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Research

Predicting the non-linear collapse of plant–frugivore networks due to habitat loss

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Habitat loss can trigger cascades of secondary extinctions, changing the organization of interacting assemblages. Until recently, most extinction models in interaction systems had limited ecological realism. Here, we estimate a realistic sequence of species extinctions resulting from habitat loss to assess its impacts on the structure of frugivory networks from the Brazilian Atlantic Forest. We show that realistic and random extinctions led to similar patterns. We also identified a threshold in the response of network structure to habitat loss. When forest cover was reduced to less than 40% of the landscape, network organization changed dramatically. Hence, the number of species being lost, rather than the order of species extinctions, is the key determinant of its impacts on the organization of frugivory networks. We highlight the need to conserve around 40% of forest cover to keep the basic organization of frugivory networks, a threshold already reached at the best-preserved Brazilian Atlantic Forest bioregion.

Keywords: extinction thresholds, frugivory networks, habitat loss, realistic extinctions

Introduction

The order in which species go extinct may affect how the structure of communities responds to biodiversity loss (Solan et al. 2004, Berg et al. 2015) and can trigger cascades of secondary extinctions throughout interacting assemblages (Colwell et al. 2012, Brodie et al. 2014). For this reason, it is crucial to estimate realistic sequences of species extinctions to unravel the potential consequences of current biodiversity loss on the organization of interacting assemblages (Srivastava 2002, Solan et al. 2004, Berg et al. 2015). Until recently, most species extinctions models in interaction systems had limited ecological realism. However, recent studies have used more realistic reasoning to model extinctions in mutualistic networks (Schleuning et al. 2016, Dalsgaard et al. 2018). Here, we add to this growing literature by estimating realistic

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extinctions to explore how coextinction cascades may emerge in a biodiversity hotspot due to one of the major threats to biodiversity, habitat loss (Pimm and Raven 2000, Sala et al. 2000, Henle et al. 2004a, Ewers and Didham 2006).

Habitat loss has important consequences on communities by changing distribution of populations, species richness but most importantly community composition (Fahrig 2003, Banks-Leite et al. 2012). This is because different species vary in the way they respond to habitat loss, with several studies now showing the existence of losers and winners in human-modified ladscapes (Laurance et al. 2006, Banks-Leite et al. 2012, Tabarelli et al. 2012). In the case of birds, a variety of traits, such as body size, habitat and diet specialization, migratory status and generation length, have been associated to extinction proneness (Newbold et al. 2013, Donoso et al. 2017). However, extinction risk is often a synergistic function of both intrinsic species traits and the nature of threat (Dirzo et al. 2014).

Simulations based on percolation theory have shown that as the area of original habitat declines in a given landscape, the pattern of fragmentation changes dramatically and there are rapid changes in the size and isolation of patches at critical proportions of habitat cover in the landscape (Andrén 1994, Gonzalez et al. 2011). If a species is sensitive to those structural aspects of the landscape, their responses to habitat proportion might also be expected to be nonlinear (Swift and Hannon 2010). Populations in fragmented landscapes may experience important reductions and below a certain amount of forest cover, non-linear responses to habitat loss emerge, leading to threshold responses as patches become smaller and more isolated (Andrén 1994, Pardini et al. 2010, Roque et al. 2018). In this context, the size of populations may decrease in forest remnants and they may be subject to more variation within and among patches, with possible local extinctions (Pardini et al. 2005, Roque et al. 2018). Such threshold responses at the population level may scale up and emerge at the organization of the interactions among different species in the community. Threshold responses are also triggered by cascading and synergistic effects of habitat change, leading to increasing levels of habitat degradation (Roque et al. 2018). Major factors affecting threshold values in the response of biodiversity to habitat loss may include matrix type and the particular metric used to describe diversity, as well as the particular organism subject of the study (Boesing et al. 2018, Roque et al. 2018).

Theoretical studies have shown that the structure of food webs changes according to the order in which species die out (Srinivasan et al. 2007, de Visser et al. 2011, Berg et al. 2015). Mutualistic assemblages too are impacted by sequential extinctions, showing different responses depending on the order in which species are lost (Memmott et al. 2004, Ramos-Jiliberto et al. 2012). For instance, habitat loss may disrupt mutualisms among plants and frugivores by leading seed dispersers to become rare or go extinct (Willson and Traveset 2000, Bascompte and Jordano 2007). The rupture in seed dispersal services, in turn, may have critical consequences to maintenance of plant populations, especially in tropical forests (Willson and Traveset 2000, Bascompte and Jordano 2007, Galetti et al. 2013).

In this study, our goal is to understand how extinctions triggered by habitat loss reorganize species-rich interacting assemblages of plants and frugivorous birds. We used as study system the Brazilian Atlantic Forest, one of the most diverse and threatened ecosystems in the world (Myers et al. 2000, Metzger 2009). To address this aim we analyzed plant–frugivore interactions from a pristine area and simulated a sequence of bird species extinctions following habitat loss, evaluating its impacts on the structure of the networks formed by plants and fruit-eating birds. To estimate this realistic sequence of species extinctions expected particularly from habitat loss, we used the minimum percentage of forest cover where each species was recorded, interpreted here as a direct assessment of species sensitivity to habitat loss, regardless of their particular traits. We explored if the simulated disassemble of the frugivory networks due to habitat loss differs from the expected when assuming random species deletions. Because some important frugivores are known to be habitat specialists that depend on forested areas to survive, we expect that forest loss would drive frugivory networks to collapse faster than expected by random extinction regimes. Finally, we extrapolated our results to assess the impacts of current forest cover on the structure of frugivory networks in the Serra do Mar bioregion of the Brazilian Atlantic Forest.

