulations so that column totals (bird species) were fixed and row totals (plant species) were allowed to vary. These constraints are biologically intuitive: a bird may select a different tree in the randomised community, but the bird cannot become a different species. Furthermore, this approach maintains the abundance distribution of the higher trophic level. This is important because it means that each randomisation event has the potential to deviate from the observed network structure while maintaining network size. All observed indices were compared to 1000 simulations generated using the *vegan* package in R (*permatfull* function; Oksanen et al. 2012) (see R code in Appendix 2).

Species accumulation underpinning network topology

Species accumulation and novel interactions were evaluated using rarefaction curves computed with the *vegan* package in R (R core team 2012). Rarefaction curves were generated from 1000 simulations and plotted with 95 % confidence intervals. A fitted spline curve was used to calculate the number of observations required to record an additional species using the *stats* package in R (*predict* function). The *predict* function approximates an unknown *Y* value by using a linear progression from the final set of data points. We consider a linear progression satisfactory because we are characterising the final shape of each curve and comparing higher and lower trophic levels. For example, after six years, both trophic levels (birds and plants) may contain a different

number of recorded species and still predict the same number of observations required to find a new species.

Variable regions of network topology

Species interactions are predicted to change annually. Inter-annual variation was evaluated using the coefficient of variation (standard deviation divided by the mean) from data recorded annually (n=6). We tested whether interactions were more variable than expected by chance by calculating their dissimilarity from expected values (i.e., observed minus expected). Positive values represent interactions that are more variable than expected by chance, while negative values represent interactions that are less variable than expected by chance. The expected variation for each interaction was computed from 1000 iterations following the same permutation protocols as network analysis (above). Non-linear regression evaluated the relationship between variation and interaction frequency. CV was depicted using a heat map produced from the *lattice* package in R (R core team 2012).

Results

The plant-animal network contained 3353 interactions between 12 bird species and 35 plant species (see Appendix 1 for species names). The final interaction matrix had 36.2% of all possible interactions (152 of 420). Each year had between 494 and 709 observations (mean = 528) which filled between 17.1% and 25.1% of the network. Annual observations included at least 9 bird species (9-11) and 20 plant species (20-25), and only 7.4% (31) of species interactions occurred consistently between years.

Network size and fill differed between years, or though we considered annual datasets comparable because the number of interactions (i.e., the number of occupied cells) were not correlated with network size (i.e., the total number of cells; $r^2 = -0.062$, $F_{1,4} = 0.705$, $p = 0.448$), and there was no correlation between standardised effect size (SES) and network size ($r^2 = 0.263$, $F_{1,4} = 2.788$, $p = 0.170$), or SES and the number of interactions ($r^2 = 0.238$, $F_{1,4} = 0.083$, $p = 0.854$).

Network analysis

Pooled data containing interactions from six years was not nested ($t = 26.675$, $z = 2.345$, $p = 0.991$ Figure 1), and was considered highly specialised ($H^2 = 0.254$, $z = 124.172$, $p < 0.001$) with fewer interactions than expected by chance (observed = 0.362, $z = -36.593$, $p < 0.001$). Observed interactions maintained a higher c-score than predicted by chance ($cs = 0.300$, $z = -1.866$, $p = 0.031$). When we considered each year independently, we found two years that were significantly nested (*year 1*: $t = 13.308$, $z = -2.163$, $p = 0.015$; *year 6*: $t = 8.122$, $z = -3.367$, $p < 0.001$, Figure 1a). However, all years were significantly specialised ($H^2 = 0.249-0.339$, $p < 0.001$) and maintained fewer interactions than expected by chance (observed = 0.171-0.225, $p < 0.001$). When data were accumulated over six years; such that year one was added to year two, which was subsequently added to year three and so on, we found only one year that was significantly nested (*year 1*: $t = 13.308$, $z = -2.163$, $p = 0.015$). All years were significantly specialised ($H^2 = 0.254 - 0.284$, $p < 0.001$) and maintained fewer interactions than expected by chance (obs = 0.225 - 0.362, $p < 0.001$).

Species accumulation

The number of bird species recorded in Zealandia rapidly approached an asymptotic point, while the number of plant species did not (Figure 2). More specifically, we were five times more likely to find another plant species (est. days = 438) than we were to finding an additional bird species (est. days = 2192). Subsequently, the number of novel links in the network did not reach an asymptotic point (Figure 2). On average, it would take a further 59 days under the current sampling regime to find a new interaction.

Network variability

Common interactions were less variable than could be predicted by random events, and connections that were furthest from the top left corner (i.e., the packed region after being reordered) had the highest rate of variation (Figure 3). That is to say, an interaction matrix that is arranged by frequency of occurrence might also indicate interactions with a higher probability of disappearing (but not going extinct) in the following year.

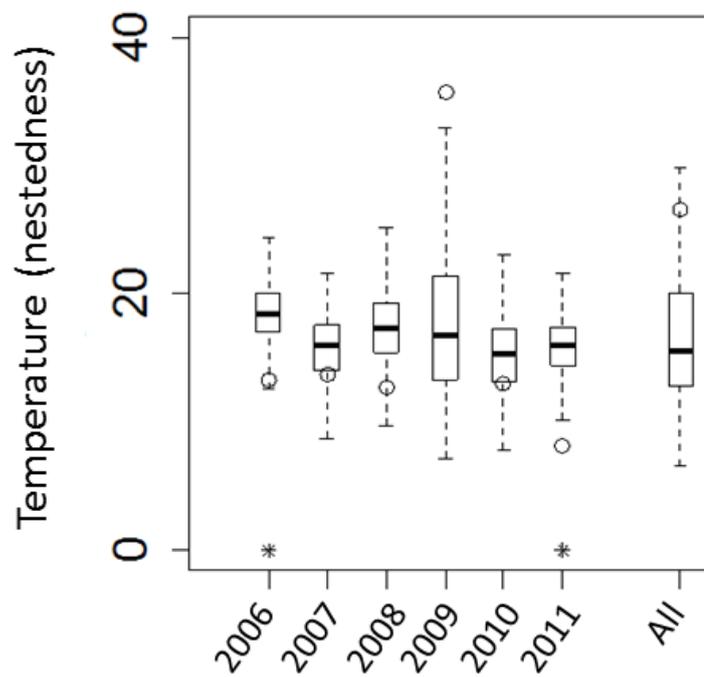


Figure 1 Patterns of nestedness for each year of this study. Temperature is an index used to measure nestedness in a reordered matrix. Species interactions were significantly nested in 2006 and 2011. All simulated data are represented by boxplots and the dashed lines indicate the distribution of expected values. Circles represent observed values, while stars indicate the years that were significantly nested.

