Genotypic variation in a foundation tree drives ecological network structure

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ABSTRACT

Biological evolution occurs in the context of complex networks of interacting species in which natural selection defines the structure of ecological networks. Fundamental to this evolutionary process is the discovery of a genetic basis to ecological network structure. Although previous work has demonstrated that tree genotype contributes to interaction network structure at the scale of forest stands, the contribution of tree genetics to localized interaction networks at the scale of individual trees has not yet been explored. To test the degree to which tree genetics can contribute to network structure across scales from trees to stands, we conducted quantitative modeling of interaction network for a community of epiphytic lichens in a long-term experimental common garden of genotyped trees of a foundation species (*Populus angustifolia*). We found three main results: 1) Tree genotype strongly contributed to network structure explaining over a third of the variation in lichen interaction networks, 2) Multiple aspects of interaction network structure varied in response to genotype, including network size, the number of interactions, linkage density and connectance, 3) At the stand scale, we also found signficant modular structure of plant-lichen networks resulting in part from the combination of trees of the same genotype tending to have similar community compositions and supporting similar lichen interaction networks dominated by positive interactions. These results support the hypothesis that variation in ecological interaction networks can result from genetically based variation in foundation species. Although these results are for a community of sessile organisms in close proximity to the tree, this study opens the possibility for a genetic basis to both direct and indirect interactions among species in complex communities.

Introduction

Evolution occurs in the context of complex networks of interacting species. In ecological communities, community dynamics depend on key interactions[1] that occur in species interaction networks, such as: trophic[2] and mutualistic[3] interaction networks. Phylogenetic patterns in ecological networks support the importance of evolutionary processes in shaping species interactions, community structure and ecosystem processes[?,9,15]. Community genetics studies[?] have shown that genetic variation in foundation species[4] plays a significant role in defining distinct communities of interacting organisms: such as, endophytes, pathogens, lichens, arthropods, and soil microbes. Multiple studies have now demonstrated that genetic variation influences numerous functional traits (e.g., phytochemical, phenological, morphological) produces a multivariate phenotype[5] tha contributes to variation in associated communities[6].

Additional work has provided support for the hypothesis that not only does composition vary among genetically distinct genotypes of foundation species but it also impacts the structure of the network of species interactions in these communities[?,?]. Also, work by Toju 2018, Toju2015, Toju2014 observed consistent patterns of centralized interactions of species modules focused around hubs of plant-fungal interactions. In other words, a small number of plant and fungal symbionts tended to have have disproportionate numbers of interactions with other species and likely are the drivers in determining community assembly, structure and dynamics.

Foundation species, by defention, have large effects on ecosystems and the networks of interacting species that make up the biotic component

[?,?,7] Ellison2005[?8]

Barbour2009

Ghering2017

Lamit2016

Hughes?

Leroy?

Busby2015

Crutsinger2016

Network structure is important for evolutionary dynamics

[?,?,9–11]

Bascompte2003

Rezende2007

Andreazzi2017

Toju2017

Dáttilo2016

Networks found to ge generally stable through time and space

34   Datillo2013

35   Díaz-Castelazo2010

36   Guimarães2007

37   Guimarães2006

38   **Genetic basis of networks**

39   Jormalainen2017

40   Fortuna et al. 2009

41   Solance2015

42   Lau2015

43   Lau2016

44   Keith2017

45   **None of the community genetics network papers have looked at species-species newtorks of associated organisms**

46   **at the scale of individual tree genotypes**

47   Here, we investigate how genetic variation in a foundation tree species determines the structure of a network of interactions

48   among a community of lichen species. Using a long-term (20 years+), common garden experiment with replicated individuals

49   of known genetic identity and a naturally established stand of *P. angustifolia*. We focused on a model community of 9

50   epiphytic lichens species, as previous research has demonstrated significant compositional responses of epiphytes to genotypic

51   variation[12, 13]. In addition, the life-history characteristics of lichen, having highly localized, direct contact interactions and

52   slow population turnover rates, allowed us to assess interactions among lichen species on individual trees. We hypothesize

53   that in natural systems evolution occurs in a community context involving interactions of complex networks of interacting

54   species[?, ?, ?, ?]. If correct, we should expect to find that network structure is genetically based in which different plant genotypes

55   support different interaction networks and that these interactions networks can function as indicators of ecological dynamics

56   important for conserving biodiversity. Applying a dual-scale (lichen-lichen and genotype-lichen interactions) network modeling

57   and analyses, we then examined the genetically based impacts of *P. angustifolia* on network structure.