Material and methods

Overall approach

We started our approach by building plant–frugivore interaction networks from three sites in a large protected area, Parque Estadual Intervales (PEI), in the Atlantic Plateau of the State of São Paulo, southeastern Brazil (Fig. 1). These networks were our templates to assess the effects of habitat loss on the organization of the interactions. The idea was to simulate species extinctions resulting from habitat loss in these networks, aiming to evaluate the potential responses of a pristine system facing habitat loss. The next step was to estimate a realistic sequence of bird species extinctions following habitat loss. We used data from two studies on bird species composition of continuous and fragmented areas to estimate a sequence of bird extinctions. The third step was to simulate the sequence of extinctions in our pristine networks, evaluating the response of network parameters to habitat loss. Finally, we analyzed the current situation of the Atlantic Forest by extrapolating our results to Serra do Mar bioregion, the most preserved Atlantic Forest region.

Plant–frugivore interaction networks in a pristine area

Our three study sites are in mature forest, exhibiting a diverse avifauna typical of undisturbed areas (Vielliard and Silva 2001). The sites are in an altitudinal gradient, at 980 m
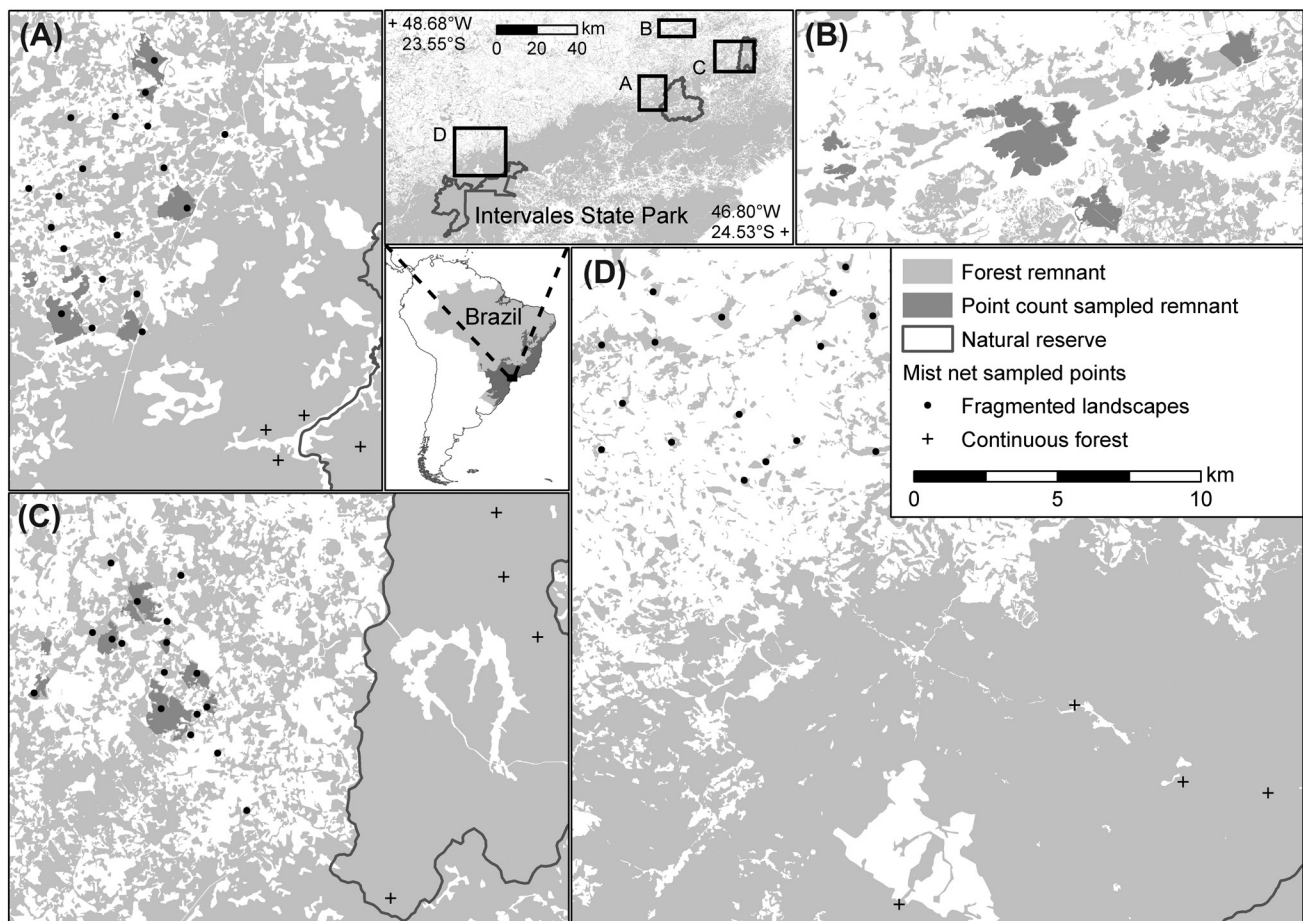
Figure 1. Location of the study sites in the Atlantic Plateau of the State of São Paulo, southeastern Brazil (central panel). Dark gray patches highlight the sites sampled with point counts, whereas dots and crosses represent sites sampled with mist nets. We built the interaction networks using data from Parque Estadual Intervales (highlighted in the upper panel). (A) Fragmented landscape in Tapiraí municipality and adjacent continuous forest. (B) Fragmented landscape in Ibiúna municipality. (C) Fragmented landscape in Caucaia do Alto municipality and adjacent continuous forest. (D) Fragmented landscape in Ribeirão Grande municipality and adjacent continuous forest.