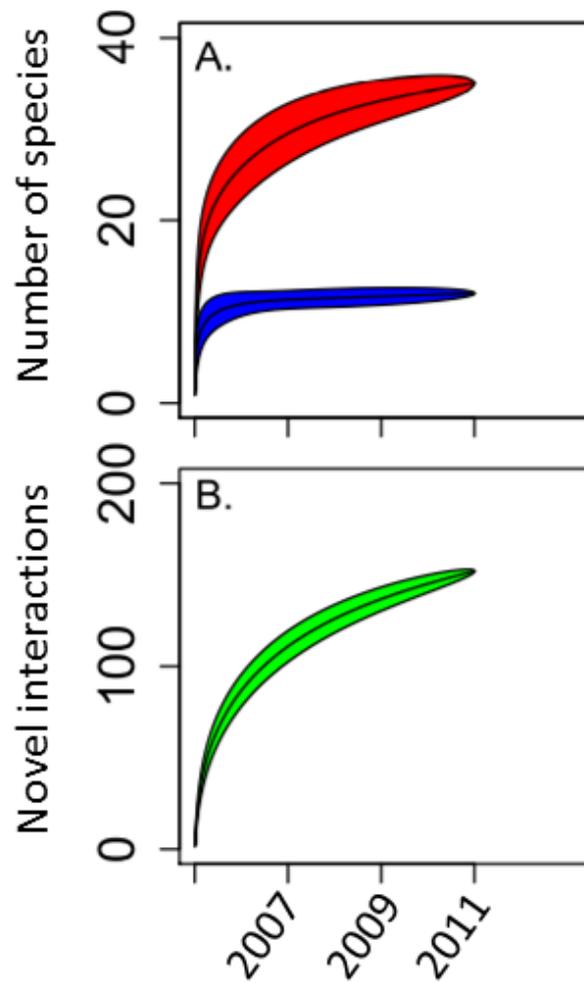


Figure 2 Patterns of species accumulation over six years of data collection. Rarefaction curves indicate different patterns of species accumulation for birds (blue) and plants (red; A). Differences in the higher trophic level (birds) had a corresponding effect on novel interactions (green; B). Coloured polygons represent 95% confidence intervals for each curve.

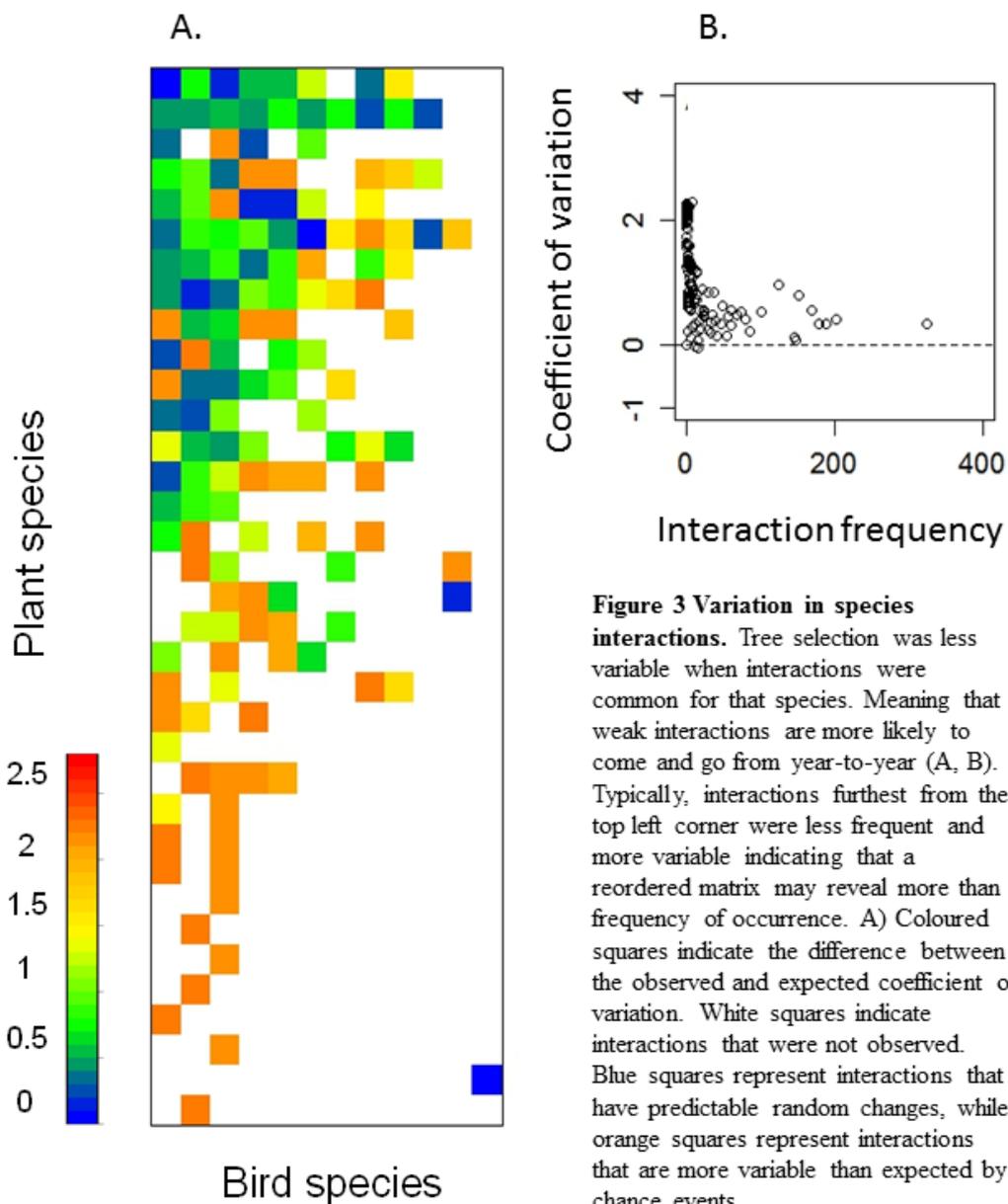


Figure 3 Variation in species interactions. Tree selection was less variable when interactions were common for that species. Meaning that weak interactions are more likely to come and go from year-to-year (A, B). Typically, interactions furthest from the top left corner were less frequent and more variable indicating that a reordered matrix may reveal more than frequency of occurrence. A) Coloured squares indicate the difference between the observed and expected coefficient of variation. White squares indicate interactions that were not observed. Blue squares represent interactions that have predictable random changes, while orange squares represent interactions that are more variable than expected by chance events.

Discussion

Our results show that a frugivore network can have very different patterns of connectivity from year to year. We found significantly different results when we considered annual data. That is, only two out of six years were considered nested, and after accumulating interactions sequentially, only one year (2006) was considered nested. After pooling all data together, frugivore interactions were considered specialised (i.e., fewer interactions than expected given the abundance of each species) which shows little correspondence with previous findings in other ecosystems (Reid and Armesto 2011, Wangworn et al. 2011). Overall, we suggest that community-level patterns, such as nestedness, might miss important changes in network architecture and resilience across the ecosystem. That is to say, finding nestedness in 2006 and web specialisation in 2007, is less informative than the frequency each species interacts with their environment.