Methods

58

Field observations in common garden and natural riparian forest stands

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60   The study was conducted along the Weber River, UT (USA), which is a cottonwood (*Populus* spp.) dominated riparian

61   ecosystem. Although two native species, *Populus angustifolia* (James) and *Populus fremontii* (S. Watson), occur here and are

62   known to hybridize, only pure or advanced generation backcrosses of *P. angustifolia* were sampled in order to avoid the effect

63   of the hybridization between these two species.

64   A common garden was used to isolate the effect of tree genotype from the effect of the localized microenvironment associated

65   with each individual and spatial autocorrelation. Asexually propagated clones of genotyped *P. angustifolia* individuals4 were

obtained from wild collections and planted randomly in a single field (0.025 km^2) at the Ogden Nature Center, Ogden, UT in 1992. A total of thirteen genotypes replicated between 3 and 8 times each, were chosen for sampling. Genotype names were previously published[?]. Observations were made in the common garden in October 2010 and May 2011.

The natural stand of *P. angustifolia* near the city of Uintah, UT (GPS: N41.13903, W110.94400) was used for the wild stand survey. We conducted sampling of the stand in May 2012. A total of 14 trees were chosen randomly over a 0.10 km^2 area with a minimal distance of 5.56 m between trees across a range of tree core based ages from 15 to 60 years.

Bark and Lichen Community Observations

On each tree, presence or absence of each lichen species was assessed in 50 total 1 cm^2 cells arrayed in a checkerboard pattern. Two adjacent 10 cm^2 quadrats centered at 50 cm and 85 cm from ground level were sampled. The checkerboard sampling pattern was chosen to isolate each cell based on an average thallus size of 1 cm^2. Samples were restricted to the northern aspect of the trunk to maximize the abundance of lichen and control for the effect of aspect. The thalli in each cell are expected to be spatially independent of the other cells in the quadrat, but exposed to similar micro-environmental conditions. Bark roughness was measured on each tree following[12].

The bark lichen community in this system is comprised of fourteen species; however, only 9 species were observed within our study quadrats. The lichen community included (abbreviations are given for species present in study): Xg = *Xanthomendoza galericulata*, Xm = *X. montana*, Ch = *Caloplaca holocarpa*, Cs = *Candelariella subdeflexa*, Rg = *Rinodina glauca*, Lh = *Lecanora hagenii*, Ls = *Lecanora* (unknown species), Pm = *Phyciella melanchra*, Pa = *Physcia adscendens*, Pu = *Physcia undulata*, *Phaeophyscia orbicularis*, *Phaeophyscia ciliata*, *Melanelia subolivacea*, *Meanelia elegantula*, including both crustose and foliose lichen species that exhibit low inter-annual variation[8]. We were able to rapidly assess lichen interactions by quantifying thalli in closed contact as assessed using 1 cm^2 cells. Species accumulation curves showed that communities in the wild and the common garden were thoroughly sampled and with similar species richness (Supplementary Materials, Fig 1).

Network modeling and analyses

We used the observations of lichen in the 1cm^2 cells on individual trees of *P. angustifolia* both in the common garden and the natural stand. Uni-partite networks were generated using an analytical procedure that removes non-significant interactions between species[14]. In brief, the approach begins by calculating the co-occurrences of all species pairs using the replicate cells. The signficance of each interaction pair is determined by comparing the observed frequency of co-occurrence to a theoretical "null" level of co-occurrence.