('hilltop'), 847 m ('middle slope') and 597 m ('bottom of valley') above sea level. Between 1999 and 2002, we collected interaction data by walking along trails and recording a feeding bout (Levey et al. 2002) every time a bird was seen eating fruits (Hasui 2003). In each site, for one year, sampling was performed for two hours for five days per month, totaling 120 h of sampling effort per site (Hasui 2003). We also collected seeds in fecal samples from birds captured in mist nets. In each site, we placed 50 mist nets that were open for six hours, from dawn to early afternoon, one day per month along one year (3600 mist-net-hours per site). Combining the recorded feeding bouts and seeds from fecal samples, we built the interaction networks corresponding to each site. We described the structure of such networks by computing 1) bird and plant species richness; 2) their connectance, that is the proportion of all possible interactions that were actually observed; and 3) their degree of nestedness, a pattern of interaction in which less-connected species tend to interact with subsets of the species that interact with the more-connected species (Bascompte et al. 2003). Nestedness was measured

using NODF (Nestedness metric based on Overlap and Decreasing Fill, Almeida-Neto et al. 2008) and we evaluated if the observed degrees of nestedness were higher than expected by a null model that keeps the heterogeneity in the number of interactions (null model 2 in Bascompte et al. 2003). Nestedness in mutualistic networks is frequently associated with robustness to species extinctions and habitat loss (Memmott et al. 2004, Fortuna and Bascompte 2006). We also evaluated the modularity of our networks, a measure associated with persistence in mutualistic networks (Thébault and Fontaine 2010). Modularity is a pattern of interaction in which species are organized in modules, or groups, where they interact more with each other than with species from other modules (Olesen et al. 2007). However, none of our networks was significantly modular and we did not include this metric in our analyses (see Supplementary material Appendix B for a description of the methods we used to assess network modularity). We evaluated our sampling effort by estimating the sampling completeness, following Ramírez-Burbano et al. (2017). We worked with the quantitative versions of our

interaction networks and used the Chao 1 estimator of species richness to estimate the total number of interactions in the community, i.e. interaction richness (see Supplementary material Appendix B for further information). We calculated sampling completeness by dividing the observed by the estimated richness of interactions (Chacoff et al. 2012).

Sequence of bird species extinctions

The three networks show how interactions among plants and frugivorous birds are organized in pristine Atlantic Forest sites. We used the three networks as templates to assess the impacts of habitat loss over the structure of mutualistic assemblages. Frugivores are considered to be at more immediate risk of extinction than plants due to particular biological traits, including their higher trophic level and short lifespan. In contrast, plants have mechanisms that promote their persistence, such as seed dormancy, and underground vegetative reproduction (Willis and Bennett 1995). Therefore, we focused our extinction simulations on bird species, assuming that plant species would die out when loosing all of their mutualistic partners.

Aiming to identify a realistic sequence of bird species extinctions resulting from habitat loss, we combined data from two studies on bird species composition of continuous and fragmented areas in the Atlantic Plateau of the State of São Paulo, Brazil (Develey 2004, Banks-Leite et al. 2011). In one of the studies (Develey 2004), we used point counts to sample birds in two areas of continuous forest and in forest remnants in three fragmented landscapes (dark gray patches in Fig. 1). During spring and summer of years 2000, 2001/2002 and 2002/2003, in each fragmented landscape, we sampled eight fragments of different sizes (between 10 and 500 ha) and surrounded by different amounts of habitat. In each fragment, we set four fixed point counts inside a 20 ha plot located at a minimum distance of 100 m from the edge, and recorded the presence of birds, by listening or visually (Reynolds et al. 1980). In each continuous area, we delimited four 20 ha plots with four fixed sample points each. We visited all the sample points five times for 10 min, totaling 20 counts per plot (160 counts in each fragmented landscape and 160 counts in continuous forest landscapes). Point counts were made between 10 min before sunrise and three hours after sunrise. We considered each plot as a sample point, so we have 32 sample points, 24 of which were in forest fragments and eight were in continuous forest. In the other study, we used mist nets to sample birds in six landscapes from the Atlantic Plateau of the State of São Paulo, three of which were fragmented and three had continuous forest. We sampled birds in 53 forest patches (from 2 to 206 ha) in the fragmented landscapes and 12 sites in continuous forest (dots and crosses in Fig. 1). In each site, we sampled bird community using 10 mist-nets per site and sampling effort was of 637 net-hours on average (Banks-Leite et al. 2011).

From the lists of bird species composition in continuous and fragmented areas, we 1) assessed the minimum amount of habitat in which each bird species was recorded, and 2) estimated the sequence of bird extinctions, based on the minimum amount of habitat necessary for each bird species to occur. For the study collecting point counts data (Develey 2004), we used land use and cover mapping from the same period when data on bird species composition were collected to assess the amount of habitat at each sample point. Mappings were done based on aerial photographs at spatial resolution of 15 m (Develey 2004, Lira et al. 2012). As the exact geographical coordinates of the point counts sites were not available, we used the 'moving window' tool of Fragstat software ver. 3.3 (McGarigal et al. 2002) to calculate the percentage of forest cover within 800 m from each pixel of the image. We chose 800 m range because bird species respond to forest cover at this spatial scale (Banks-Leite et al. 2011). Then we used the average values of the pixels corresponding to each forest fragment, excluding the pixels located in the 100 m buffer from the edges, as a descriptor of the amount of habitat at each sample point. In the case of the study that collected mist net data (Banks-Leite et al. 2011), geographical coordinates of sampling points were available. This way, we used SPOT satellite images to calculate the percentage of forest cover in an 800 m radius around each sampling point. With these data, we calculated the minimum percentage of habitat around the points where each bird species was recorded (n = 53 species). When the same forest fragment was sampled by both studies, in a conservative approach we used the lowest estimate of habitat cover. For all the sample points from continuous areas, we considered 100% of habitat cover. On the basis of the lowest percentages of habitat in which each species was recorded, we established the expected order of extinction, such that species with larger minimum percentages were extinct before the species with smaller minimum percentages.