How novel interactions affect network topology is poorly understood. In this study we show that the accumulation of novel interactions was mostly due to interactions from bird species already considered part of the network. In fact the total number of bird species was exhausted after two years, while the number of plant species continued to increase over six years. The accumulation of novel interactions from existing bird species is logically predicted to influence network topology, by increasing the number of occupied cells in a data matrix, and subsequently decreasing the likelihood of web specialisation. This was not the case in our dataset; even after six years network topology remained more specialised than predicted by chance. Importantly, network topology did change from a nested pattern to web specialisation over the first two years

of this study suggesting that the accumulation of data over an extended period can have a powerful effect on how we perceive community structure.

In this study we expected to find nested patterns (Reid and Armesto 2011 and Wangworn et al. 2011) because an increase in field time and accumulation of data would lead to the detection of common bird species visiting rare, less abundant plant species in the community; rather than the accumulation of new species altogether. In six sequential years we found just 7% of all species interactions reoccurring each year. This result is consistent with other mutualistic networks that have found between 5% and 31% of reoccurring interactions (reviewed by Vázquez et al. 2009a). However, an important distinction is that plant-pollinator networks have been shown not to change the degree of order (ranked position of each interaction) from year to year (Alarcon et al. 2008, Díaz-castelazo et al. 2010) – a result that was found in this study. We suggest that an increase in novel interactions and changing network topology from year to year indicates that interactions are both gained and lost annually.

Analysis of network variability showed that some interactions were more likely to persist than others. Network interactions that were most abundant in the frugivore network were less variable and positioned in the top left corner of the interaction matrix. We suggest that species turnover and the position of novel interactions in an interaction matrix are important factors underpinning network topology and we hypothesise that variability between species pairs that are present from one year to the next indicates the degree of determinism in the community. The dominant pattern observed for this network, web specialisation, may occur because forbidden links inhibit the possibility of nestedness. That is, fruit removal by different bird species may be constrained by gape

size, fruit phenology, and preferential selection (Wheelwright 1985). Forbidden interactions are an important factor determining other networks, including ant assemblages (Olesen, J.M. et al. 2011, Guimarães, P.R. et al. 2007.), and pollination events (Stang et al. 2006, 2009).

An equally plausible hypothesis regarding network structure follows that sampling effort inhibited our ability to fully resolve network interactions. For example, the data may underestimate true interactions among birds and plants. The study area surrounds an inactive reservoir that no longer feeds the metropolitan area – an area that is occupied by a wide range of bird species with diets that do not extend to native fruit. However, with enough observations it is likely that some of these species that occupy open visible areas would eventually be observed exploring less common food choices. This occurred in our dataset; we observed an unlikely association between the Mallard duck (*Anas platyrhynchos*) and *Pratia* spp. This is a striking result when we consider other unobserved bird species, including the Shining Cuckoo (*Chrysococcyx lucidus lucidus*) which is common to the area but less obvious to see and observe foraging among native trees. However, in this study we did not measure sampling effort or the uncertainty underpinning each interaction. Uncertainty in data collection may also extend to our method of standardizing interactions between flocks and solitary birds. These limitations in sampling effort are untested hypotheses regarding the dataset and require further testing and evaluation. However, this study remains one of the longest of its kind, spanning six years, and we suggest that interaction variability must be explored in further experimental studies to ensure network stability is not overestimated.

Many ecological networks evaluate interactions across a single season or year. In this study we increased our sampling effort to a period spanning six years which increased our chances of observing rare interactions (Petandidou, T. et al. 2008). Rare interactions, such as long distance dispersal events (Nathan 2006 and references within) or resource mediated selection (i.e. following optimal diet theory or an ideal free distribution; see Pires et al. 2011) are ecologically important and meaningful to the interpretation of community structure. Quantifying the amount of data necessary to evaluate the role of rare events on network structure is an important goal for future work.

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References

- Alarcon, R. et al. 2008. Year-to-year variation in the topology of a plant–pollinator interaction network. - *Oikos* 117: 1796–1807.
- Baldock, K.C.R. et al. 2011. Daily temporal structure in African savanna flower visitation networks and consequences for network sampling. – *Ecology* 92: 687-698.
- Bascompte, J. et al. 2003. The nested assembly of plant-animal mutualistic networks. - *Proceedings of the National Academy of Sciences USA* 100: 9383-9387.
- Blick, R. et al. 2008. Successional dynamics after pest eradication in the Karori Wildlife Sanctuary. - *New Zealand Natural Sciences* 33: 3–14.
- Blick, R. & Burns, K.C. 2009. Network properties of arboreal plants: Are epiphytes, mistletoes and lianas structured similarly? - *Perspectives in Plant Ecology, Evolution and Systematics* 11: 41-52.

- Blick, R.A.J. & Burns, K.C. 2011. Liana co-occurrence patterns in a temperate rainforest. – *Journal of Vegetation Science* 22: 868-877.
- Burns, K.C. & Zotz, G. 2010. A hierarchical framework for investigating epiphyte assemblages: networks, meta-communities, and scale. – *Ecology* 91: 377-385.
- Burns, K.C. 2012. Are introduced birds unimportant mutualists? A case study of frugivory in European blackbirds (*Turdus merula*). – *New Zealand Journal of Ecology* 36: 171-176.
- Blüthgen, N. et al. 2008. What do interaction network metrics tell us about specialization and biological traits? - *Ecology* 89: 3387-3399.
- Blüthgen, N. 2010. Why network analysis is often disconnected from community ecology: A critique and an ecologist's guide. - *Basic and Applied Ecology* 11: 185-195.
- Campanello, P.I. et al. 2007. Lianas in a subtropical Atlantic forest: host preference and tree growth. - *Forest Ecology and Management* 242: 250-259.
- Carlo, T. & Yang, S. 2011. Network models of frugivory and seed dispersal: Challenges and opportunities. - *Acta Oecologica* 37: 619-624.
- DeWalt, S.J. et al. 2006. Liana habitat associations and community structure in a Bornean lowland tropical forest. - *Plant Ecology* 186: 203-216.
- Dormann, C.F. et al. 2009. Indices, Graphs and Null models: Analyzing Bipartite Ecological Networks. - *The Open Ecology Journal* 2: 7-24.
- Díaz-castelazo, C. et al. 2011. Changes of a mutualistic network over time: reanalysis over a 10-year period. - *Ecology* 91: 793-801.
- Encinas-Viso, F. et al. 2012. Phenology drives mutualistic network structure and diversity. – *Ecology Letters* 15: 198-208.
- Gotelli, N.J. 2000. Null model analysis of species co-occurrence patterns. - *Ecology* 81:

2606-2621.