Data and Analytical Software Availability

All analyses were conducted using the R statistical programming language[?]. Code for the analyses is available on github and data are available at Figshare.

₉₆ ## Results

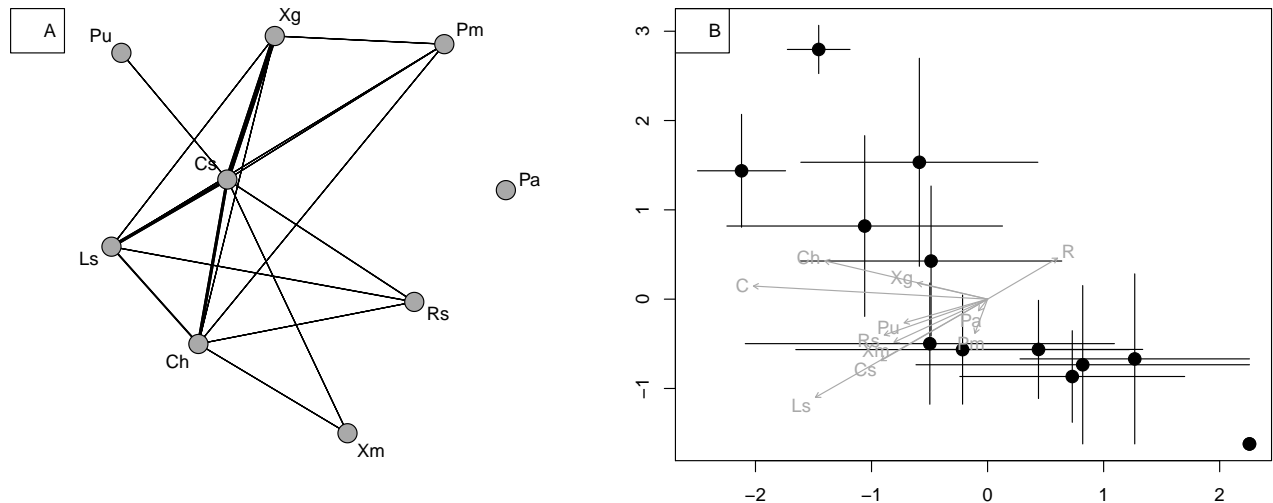₉₇ ### Tree Genotype Influences Ecological Network Structure

Figure 1. Significant lichen interaction network structure resulting from tree genotypic variation was observed in the common garden. A) A network diagram showing significant interactions averaged over all trees shown as edges connecting lichen species shown as vertices. B) Genotype centroids (points) of NMDS ordinated lichen networks ($\pm$ 1 S.E.). Arrows show the magnitude and direction of correlation of the ordinated networks with tree bark roughness (R), network connectance and lichen species abundances (Xg = *Xanthomendoza galericulata*, Xm = *X. montana*, Ch = *Caloplaca holocarpa*, Cs = *Candelariella subdeflexa*, Rs = *Rinodina* (unknown species), Ls = *Lecanora* (unknown species), Pm = *Phyciella melanchra*, Pa = *Physcia adscendens*, Pu = *Physcia undulata*).

	Df	SumsOfSqs	MeanSqs	F.Model	R2	Pr($>$F)
onc.geno	12.00000	163.74158	13.64513	1.87165	0.33795	0.04100
Residuals	44.00000	320.77902	7.29043		0.66205	
Total	56.00000	484.52060			1.00000	

Table 1. Pseudo-F Table for the perMANOVA test of genotype effect on lichen network similarity.

₉₈ ### Network Response to Tree Variation

₉₉ ### Genetic Structure Generates Forest Scale Network Structure

₁₀₀ ## Discussion

₁₀₁ Trait variation + assembly + ecosystem function

₁₀₂ These findings support the hypothesis that genotypic variation in a foundation species contributes to the structure of a

₁₀₃ network of interacting species that might be least expected to exhibit such structure.