Fourteen bird species (ca 20%) comprised in the interaction networks from PEI were not recorded in the fragmented landscapes nor in the continuous areas sampled with point counts (Develey 2004) or mist nets (Banks-Leite et al. 2011). For these bird species, we used the IUCN threat categories (IUCN 2017) to assign their position in the sequence of extinctions. Species classified as least concern persisted in the networks all over the simulations of species extinctions, whereas endangered and near threatened species were put among the birds with the highest values of habitat requirements, as they are all threatened by habitat loss (IUCN 2017). One species (*Tangara* sp.) was still unrated because it was identified only at genus level. Thus, in a conservative approach, we included this bird among those that remained in the community until the end of the simulations.

To assess the robustness of our approach, we tested if the minimum percentage of habitat cover where each bird species occurred was related to independent assessments of bird extinction proneness, i.e. bird sensitivity to human disturbances and level of habitat specialization. In general, species restricted to areas with greater percentages of habitat cover tend to be more sensitive to human disturbances and also

more specialists in their habitat use (Supplementary material Appendix A).

Habitat loss simulations

We simulated increasing habitat loss (from 100% to 10% forest cover, 10% by 10%) in the three networks from PEI, seeking to assess the impact of habitat destruction on the organization of the interactions. We simulated bird extinctions according to their requirements of available habitat. For instance, species that were present in areas with at least 90% of forest cover went extinct when we reduced habitat cover to less than 90%. Plant species that lost all their seed dispersers were considered extinct, given the critical role seed dispersal plays for the persistence of plant populations (Cain et al. 2000, Howe and Miriti 2000, Jordano et al. 2010).

We assessed the effects of bird species extinctions on the interaction networks by computing, at each step of the simulation: the number of remaining species, the number of remaining interactions in the networks, network connectance, nestedness (NODF) and mean degree of bird and plant species. We performed piecewise linear regressions to evaluate potential thresholds in the response of network structure to species extinctions driven by habitat loss. We tested for a significant breakpoint by using Davies' test (Davies 2002). Piecewise regressions and Davies' tests were run using package 'segmented' (Muggeo 2008) in R (R Core Team). We also assessed the robustness of the frugivory networks by computing the area under the curve describing the percentage of plant species remaining in the network against primary bird extinctions (Burgos et al. 2007, Evans et al. 2013). Areas that are close to one represent networks that are robust to primary extinctions, since large percentages of extinctions are needed until significant secondary extinctions are observed. On the other hand, areas that are close to zero correspond to very fragile communities, in which extinction of even a small portion of species from one group leads to important extinctions in the group with which they interact (Burgos et al. 2007).

We developed three theoretical benchmarks to compare against the extinction regime based on habitat loss. In the first scenario, bird species were randomly deleted from the networks (1000 replicates), representing the effects of nonselective extinction events. We randomized the different species for a fixed number of species extinctions resulting from habitat loss. In the second scenario, we simulated extinctions from the most to the least connected bird species. In the third scenario, we simulated extinctions from the least to the most connected bird species. Least to most-connected and most to least-connected scenarios represent endpoints in the continuum describing how threatening an extinction sequence can be. This way, seeking to assess the resistance of the studied networks to habitat loss in relation to a critical scenario, we calculated the difference between robustness to extinctions from most to least connected species and robustness to extinctions due to habitat loss. The greater the

difference between scenarios, the higher is the resistance of networks to habitat loss provided by its nonrandom patterns of interaction. The effects of the simulated species extinction may differ because species play different roles in structuring networks. By exploring the distribution of contribution to network structure (Vidal et al. 2014) among birds, we also assessed whether particular structural roles were lost at different stages of the habitat loss process (Supplementary material Appendix B).

Current forest cover in the Atlantic Forest

In order to analyze the current situation of the Atlantic Forest, we extrapolated our results to Serra do Mar bioregion, the most preserved Atlantic Forest region (Ribeiro et al. 2009), where our study area belong to. First, we used the 2016 forest cover map from MapBiomas (Projeto MapBiomas 2018) to quantify the percentage of forest cover around each pixel from the Atlantic Forest. Then, we used our piecewise regression models to predict network metrics values that correspond to actual percentages of forest cover found in the Serra do Mar bioregion (see details in Supplementary material Appendix C). We considered the estimated breakpoints and evaluated the percentages of the Serra do Mar bioregion that are below such breakpoints.

Data deposition

Data available from the Dryad Digital Repository: <https://doi.org/10.5061/dryad.b47n414> (Vidal et al. 2019).

Results

Plant–frugivore interaction networks in a pristine area

The structure of the three frugivory networks was similar (Supplementary material Appendix D Table D1). All networks comprised slightly more plant than bird species, showed low connectance (0.07) and were significantly nested (p < 0.01), with similar values of NODF. Sampling completeness was of 56.6% in hilltop, 47.4% in middle slope and 59.1% in bottom of valley network. Besides their similar structure, the general pattern of structural response to bird extinctions were consistent among the three networks (Supplementary material Appendix E Fig. E1–E3). Therefore, we decided to combine the information from the three networks in one single network, hereafter called 'combined network'. To generate the combined network we pooled all the species and interactions recorded in the three networks together in a single network. We believe this would make our message clearer and straightforward, since the responses of each network resembled each other (Supplementary material Appendix E Fig. E1–E3) and the combined network represented the general patterns. The combined network comprised 67 bird species and 127 plant species, showed low connectance (0.06) and was also significantly nested (NODF = 17.93, p < 0.01,

Supplementary material Appendix D Table D1). Sampling completeness of the combined network was of 57.7%. The initial structural patterns regarding the number of species and interactions, connectance, nestedness and mean degree of bird and plant species were benchmarks for us to assess the impacts of sequential bird species extinctions on the organization of the network.