- Guimarães, P.R. et al. 2007. Interaction intimacy affects structure and coevolutionary dynamics in mutualistic networks. - *Current Biology* 17: 1797-1803.
- Hijmans, R.J. et al. 2005. Very high resolution interpolated climate surfaces for global land areas. – *International Journal of Climatology* 25: 1965–1978.
- Ings, T.C. 2009. Ecological networks – beyond food webs. - *Journal of Animal Ecology* 78: 253-269.
- Jordano, P. 1987. Patterns of mutualistic interactions in pollination and seed dispersal: connectance, dependence asymmetries, and coevolution. - *American Naturalist* 129: 657-677.
- Morales, J.M. & Vázquez, D.P. 2008. The effect of space in plant-animal mutualistic networks: insights from a simulation study. – *Oikos* 117: 1362-1370.
- Nathan, R. 2006. Long-distance dispersal of plants. *Science* 11: 786-788.
- Petandidou, T. et al. 2008. Long-term observation of a pollinator network: fluctuation in species and interactions, relative invariance of network structure and implications for estimates of specialization. – *Ecology Letters* 11: 564-575.
- Oksanen, J. et al. 2012. vegan: Community Ecology Package. R package version 2.0-3. <http://CRAN.R-project.org/package=vegan>.
- Olesen, J.M. et al. 2011. Missing and forbidden links in mutualistic networks. - *Proceedings of the Royal Society B-Biological Sciences* 278: 725-732.
- Pires, M.M. et al. 2011. The nested assembly of individual-resource networks. *Journal of Animal Ecology* 80: 896-903.
- R Development Core Team (2010) R: a language and environment for statistical computing. Vienna, Austria. <http://www.R-project.org>.

- Reid, S. & Armesto, J.J. 2011. Interaction dynamics of avian frugivores and plants in a Chilean Mediterranean shrubland. – *Journal of Arid Environments* 75: 221-230.
- Rhodes, J.R. & Jonzén, N. 2011. Monitoring temporal trends in spatially structured populations: how should sampling effort be allocated between space and time? – *Ecography* 34: 1040-1048.
- Schnitzer, S.A. 2005. A mechanistic explanation for the global patterns of liana abundance and distribution. - *American Naturalist* 166: 262-276.
- Schnitzer, S.A. & Bongers, F. 2002. The ecology of lianas and their role in forests. - *Trends in Ecology and Evolution* 17: 223-230.
- Sfair, J.C. et al. 2010. Nested liana-tree network in three distinct neotropical vegetation formations. - *Perspectives in Plant Ecology, Evolution and Systematics* 12: 277-281.
- Stone, L. & Roberts, A. 1990. The checkerboard score and species distributions. - - *Oecologia* 85: 74-79.
- Wangworn, S. et al. 2011. Plant-frugivore interactions in an intact tropical forest in north-east Thailand. – *Integrative Zoology* 6: 195-212.
- Wheelwright, N.T. 1985. Fruit-Size, Gape Width, and the Diets of Fruit-Eating Birds. - *Ecology* 66: 808-818.
- Tylianakis, J.M. et al. 2010. Conservation of species interaction networks. - *Biological Conservation* 143: 2270-2279.
- Ulrich, W. & Gotelli, N.J. 2007. Disentangling community patterns of nestedness and species co-occurrence. - *Oikos* 116: 2053-2061.
- Vázquez, D.P. et al. 2009a. Uniting pattern and process in plant-animal mutualistic networks: a review. - *Annals of Botany* 103: 1445-1457.
- Vázquez, D.P. et al. 2009b. Evaluating multiple determinants of the structure of plant-animal mutualistic networks. - *Ecology* 90: 2039-2046.

Vázquez, D.P. et al. In press. The strength of plant-pollinator interactions. – *Ecology*
[doi:<http://dx.doi.org/10.1890/11-1356.1>].

Zahng, F. et al. 2011. An interaction switch predicts the nested architecture of
mutualistic networks. – *Ecology Letters* 14: 797-803.

Appendix 1

Species names

Birds: *Turdus merula* (Blackbird), *Anthornis melanura* (Bellbird), *Notiomystis cincta* (Stitchbird), *Nestor meridionalis* (Kaka), *Anas platyrhynchos* (Mallard), *Hemiphaga novaeseelandiae* (Wood Pigeon), *Philesturnus carunculatus* (Saddleback), *Turdus philomelos* (Song Thrush), *Sturnus vulgaris* (Starling), *Prosthemadera novaeseelandiae* (Tui), *Zosterops lateralis* (Waxeye), *Mohoua albicilla* (Whitehead).

Plants: *Aristotelia serrata*, *Berberis darwinii*, *Coprosma repens*, *C. grandifolia*, *C. robusta*, *Cordyline australis*, *Coriaria arborea*, *Corokia cotoneaster*, *Elaeocarpus dentatus*, *Fuchsia excorticata*, *Geniostoma rupestrae*, *Hedycaria arborea*, *Lonicera japonica*, *Melicytus ramiflorus*, *Muehlenbeckia sp.*, *Myoporum laetum*, *Myrsine australis*, *Passiflora sp.*, *Pennantia corymbosa*, *Piper excelsa*, *Pittosporum eugenoides*, *P. tenuifolium*, *Podocarpus totara*, *Pratia*, *Pseudopanax arboreus*, *Pseudopanax crassifolius*, *Ripogonum scandens*, *Rubus cissoides*, *R. ideal*, *R. procerus*, *Schefflera digitata*, *Solanum sp1*, *Solanum sp2*, *Fragaria sp.*, *Syzygium sp.*.

Appendix 2

R code

Randomisation script

requires the 'vegan' and 'bipartite' packages

```
for (i in 1:1000) {
  data.set<-mydata
  randomise<-permatfull(x, fixedmar = "column", shuffle = "ind", strata = NULL,
    mtype = "count", times = 1)

  nest<-nestedness((as.data.frame(randomise$perm)),null.models=FALSE,n.nulls=0)
  cs<-C.score((as.data.frame(randomise$perm)),normalise=TRUE)
  Web.spec<-as.vector(H2fun((as.data.frame(randomise$perm))))
  comp<-compart(as.data.frame(randomise$perm))
  connectance<sum(ifelse(as.data.frame(randomise$perm)>=1,1,0))/
    (length(data.set[1,])*length(data.set[,1]))

  indices<-c(nest$temperature,cs,Web.spec[1],comp$n.compart,connectance)
  write(indices, file="C:\\ Desktop\\results.txt",append=TRUE)
}
```

Heat map script

requires the 'lattice' package

```
rgb.palette <- colorRampPalette(c("blue", "green","yellow","orange","red"), space =
"rgb")
levelplot(as.matrix(t(mydata[35:1,1:12])),main="",ylab="Host trees", xlab="Vines",
scale=list(y=list(draw=F), x=list(draw=F)), col.regions = rgb.palette(120),
at=seq(0,2.8,0.1), colorkey=FALSE)
```

Chapter Six

Discussion: challenges and opportunities

“[John Tukey] complained of “a natural, but dangerous desire for a unified approach” [and explained that] “the greatest danger I see from Bayesian analysis stems from the belief that everything that is important can be stuffed into a single quantitative framework”.*”

*Quote from Sharon Bertsch Mcgrayne (2011). The theory that would not die: how Bayes’ rule cracked the enigma code, hunted down Russian submarines and emerged triumphant from two centuries of controversy, pp 170.

Synthesis, concepts and writing by RB.