₁₀₄ **TGW: MIGHT BE GOOD TO CITE PAPERS ON COMEPTITION IN LICHENS OR OTHER ORGANIZING**

₁₀₅ **FACTORS TO BACK UP THE LEAST EXPECTED STATEMENT. AS EPIPHYTES WE MIGHT NOT EXPECT**

₁₀₆ **THEM TO CARE.**

MKL: This is a job for Lamit and Rikke.

Several lines of evidence support this conclusion. First, the wild stand showed significant interaction network structure (Fig. 1a and b); and both tree genotype and the genetically based tree trait, bark roughness, was a strong predictor of co-occurrence patterns (Fig. 2a).

TGW: I THINK WE NEED TO EMPHASIZE THE LONG-TERM NATURE OF OUR COMMON GARDEN STUDY AS VERY FEW COMMON GARDEN STUDIES OF LICHENS LIKELY EXIST. ANY REFS ON THIS? IF TRUE MIGHT WANT TO MENTION THIS UP FRONT IN INTRO.

MKL: Same here. This is a job for Lamit and Rikke.

Second, in a long-term common garden study, network (Fig. 1b) structure showed a high degree of similarity to the wild stand network structure (Fig. 1c and d). Third, tree genotype was a significant predictor of SES values (Fig. 2a), displaying significant correlation with a genetically linked trait, bark roughness, both in the common garden (Fig. 2a) and in a naturally established stand of trees (Fig. 2b). Last, both of the bipartite genotype-species networks in the common garden and natural stand displayed significant modularity, suggesting that genotypic variation is leading to the formation of evolutionarily dynamic compartments within the community. Thus, just as numerous studies have shown that plant genotype can affect species richness, abundance, diversity, and composition and previous work has demonstrated that evolutionary processes shape ecological networks[10, 11], our study includes genetics in an empirical investigation that combines both experimental common garden findings along with studies in the wild that are in close agreement.

Our results point to the importance of understanding the community level effects of genetic variation and corroborate previous findings of the importance of plant genetics in shaping community structure and ecosystem processes[15]. This study highlights the potential for indirect effects of genetic variation to propagate through networks of interacting species and trophic levels. Altering the structure of interaction networks presents a means for genetic effects to be magnified within the system of interacting species. For example, Keith et al. (2017) showed that the genetics based interations of aphid resistant and aphid susceptible trees resulted in different interaction networks of their associated arthropod communities composed of 139 species. At the scale of ecosystems, trophic networks or food webs direct and control the rates of energy and nutrient flux[16]. Furthermore, in a predator-prey-plant study, Smith[17], showed that the interactions among species across trophic levels depended on plant genotype.

0.1 Units of evolutionary potential: Moving beyond species pairs

Tylianakis 2010 Conservation of species interaction networks.

- Functions-Metrics (Figure 2:

    – Richness/Connectance = Increased function and function stability

    – Nestedness = Buffer extinctions in mutualistic networks

- – Compartmentalization = greater stability, slow spread of disturbance (i.e. trophic cascades)

- – Proportion of Weak Links = stability, fewer cascades

- – Connectivity Distribution = Indicate Assembly, Robustness to 2nd extinctions

- Focus on metrics that saturate quickly with sampling

- Connectivity, Compartmentalization, Nestedness

- Need more research on the impacts of perturbations on these networks

- Networks working at multiple scales re-inforce connectivity

- Gene networks (Zink)

- Community networks (Keith2017, Lau 2016)

- Stand scale (This work)

- Landscape scale (Bothwell2017)

Although our study was conducted with a community of lichens, these results should be generalized to other groups of diverse organisms around the world that also exhibit significant genetic signals at the community level[7, 18], although spatial scale of interactions should be considered[19] Bangert et al. 2006. As heritable variation is the raw material for natural selection to act upon, a genetic basis for interaction network structure indicates evolutionary dynamics should be considered at the community level and that conserving genetic variation is important to consider in efforts to restore or preserve complex species interactions and their associated ecosystem functions[20]. With such findings, it appears that we are closer to understanding the evolutionary drivers of Darwin's entangled bank and the interconnectedness of species in complex communities.