Sequence of bird species extinctions

By ordering the bird species according to their minimum requirements of habitat area (Supplementary material Appendix D Table D2), we identified, in each frugivory network, the birds that would go extinct in scenarios of increasing habitat loss. Twenty bird species from the combined network occurred in low-forested areas, with less than 10% of habitat cover, persisting in the network until the end of habitat loss simulations (Supplementary material Appendix D Table D2). Large frugivorous birds, such as cotingas (Cotingidae), trogons (Trogonidae) and toucans (Ramphastidae), were absent from those low-forested areas, where habitat generalists, omnivorous birds that occasionally feed on fruits prevailed. On the other hand, twenty-one bird species from the combined network occurred in areas with at least 60% of habitat cover, including those large-bodied frugivores, which are important seed dispersers.

Among birds of each frugivory network, we found considerable variation in their contribution to network structure and this variation occurs across the range of sensitivity to the loss of habitat cover (Supplementary material Appendix B Fig. B1). For example, birds restricted to areas with high percentages of habitat cover (more than 40%), comprise both species with high and low values of contribution to network structure. In each site, median values of contribution to network structure were also similar among birds from different categories of percentage of habitat cover (Supplementary material Appendix B Fig. B1).

Habitat loss simulations

In our approach, the simulated reductions in habitat cover led to sequential bird species extinctions according to their minimum requirements of habitat amount. Bird extinctions, in turn, led to secondary extinctions of the plants that lost all their seed dispersers. Therefore, increasing habitat destruction led to increasing species loss in the networks, affecting their structure. For all network descriptors, we found a qualitative change in network structure from all the studied sites when habitat was reduced to around 40% or 30% of its original cover. For example, the realistic sequence of species extinctions produced a threshold in the number of remaining species in the combined network, with a significant breakpoint at 39% of habitat cover (Fig. 2). Similar thresholds in the number of remaining species were found when the three interaction networks were analyzed separately (Supplementary material Appendix E Fig. E1–E3). Accordingly, all the networks showed a decrease in the number of interactions, as

interacting birds and plants died out. We identified a threshold in the number of interactions at 41% of habitat cover in our combined network (Fig. 2). Connectance increased in the networks, with a threshold at 32% of habitat cover in the combined network. Similarly, network nestedness tended to increase with habitat loss (Fig. 2), with a threshold at 31% of habitat cover in the combined network. Mean degree of bird species increased with habitat loss, with a breakpoint at 29% of habitat cover, whereas mean degree of plants tended to decrease, with a threshold at 42% of forest cover. In general, structural consequences of the expected sequence of extinctions did not differ from the results obtained with random extinctions (Fig. 2). Moreover, random species deletions also led to threshold responses, with significant breakpoints very similar to the ones found when species were removed according to their habitat requirements. The exception was the mean degree of bird species, which increased in the realistic model and was kept nearly constant in the random model (Supplementary material Appendix D Table D3). Thus, habitat loss likely leads to a pattern similar to the expected by random extinctions, but even so the network structure shows a sudden qualitative change in the patterns of interaction.

Robustness of our combined network to the realistic sequence of extinctions considerably departs from the critical scenario of deletions from most to least connected bird species (difference between areas under curves = 0.32, Fig. 3). The three networks, consistently with the combined network, are relatively robust to the sequence of bird extinctions when compared to the critical scenario (Supplementary material Appendix E Fig. E4). Moreover, for all the interaction networks, the curves generated by the realistic sequence of bird extinctions are between the 2.5% and 97.5% quantiles of the areas under curves produced by the random sequences of extinctions (Fig. 3, Supplementary material Appendix E Fig. E4). Therefore, the realistic and random extinctions are not so drastic to system's robustness as extinctions targeted at the most connected species.

Current forest cover in the Atlantic Forest

Between 35% and 45% of the Serra do Mar bioregion is below the forest cover thresholds we identified for frugivory networks (Fig. 4). Currently, 43% of the Serra do Mar bioregion is below the threshold of forest cover for the number of species (Supplementary material Appendix C Fig. C1). Similarly, 44% of the Serra do Mar bioregion is below the threshold we identified for number of interactions (Supplementary material Appendix C Fig. C2). With respect to connectance and nestedness, 37% and 36% of the Serra do Mar bioregion, respectively, are below the thresholds we identified for such metrics (Supplementary material Appendix C Fig. C3, C4).