The aim of this thesis was to evaluate ecological networks, discuss their limitations, and advance the understanding of plant-plant interactions. First I asked whether biological traits can predict species interactions. Here I applied a deterministic model to test leaf trait similarity between mistletoes and host trees. Second, I asked whether one trophic level could influence network topology. Resource limitation is an important aspect of all networks; subsequently I tested for the first time whether plant-plant networks (mistletoes) were dependent on how many suitable host trees were present in the community. To extend our understanding in plant-plant networks I asked in a different ecosystem whether abundant trees and coincidental associations could lead us to make a mistake about deterministic processes. That is, I asked whether vine-host interactions were in fact deterministic or whether interactions were correlated with overlapping distributions. Even though a major focus of this thesis was to understand plant-plant interactions, I also tested variability in a plant-animal network to explore a less known area of network analysis; temporal variation of species interactions. The results from this thesis show that ecological networks depict specialised, sometimes coincidental and variable interactions for different species in different environments.

Network analysis: considerations

Results from my thesis show that ecological networks are a useful statistical and graphical tool for assessing species interactions in ‘finite’ communities (when ecological boundaries are known), offering exciting potential for mapping functional characteristics, such as species interactions in conservation monitoring. However, researchers implementing network theory must make several assumptions that limit universal application, especially in plant-plant networks. Arguably the most important limitation of network theory is that a mechanistic explanation for network structure

must be inferred from pattern analysis, rather than from an explicit test of ecological processes. For example, in chapter two I investigated leaf shape similarity between mistletoes and host trees, while in chapter three I focussed on interactions between mistletoes and trees according to host availability. In both studies, I concluded that mimicry and tree availability were not correlated, or did not determine, interaction strength. These conclusions were inferred from spatially implicit datasets (i.e. no spatial data was included) thereby assuming that all players in the network have the opportunity to interact. This assumption is realistic for ‘mobile’ networks, such as plant-pollinator networks; however it may be less realistic for plant-plant networks.

The deterministic approach: leaf traits of mistletoe and host trees

Prior to my thesis, very few studies using network analysis had used biological traits to predict species interactions (e.g. floral depth; Stang et al. 2009). In this thesis I tested a ‘deterministic’ model that predicted species interactions according to leaf trait similarity (size, shape and colour). Rather than using a null model approach, I hypothesised that ecological network analysis could reveal ‘unique’ similarities in leaf traits - a theoretical equivalent to finding forbidden links in plant-animal networks (e.g. a proboscis length too short for some floral tubes). Importantly, I showed that ecological networks can be used in such a way that trait variation could be analysed statistically across species interactions. Subsequently this chapter has been published in *Oikos*; providing a conceptual framework for future research.

In chapter two I used leaf traits to test the deterministic model. To do this I used the hypothesis that herbivory can select for similar leaf traits (concealing high nutrient load) or dissimilar leaf traits (advertise poor nutrient concentration) between mistletoes and

host trees (Barlow and Wein 1977, Ehleringer et al. 1986, Canyon and Hill 1997). There are however, alternative hypotheses that also explain mistletoe mimicry. For example, avian foraging behaviour may better explain leaf shape similarity in areas that are absent of herbivores (Calder 1983). That is, mistletoe leaves with similar shape and colour to host trees are harder to find which increases the foraging time in neighbouring trees and facilitating dispersal. However, even if the overarching hypothesis is changed from marsupial herbivory to avian foraging, the parameters assigned to the deterministic model remain unchanged; the deterministic model keeps the prediction that unique combinations of leaf traits can be matched to host trees.

Identifying the most plausible hypothesis for leaf shape in mistletoe is worth further investigation; however it was not the primary purpose of this study. More importantly to my study are interpretations derived from the results in light of the relevant hypotheses proposed above. Perhaps, a more plausible inference from these results (that unique traits were not correlated with species interactions) is that leaf traits do not need to be ‘unique’ combinations in the community and only a small amount of concealment can provide adequate visual cues that increase dispersal success in mistletoe populations. In addition, the alternative hypothesis (that avian foraging selects for leaf traits) predicts that easy detection and longer foraging times per tree can lead to clumping and overloading on some, but not all individuals. This might be one explanation why some trees of *Acacia aneura* (needle/terrete leaves) receive disproportionately larger numbers of *Amyema maidanii* (round/spathulate leaves) in certain parts of the landscape surrounding Fowlers Gap (Chapter four).

In this thesis I focused on the methodological approach; can networks be applied in such a way that biological traits can be mapped and depicted at the community level. I showed that leaf traits (such as *Amyema linophylla* on its host *Casuarina glauca*) were no different from ‘hypothetical’ mistletoe-host interactions; such that they do not predict pairwise interactions using network theory.

However, there are several factors (other than visual cues) that may have been more successful in predicting network interactions, including olfactory concealment, fruit conspicuousness (e.g. chromatic and achromatic visual cues), and the distribution of bird species.

Coincidental associations, sampling effort, and the role of chance

Ecological networks need to be interpreted with caution. In chapter three, I asked whether common interactions were determined from tree abundance at Fowlers Gap. More specifically; can one trophic level influence interactions of an ecological network? I concluded that network structure was not determined by tree availability. To examine this question more closely I evaluated a specialised interaction between the pale leaf mistletoe (*Amyema maidenii*) and *Acacia aneura*. Both studies showed support that tree abundance does not limit infection prevalence among trees at Fowlers Gap. Prior to these analyses, all plant-plant interaction networks had omitted the possibility that tree availability could influence interaction frequency. This chapter provides the conceptual framework and the first test of this idea. However, I note here that ecological networks must analyse community-level data by omitting information from a wider geographic area. While this is a common problem to all areas ecology; it remains less clear how community boundaries might affect network topology.

Research carried out in this thesis has uncovered important differences between plant-plant interactions. Previous research found that mistletoes and vines have similar network properties (Blick and Burns 2009, 2011). However, analyses from chapters three, four and five show that these two functional groups interact with individual trees in very different ways regardless of host identification. More specifically, the results show that vines form coincidental interactions with abundant trees in the same location as vines. That is, when vines are abundant or cover a wide distribution they are highly connected in the network – a result not found for mistletoes. Previous hypotheses proposed that host preferences could explain network properties of vines. These results show for the first time that plant-plant networks, such as vines growing on host trees, can lead one to make the mistake that deterministic interactions or causal mechanisms are at play rather than coincidental association between neighbouring plants.

In addition to finding differences between ecosystems with similar network properties, I also show the importance of individual interactions which vary annually (for birds; chapter six) or between forests (for vines; chapter five). The important finding here is that network interactions that are observed and ‘summed’ over short time periods can inhibit any ability for identifying real change in the community. Arguably, this is obvious for vine-host interactions in different forests, because vine species form different associations with host species according to their abundances in different environments. However, the suggestion that plant-animal networks vary from year to year is less clear, and contrasts markedly from other studies. One possible explanation for finding vastly different patterns between years is due to sampling effort and efficiency. Under this hypothesis, we might predict that interactions that were observed during each day were largely coincidental interactions between obvious bird species.