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Acknowledgments

Author contributions statement

M.L. and L.L. conceived the study, M.L. and L.L. conducted the field work, R.N. assisted in lichen identifications, M.L. wrote the first draft of the manuscript, S.B. and T.W. contributed substantively to the conceptual development, T.W. established the common garden. All authors contributed to revisions of the manuscript.
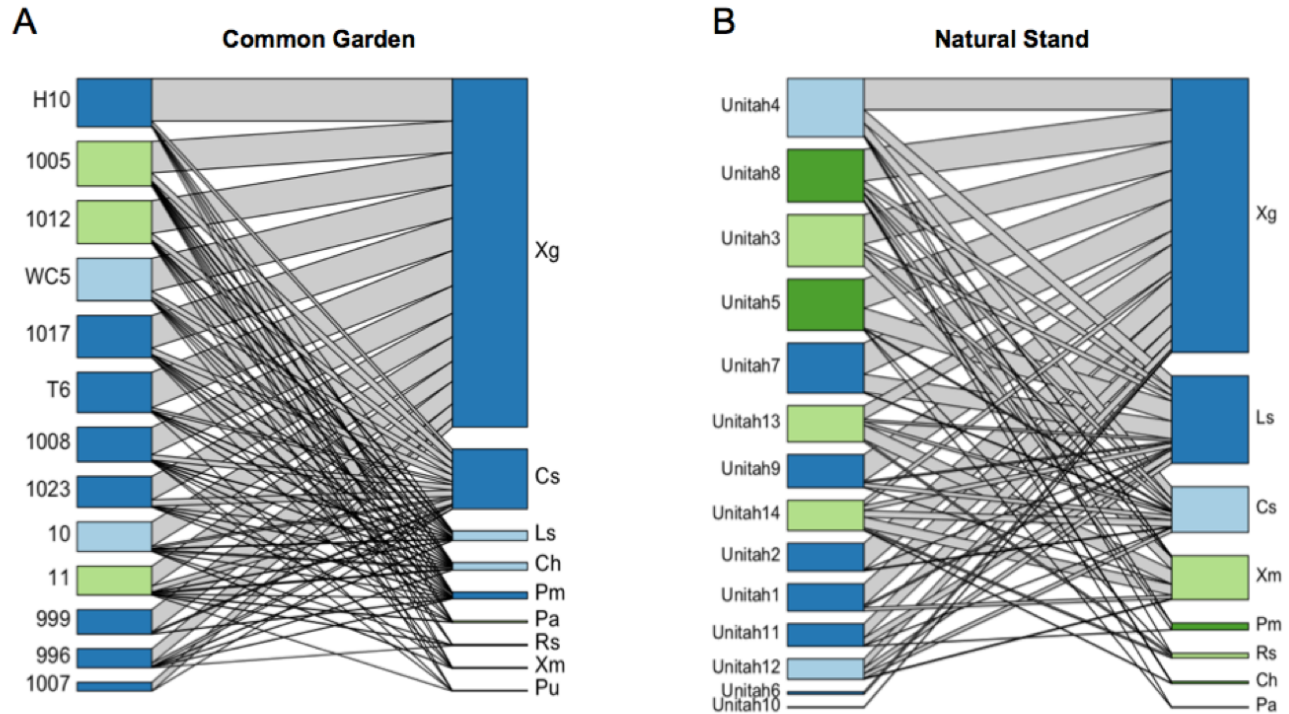
Additional information

Figure 2. Bipartite networks displayed significant modularity with modules comprised of both genotypes and species. The left most set of nodes shows tree genotypes (see Methods for genotype names) for the common garden (A) or individuals in the natural stand (B) connected to lichen species on the right. Both sets of nodes are scaled by their marginal totals (i.e., total observed individuals for tree nodes and total abundance for lichen species) and arranged by ascending totals from bottom to top. Node color shows the significant module membership for both trees and lichen species with module color having no direct relationship between the two networks, as modules were determined for each network independently.