Discussion

Simulated bird species extinctions expected from habitat loss led to substantial changes in the organization of the
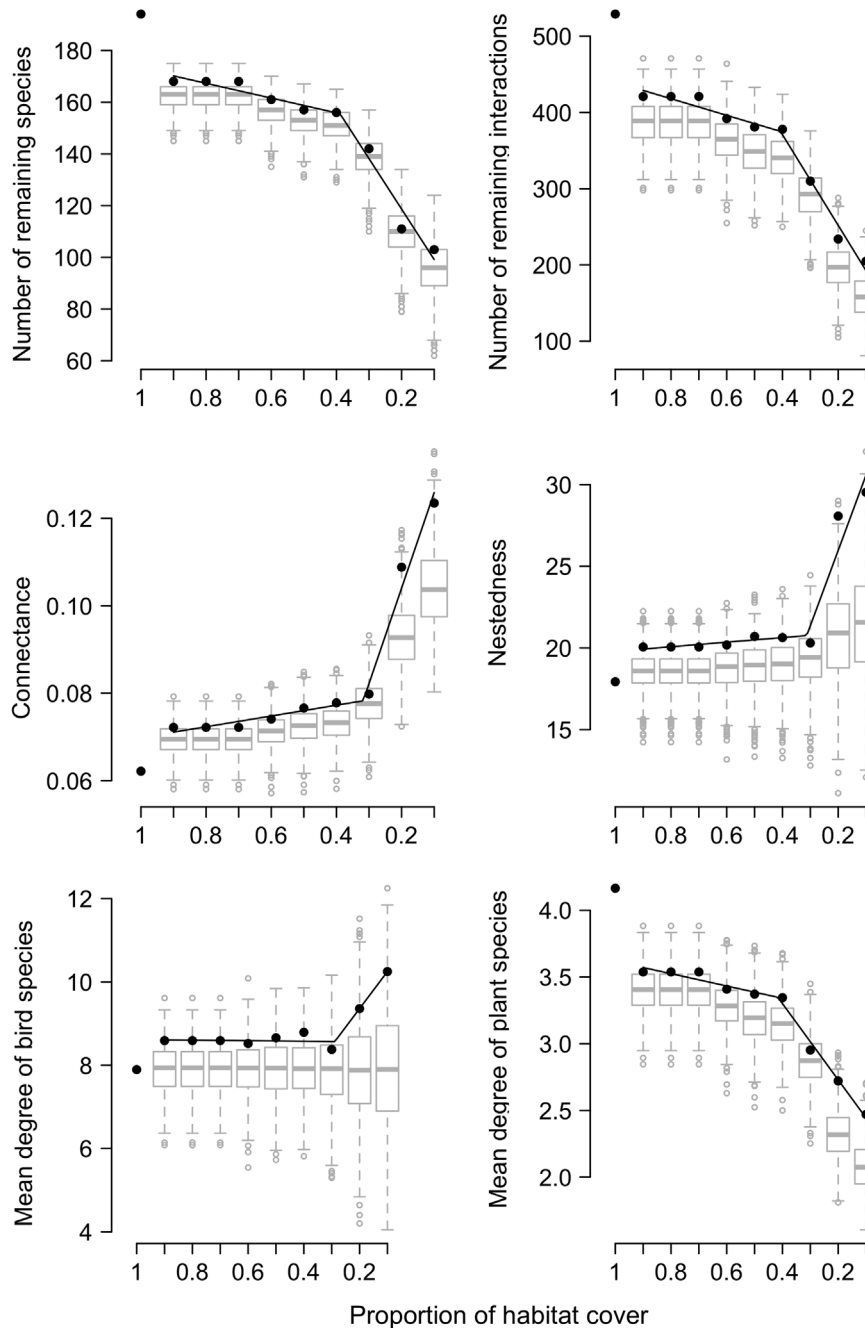
Figure 2. Effects of habitat loss on the structure of the combined network. Black dots depict structural values of the networks as birds, and secondarily plants, were lost in simulations of habitat destruction. Gray boxplots represent results from random deletions of bird species. Solid black lines are the fitted linear models with breakpoints, representing the threshold in the response of network parameters to habitat loss.

plant–frugivore networks. Bird species losses resulted in secondary plant extinctions, increasing network connectance and nestedness. In our approach, unconnected species are always removed, increasing connectance. Besides that, most bird and plant species have few interactions (Supplementary material Appendix F Fig. F1), in relation to the number of possible interactions they could have, increasing connectance when they are removed from the system (Soares et al. 2017).

So that, when one species with few interactions is lost, the number of interactions in the network keeps nearly the same, whereas the number of possible interactions is reduced, increasing network connectance. The combined network, in agreement with the three frugivory networks, originally shows low connectance, and low mean degree, for both bird and plant species. Therefore, species with few interactions prevail and network connectance increases when these
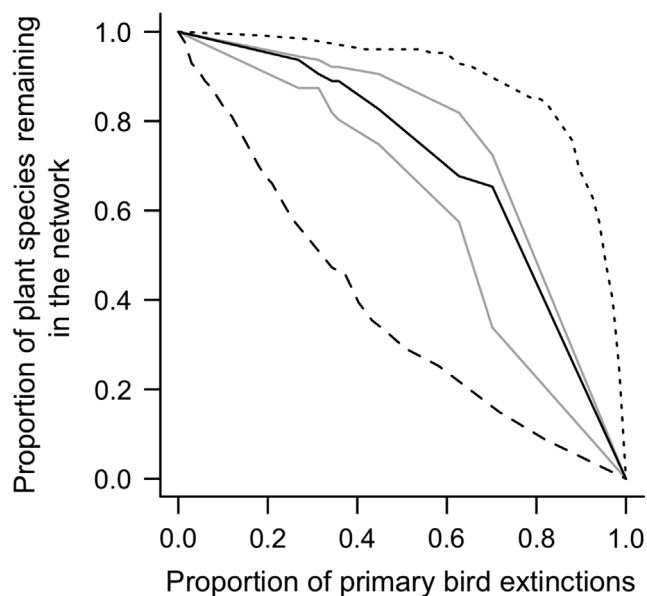
species are lost from the system. Similarly, an empirical study of plant–pollinator systems showed that habitat loss, by reducing species richness, also led to increased connectance (Spiesman and Inouye 2013). In contrast to our results, however, the same study found no effect of habitat loss on nestedness (Spiesman and Inouye 2013). We found that nestedness tended to increase with habitat loss, suggesting a concentration of interactions by the most connected species (Soares et al. 2017).

Connectance and nestedness both promote the persistence of populations in mutualistic systems (Lever et al. 2014), but see (Allesina and Tang 2012). Therefore, these metrics may indicate greater resilience of the system to species loss (Soares et al. 2017). As we observed greater connectance and nestedness when habitat was reduced to less than 30% of its original cover, we may infer that simplified frugivory systems, with fewer species and interactions, are more resilient to further extinctions than richer systems (Soares et al. 2017).