However, it is more plausible that infrequent interactions that are more variable in the network (as depicted in chapter five) are likely to be coincidental interactions. Even so, this dataset spanning six years is one of the largest of its kind and has provided valuable insight into the variation of network interactions; a previously unexplored area of research to network analysis.

Null models: application to ecological network analysis

Since the inception of null models in ecology, researchers have raised concerns about the pitfalls of analyses that employ inappropriate null models (Ulrich 2004). For example, Diamond and Gilpin (1982) suggested that the dilution effect might occur during the null modelling procedure in tests of Diamond's (1975) assembly rules, leading researchers such as Connor and Simberloff (1979), to incorrectly accept the null hypothesis. Similarly, not accounting for spatial constraints can lead researchers to incorrectly reject null hypotheses, for instance in explaining the checkerboard distribution of hunting spider species in dune ecosystems (Peres-Neto et al. 2001). Thus, a modified null model is required to avoid type I and type II errors in the interactions between functional groups. In this thesis I used weighted probabilities to predict species interactions – an approach previously untested among network analyses and the mapping of plant-plant interactions.

It is widely recognised that species with different abundance and spatial distributions can influence ecological networks (Morales and Vázquez 2008, Ulrich and Gotelli 2010, Gilarranz et al. 2012). In chapter four, I demonstrate how species with wider distributions can lead to higher measures of connectance between vines and trees – a finding that is consistent with results from other ecosystems (Morales and Vázquez

2008). However the important contribution from this study involves the analytical procedure prior to assessing network indices. To test the structure of vine-host interactions I developed a modified null model that only allowed species from the same part of the landscape to interact. From this study I showed that a vine host interaction network revealed negative co-occurrence patterns, even after accounting for patterns of distribution and abundance. This result was not supported by individual analyses in three out of nine forests. Together, these results show that species interactions can generate different patterns of connectivity when examined at different spatial scales.

Application of a modified null model is theoretically and biologically important. In this thesis I integrated biological information to test ecological theories such as mimicry (chapter two) and the host-availability hypothesis (chapter three). By adjusting the null model, the network paradigm can provide a means of understanding biological structure in ecological communities. For example, the mistletoe network showed a clumped structure suggesting that certain trees act as a focal point for the facilitation and spread of mistletoe populations. One plausible explanation may be that the movement of frugivorous birds (Rawsthorne et al. 2012) between large trees in the landscape (keystone structures; Manning et al. 2006, Stagoll et al. 2012) can describe the movement of mistletoe seeds to new trees. Resolving which mechanisms foster successful propagation of mistletoe seeds is important for conservation of fragmented landscapes; however, the results from chapter four suggest that tree size is less important than position. In addition, I suggest that the stability of mistletoe-host networks is determined by the quality of individual trees (Watson 2009) that are infected disproportionately across the landscape.

Results from this thesis are important for assessing the stability of ecological networks - which has come into focus over recent years (Thébault and Fontaine 2010). I showed that the distribution of individuals (chapter three) and spatial constraints (chapter four) are important attributes of ecological networks that change frequently (chapter five). Specifically, in chapter five I showed that nestedness was a transient feature of a plant-animal network suggesting that a general relationship between nestedness and stability may not be appropriate. Importantly, topological plasticity may improve community stability because species can switch partners when resources are lost (Ramos-Jiliberto et al. 2012). In support of this concept, I found new interactions forming in the plant-animal network even after six consecutive years of observations, supporting the idea that ecological networks are continuously changing naturally on temporal and spatial scales for different species (see heat map in chapter six).

Opportunities: an experimental approach to ecological networks

In network analyses the structure of the community is considered deterministic if an observed index deviates from random expectations (Bascompte et al. 2003). However, the observational null model approach has recognised limitations and often assumptions need to be made during analysis. For example, the analytical approach must make the assumption that (i) the rank abundance of species does not change through time and (ii) the ecosystem is in a 'final state' of order (Climax; Clements 1936). An important contribution that could be made to test these assumptions is to use an approach that manipulates species interactions either experimentally or through natural observations after disturbance. Understandably, ecological networks are composed of observations that are difficult to acquire without adding the complexity of experimental

manipulation. I outline two feasible studies for testing plant-plant interaction networks below.

Natural changes in the landscape: A case for vine-host interactions

Disturbance is a prominent factor in all ecosystems and an ecological phenomenon that lends itself to studies under the network paradigm. Here I suggest a comparison between pre- and post-disturbance networks to evaluate natural patterns of connectivity between species interactions (Figure 1). This would allow one to ask: if we reset the system repeatedly, does it reassemble in the same way each time, or is it more random?

The Australian flora is well adapted to fire; where large trees survive frequent burning events, and vines and small herbaceous plants are killed (Pekin et al. 2012). During re-establishment, vines could be tracked through time where the pre-disturbed patterns of connectivity are compared statistically with natural recolonisation events. A Mantel test of matrix similarity could be used to compare the connectivity of interactions between pre- and post-disturbance at an individual- and species-level in different habitats and landscapes. A natural observational approach could then evaluate the mechanisms involved in generating network indices such as negative co-occurrence or nestedness, and used to analyse disturbance such as (i) time-since-fire in forest ecosystems and (ii) time-since-closure of mined land.