In general, the consequences to levels of robustness of the realistic sequence of extinctions and to network organization did not differ from those observed when bird species were removed randomly from the system. Therefore, the specific order in which birds are lost following habitat loss seems to play a minor role in determining the structure of remaining networks during the process of habitat destruction. The number of primary extinctions, regardless the order in which they occur, however, seems to be critical in determining the impacts over the organization of the frugivory networks.

Figure 3. Robustness of the combined network. Solid black line depicts the tolerance curve for the realistic sequence of bird extinctions following habitat loss. Gray lines are the 2.5% and 97.5% quantiles of the areas under curves produced by the random sequences of extinctions. Dashed black line corresponds to the tolerance curve for the removal of the most-linked to least-linked bird species. Dotted black lines correspond to the tolerance curve for the removal of the least-linked to most-linked bird species.
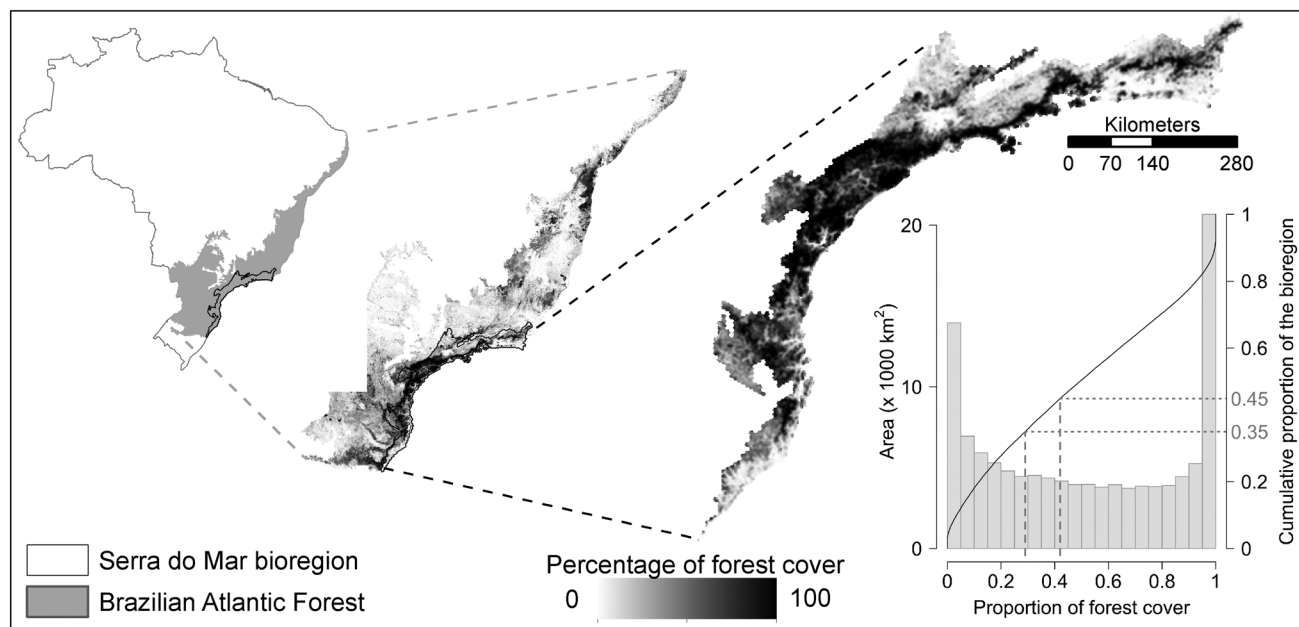

Figure 4. Brazilian Atlantic Forest biome (dark gray), highlighting the Serra do Mar bioregion and current percentages of forest cover. Inner graph comprises the histogram of the area (bars) and cumulative proportion of the Serra do Mar bioregion (solid line) within classes of proportion of forest cover. We highlight the largest and the smallest forest cover values we identified as breakpoints (dashed gray lines): 0.29 for mean degree of bird species and 0.42 for mean degree of plant species, and corresponding proportions of the Serra do Mar bioregion that are below such breakpoints (0.35 and 0.45, respectively). See Supplementary material Appendix C for details.

Indeed, connectance and nestedness seems to be much more dependent on the number and abundance of species than on the identity of species in a network (Spiesman and Inouye 2013). In this direction, random primary removals, just as the realistic sequence of extinctions, had major impacts on community organization, producing similar thresholds responses to habitat destruction. Thus, the number of species expected to be lost in scenarios of increasing habitat loss could be used to estimate the impacts of such extinctions on the organization of plant–frugivore communities. The correlation between species richness and the network metrics we used may affect the threshold values we found in the response of network structure to habitat loss. However, the reduction of species richness resulting from habitat loss is not the only responsible for the observed patterns. The identity of the lost species and their contribution to network structure are key factors determining the threshold response. A possible explanation for the similarity between simulated extinctions based on habitat loss and random extinctions is the fact that birds with different structural roles were observed in each category of percentage of forest cover. This way, structurally important bird species, i.e. those with many interactions, both within and among modules, and high contribution to network nestedness (Vidal et al. 2014), were recorded in sites with high percentages of forest cover and also in less forested areas. This finding suggests that species vulnerability to habitat loss is not related to species importance to network structure, so that we may loose important components of frugivory networks at different moments of the process of habitat destruction.