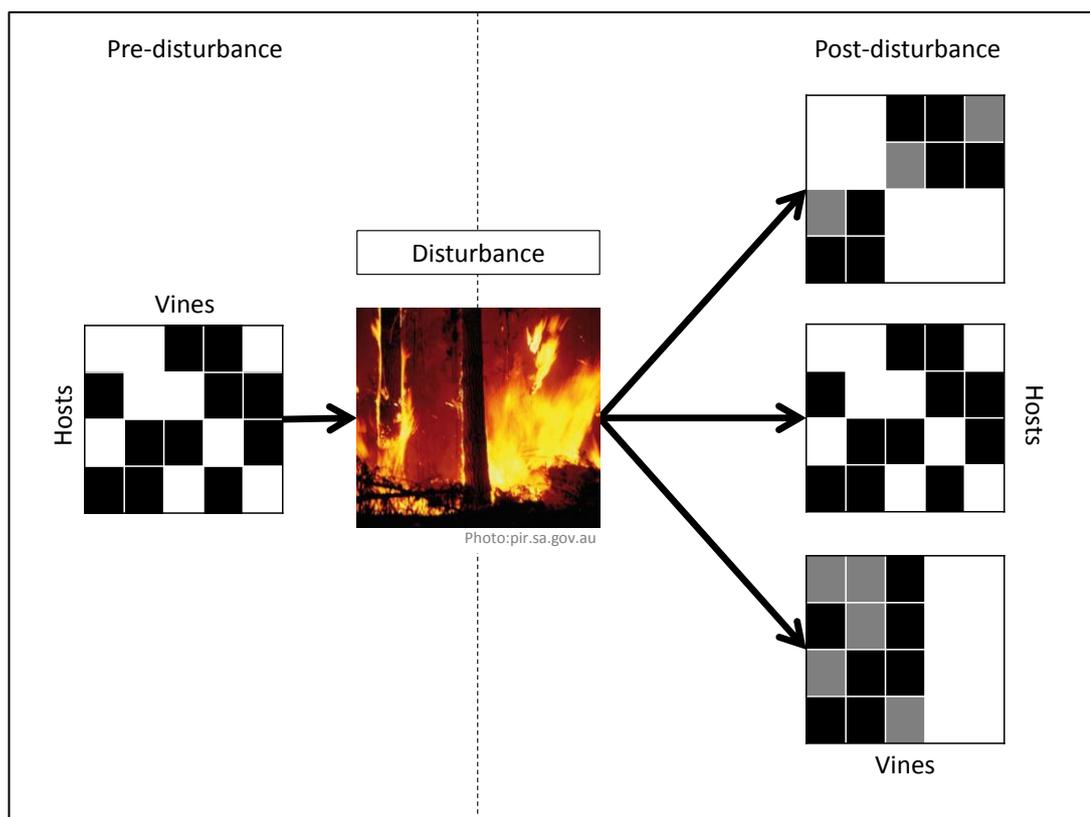


Figure 1 A framework for testing disturbance and ecosystem structure. Filled cells represent interactions and unfilled cells represent absences. Grey cells represent new species interactions in post disturbance networks.

Networks in reverse: A case for mistletoe-host interactions

Ecological networks are all observation based estimates of community structure. An exciting variation of this idea is to integrate an experimental procedure which determines the successional stages of network development. To do this one could test the role of seed dispersal and host immunity in structuring a mistletoe-host network using an experimental manipulation of connectivity. Mistletoe develop through four fundamental stages prior to becoming reproductively mature; (i) dispersal to an appropriate host species, (ii) germination and avoiding seed predators, (iii) making a connection with the host, and (iv) establishing as a seedling and surviving herbivory

(Figure 2). An experimental approach could test the role of each biological process in structuring the network (Figure 3).

To establish the experimental network, fruit from each mistletoe species are collected and artificially dispersed on all available tree species in the community to form a saturated community (removing the dispersal vector). By observing all transplanted seeds through time (one week, three months, one year), the loss in connectivity between species could be attributed to each factor underlying network structure. A Mantel test of matrix similarity could evaluate the structural similarities between the observed network and the artificial network (resolved community). Results from this experiment would indicate the importance of dispersal in structuring keystone species of forest ecosystems. Moreover, this experiment would determine which biological mechanisms are involved in generating negative co-occurrence patterns, web specialisation or modularity in mistletoe-host interaction networks, and advance our understanding of network analysis more clearly.

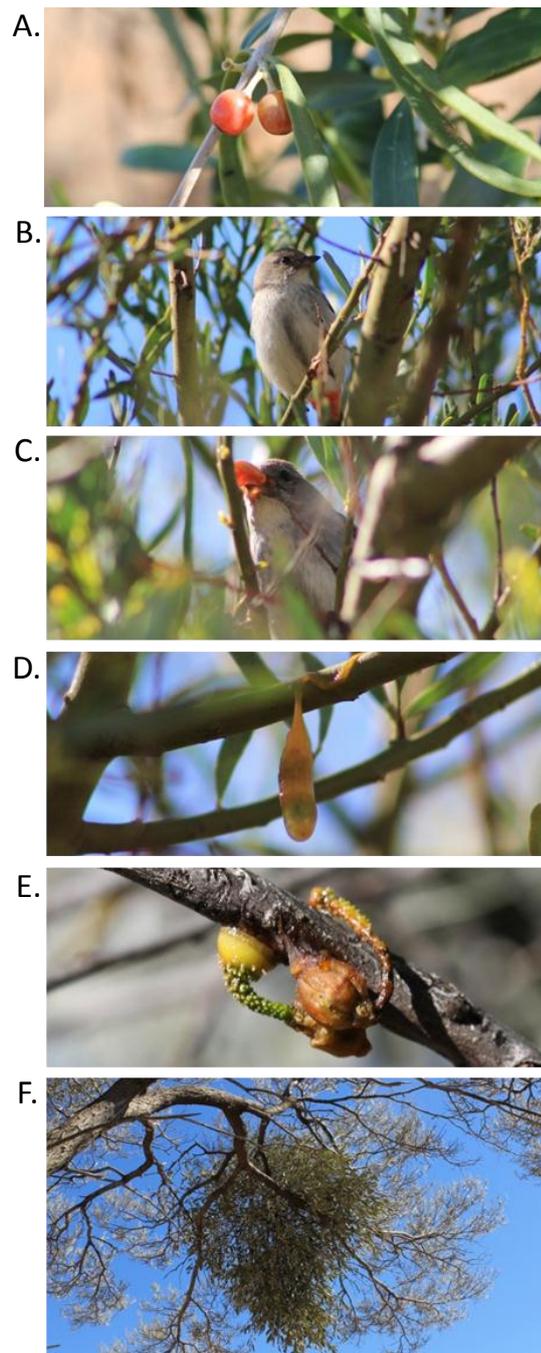


Figure 2 The mistletoe life cycle. Fruit are produced by mistletoe (A; *Lysiana exocarpi*) and consumed by frugivores (B and C; Mistletoe bird). Seeds are dispersed to neighbouring trees (D; mistletoe seeds on *Acacia victoriae*) and establish a parasitic connection (E; *Lysiana exocarpi* seeds infecting *Acacia tetragonophylla*). Plants that survive post seedling herbivory form large clumps in host trees (F; *Amyema maidenii* on *Acacia aneura*).