Changes in the structure of the networks are relatively mild until enough habitats is lost to cause the collapse of the system, which occurs suddenly. Between 40% and 30% of habitat cover, we can observe an abrupt change in the number of species and interactions, connectance, nestedness of the networks and mean degree of bird and plant species, suggesting a threshold of habitat cover for structural patterns of frugivory networks. In our simulations, when habitat cover shrinks to 40%, large numbers of bird species are lost as they need more habitat to occur, and this leads to a massive extinction of birds and interacting plants. Indeed, we found significant breakpoints (around 40–30% of habitat cover) in the relationship between network structure and habitat amount in the landscape. Theory predicts that a disproportionate loss of species occurs when habitat cover in the landscape decreases to approximately 10–30% (Andrén 1994), and empirical data from the same sites indicate a threshold for the integrity of vertebrate communities (birds, mammals and amphibians) around 30% of habitat cover (Banks-Leite et al. 2014). At this point, the community integrity, i.e. the similarity in community composition between fragmented and continuously-forested landscapes, is drastically reduced, as large proportions of forest-dependent species are lost while habitat generalists enter the system (Banks-Leite et al. 2014). Such changes in bird species composition may lead to the collapse of seed dispersal networks when habitat cover is reduced to 40% or 30% of the landscape. Whether a species will perish or persist in altered landscapes is often determined by their traits (Henle et al. 2004b), with endemics being more likely to be losers, while species with broad geographical ranges are more likely to be winners (Banks-Leite et al. 2014). However, little is known about how this replacement of losers by winners impacts interaction networks.

Our result is consistent with a recent review on threshold responses of birds to habitat loss, which found a mean habitat cover threshold at 29.5% in tropical landscapes (Melo et al. 2018). At low habitat levels, fragmentation effects compound those of habitat loss, such that the rate of change in the ecological response is greater than expected from habitat loss alone (Swift and Hannon 2010). Below 30% of habitat cover, there are non-linear changes in population size that result from patches becoming smaller and more isolated (Andrén 1994, Pardini et al. 2010, Roque et al. 2018). Shrinking populations may weaken mutualistic interactions, as the frequency of interactions is reduced, what can cause species extinctions until the collapse of the whole community (Dakos and Bascompte 2014). In this context, our findings suggest that those threshold responses of population sizes emerge at the network level, implying subtle changes in the organization of frugivory interactions.

Indeed, specialized frugivores, such as toucans, *Euphonia* spp., most trogons and cotingas (Snow 1981, Jordano 2000) were missing from the frugivory networks when habitat cover was reduced to 30% of its original cover. In these networks, tanagers, thrushes and tyrant-flycatchers prevail. Many of these species show limited effectiveness as seed dispersers, as they are habitat and diet generalists, eating fruits only occasionally. These results reinforce the need to maintain forest cover over 30%, preferably 40%, not only to maintain the integrity of species composition (Banks-Leite et al. 2014), but also to secure the structural patterns of frugivory networks. It is worth noting that threshold response in species richness represents the end point of the extinction process, i.e. the corollary of multiple single-species extinction events (Radford et al. 2005). Therefore, for conservation purposes, target level of habitat cover should be well above the threshold level. Currently, around 40% of the Serra do Mar bioregion is below the threshold values we estimated for network metrics. Serra do Mar bioregion is the best-preserved biogeographical sub-region of the Atlantic Forest, holding 36.5% of its original vegetation (Ribeiro et al. 2009). Therefore, even in the best-preserved region of the Atlantic Forest, a significant area may be under important changes in the organization of their frugivory systems. Future studies could evaluate the structure of frugivory networks in other Atlantic Forest bioregions, investigating how to avoid or revert changes in the organization of the interactions due to habitat loss.

Although our model of species deletions is not dynamic, we found relative robustness to realistic species loss, indicating that the observed structure of frugivory networks promote the system stability in landscapes facing habitat loss. Our topological approach also does not consider possible secondary extinctions due to changes in species

abundances, what may underestimate the number of co-extinctions (Berg et al. 2015). Hence, approaches that consider population dynamics may bring new insights on the effects of habitat loss on mutualistic systems. We used the minimum percentage of forest cover where each species was recorded as a direct assessment of species sensitivity to habitat loss. However, it would be interesting to investigate other sequences of extinctions built as functions of different variables that have been associated with extinction proneness, such as body size (Donoso et al. 2017). Regarding particular species traits, another issue to be better explored in future studies is the phylogenetic relationship among species that go extinct (Rezende et al. 2007). Coextinction of phylogenetic related species may cause the loss of entire functional groups, since closely related species share a number of traits affecting particular functions of organisms, such as seed dispersal. With respect to our measure of habitat amount, we used the percentage of forest cover in a defined area, an important measure that is widely used in studies that investigate threshold responses to land cover change (Melo et al. 2018). Although we recognize that spatial configuration and species traits may interact and affect thresholds (Villard and Metzger 2014), habitat cover plays a critical role in defining biodiversity persistence (Fahrig 2013, Banks-Leite et al. 2014, Melo et al. 2018).

Our results indicate relative robustness of the frugivory networks to a realistic sequence of bird extinctions following habitat loss. The order in which species are lost seems to play a minor role in defining the ultimate consequences to the organization of interacting assemblages. This way, the number of species expected to go extinct following habitat loss, rather than their specific order of extinction, may suffice to infer about the structural impacts over frugivory systems. However, the structure of the networks seems to change abruptly when habitat cover goes below 40% to 30%, revealing a threshold response. Future studies should explore the generality of these results to other mutualistic networks. In our study system, the alterations habitat loss caused to interaction organization are not so drastic as it would be if the most important species for network structure were lost first. Fortunately, this result suggests that we are not in the worst-case scenario. Although there are opportunities to avoid further extinctions in areas that were recently subject to deforestation, we already reached critical conditions in great part of the Atlantic Forest.

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Supplementary material (available online as Appendix ecog-04403 at <www.ecography.org/appendix/ecog-04403>). Appendix A–F.