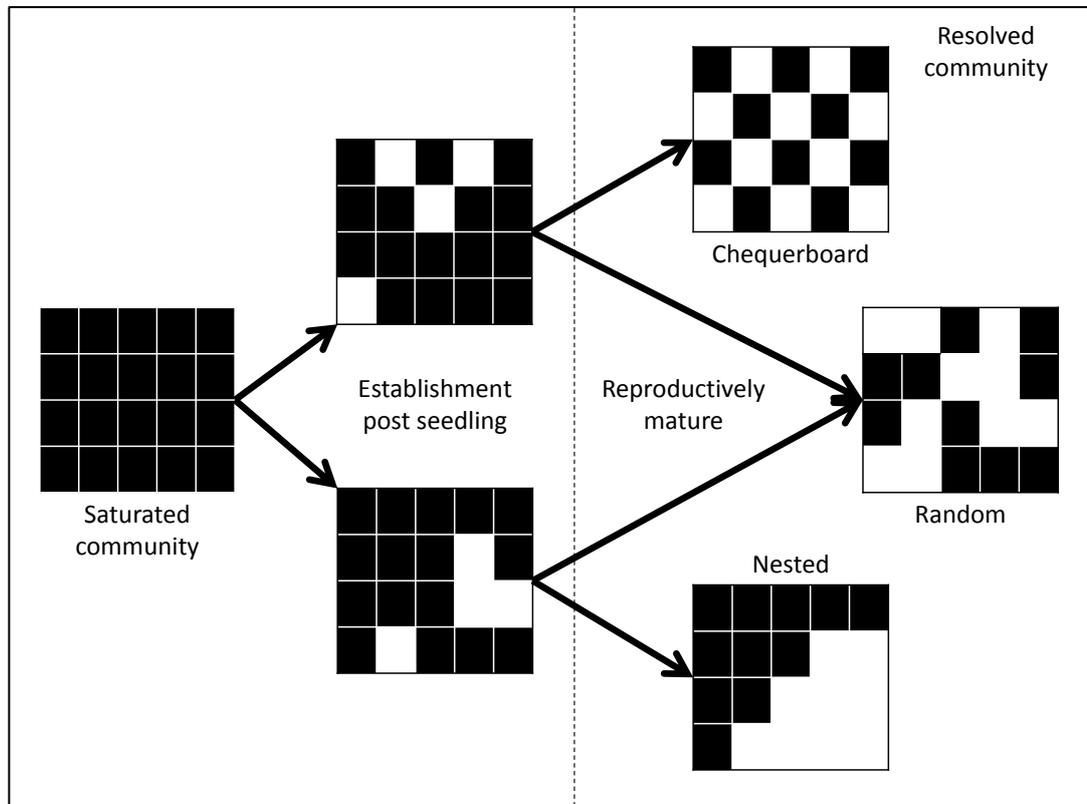


Figure 3 An experimental framework for testing the role of dispersal and host immunity in a mistletoe network. A saturated community is established through artificial dispersal (Filled cells), that are observed through time until all plants have died (unfilled cells) or become reproductively mature.

Summary statement

In this thesis I tested four different factors predicted to influence ecological networks, and I advanced the conceptual framework to investigate plant-plant interactions.

Overall, I showed that ecological networks are a valuable tool used to map species interactions, but require careful interpretation; including spatial scaling, sampling effort and distributional data. Prior to this thesis, network theory was applied to plant-plant networks with little consideration for biological traits, tree availability and species turnover. In turn, I showed that these factors influence network topology at the finest scale; an individual. These advances are fundamental to ensure that successful application of graph theory is applied to a wide range of communities, including aquatic and terrestrial, across continents and bioregions. Individual organisms (such as trees in the case of this thesis) that are generally ignored in network theory (because a connection needs to be made to be included) need to be considered during the interpretation of species interactions if network analysis is to reveal important mechanistic processes underlying the complex structure of very different ecosystems around the world. As development in the technology sector continues and advances are made in other fields of science, more complex machine learning approaches will be possible. Integrating artificial neural networks and feedback loops under the ideas of the Connectome project (mapping the human brain) will explain ecosystems in much greater detail. During these advances, it is essential that individual interactions, as I showed in this thesis, whether they are directly involved or considered a subsidiary part of an interaction network, are required to clearly explain structure.

References

- Bascompte, J. et al. 2003. The nested assembly of plant-animal mutualistic networks. - *Proceedings of the National Academy of Sciences USA* 312: 431-433.
- Barlow, B.A. & Weins, D. 1977. Host-parasite resemblance in Australian mistletoes: The case for cryptic mimicry. - *Evolution* 31:69-84.
- Canyon, D.V. & Hill, C.J. 1997. Mistletoe host resemblance: A study of herbivory, nitrogen and moisture in two Australian mistletoes and their host trees. - *Australian Journal of Ecology* 22: 395-403.
- Clements, F.E. 1936. Nature and structure of the climax. - *Journal of Ecology* 24: 253-284.
- Connor, E.F. & Simberloff, D.S. 1979. The assembly of species communities: chance or competition? - *Ecology* 60: 1132-1140.
- Diamond, J.M. & Gilpin, M.E. 1982. Examination of the "null" model of Connor and Simberloff for species co-occurrences on islands. - *Oecologia* 52: 64-74.
- Ehleringer, J.R. et al. 1986. Mistletoes: a hypothesis concerning morphological and chemical avoidance of herbivory. - *Oecologia* 70: 234-237.
- Gillarranz, L.J. et al. 2012. The architecture of weighted mutualistic networks. - *Oikos* 121: 1154-1162.
- Gotelli, N.J. 2000. Null model analysis of species co-occurrence patterns. - *Ecology* 81: 2606-2621.
- Gotelli N. J. & Ulrich W. 2012. Statistical challenges in null model analysis. - *Oikos* 121: 171-180.
- Harvey, P.H. et al. 1983. Null models in ecology. - *Annual Review of Ecology, Evolution, and Systematics* 14: 189-211.

- Ings, T.C. et al. 2009. Ecological networks – beyond food webs. - *Journal of Animal Ecology* 78: 253-269.
- Manning, A.D. et al. 2006. Scattered trees are keystone structures - Implications for conservation. - *Biological Conservation* 132: 311-321.
- Morales, J.M. & Vázquez, D.P. 2008. The effect of space in plant-animal mutualistic networks: insights from a simulation study. - *Oikos* 117: 1362-1370.
- Pekin, B.K. et al. 2012. Response of plant species and life form diversity to variable fire histories and biomass in the jarrah forest of south-west Australia. - *Austral Ecology* 37: 330-338.
- Peres-Neto, P.R. et al. 2001. Environmentally constrained null models: site suitability as occupancy criterion. - *Oikos* 93: 110-120.
- Pires, M.M. et al. 2011. The nested assembly of individual-resource networks. - *Journal of Animal Ecology* 80: 896-903.
- Roxburgh, S.H. & Matsuki, M. 1999. The statistical validation of null models used in spatial association analyses. - *Oikos* 85: 68-78.
- Roxburgh, S.H. & Chesson, P. 1998. A new method for detecting species associations with spatially autocorrelated data. - *Ecology* 79: 2180-2192.
- Rawsthorne, J. et al. 2011. Implications of movement patterns of a dietary generalist for mistletoe seed dispersal. - *Austral Ecology* 36: 650-655.
- Ramos-Jiliberto, R. et al. 2012. Topological plasticity increases robustness of mutualistic networks. - *Journal of Animal Ecology* 81: 896-904.
- Stagoll, K. et al. 2012. Large trees are keystone structures in urban parks. - *Conservation Letters* 5: 115-122.
- Strong, D. 1980. Null hypotheses in ecology. - *Synthese* 43: 271-285.
- Thébault, E. & Fontaine, C. 2010. Stability of Ecological Communities and the

- Architecture of Mutualistic and Trophic Networks. - *Science* 329: 853-856.
- Sweetapple, P.J. 2008. Spatial variation in impacts of brushtail possums on two Loranthaceous mistletoe species. - *New Zealand Journal of Ecology* 32: 177-185.
- Ulrich, W. 2004. Species co-occurrences and neutral models: reassessing J. M. Diamond's assembly rules. - *Oikos* 107: 603-609.
- Ulrich, W. & Gotelli, N. 2010. Null model analysis of species associations using abundance data. - *Ecology* 9: 3384-3397.
- Watson, D.M. 2009. Determinants of parasitic plant distribution: the role of host quality. - *Botany* 87: 16-21.