Title: Trophic complexity alters the diversity-multifunctionality relationship in experimental grassland mesocosms

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Abstract

Higher levels of diversity within trophic levels are necessary to sustain multiple ecosystem functions, but this diversity-multifunctionality relationship peaks at intermediate percent-function thresholds. The presence of multiple trophic levels, or trophic complexity, affects ecosystem multifunctionality but its effect on the diversity-multifunctionality relationship has not been experimentally tested. To test the sensitivity of the diversity-multifunctionality relationship to trophic complexity, we simultaneously manipulated plant diversity and trophic complexity in a multifactorial tall-grass prairie mesocosm experiment at Cedar Creek, Minnesota, USA. Trophic complexity altered the diversity-multifunctionality relationship, by lowering the height of the peak as well as by shifting to a negative effect at lower thresholds. These outcomes are consistent with decreases in both the “jack-of-all-trades” effect and complementarity among plant species. Our findings suggest that trophic complexity, which is experiencing widespread declines on a global scale, is important for sustaining ecosystem multifunctionality in the face of equally widespread declines in biodiversity.

Introduction

Increasing biodiversity within a trophic level, both along experimental and naturally occurring gradients, frequently show positive relationship with single ecosystem functions[1–4]. When this diversity-function effect is tabulated across multiple functions, depending on the percent-function threshold selected, higher levels of biodiversity are needed to sustain multiple functions than what is needed to sustain any single function[4,5]. Empirical support for this diversity-multifunctionality relationship, across taxa and habitats, suggests that biodiversity loss can have larger consequences on ecosystem functioning than previously assumed based on single-function studies[2,6,7].

This diversity-multifunction relationship has been shown to be sensitive to two key factors; (1) the percent-function threshold selected[7–9] and (2) biodiversity within trophic levels[4,7]. Selecting low thresholds (e.g., obtaining at least 10% of every function measured) generally yields positive diversity-multifunction effects, the strength of which increases with increasing value of the selected threshold. This pattern of a rise in the positive association between diversity and multifunctionality is referred to as the “jack-of-all-trades” effect[8] (Fig 1). This jack-of-all-trades effect occurs because species vary in their contribution to different functions and increasingly diverse communities will contain species that are masters of some functions but not of others. This has been explained through physiological trade-offs that limit the potential of any species to maximise all functions simultaneously. However, as one increases the threshold selected (e.g., obtaining over 80% of every function measured), the relationship switches to a negative relationship. The negative association between diversity and multifunctionality above moderate thresholds occurs because the likelihood that any species will provide moderate to high levels of functioning across all functions becomes increasingly low; a pattern termed “the-master-of-none” effect. In general, diverse communities sustain most functions at moderate thresholds because of

the jack-of-all-trades effect, but above moderate thresholds, diverse communities cannot sustain multiple functions at high levels because of the master-of-none effect.

Most diversity-function studies have focussed on a single trophic level, most often the plant or producer community, but changes in diversity of non-producer trophic levels can also have significant impacts on several ecosystem functions[10–16]. The number of trophic levels, or trophic complexity, can thus potentially impact the diversity-multifunctionality relationship, but this effect is poorly understood. In grassland communities, for example, the presence of multiple trophic levels was shown to decrease the effect of biodiversity on a single ecosystem function over a long term experiment[17], while an observational study showed the opposite effect[18]. Current studies, however, do not allow for a direct comparison of ecosystems that vary in trophic complexity because such explorations would require simultaneously manipulating plant diversity as well as the number of non-producer trophic groups.

Here, we explore by a direct manipulative experiment, the hypothesis that trophic complexity affects the diversity-multifunctionality relationship through impacts on the jack-of-all-trades effect (i.e., change in the value of the threshold at which there is a switch from a positive to a negative diversity-multifunctionality relationship) *across* functions and complementarity and selection effects *within* functions. Quantitatively, these effects can be measured as the slope of the per-species gain or loss in functions above a given threshold, which we define as the diversity-multifunctionality effect (the DME). We hypothesised two possible effects of changes in trophic complexity: (1) a change in the magnitude (or height) of the peak in DME along the threshold axis attributable to the jack-of-all-trades effect and (2) a change in the location of the peak in DME along the threshold axis attributable to changes in complementarity or selection. Figure 1 provides a conceptual framework for these predicted

outcomes. We note that these predicted responses to trophic complexity are not mutually exclusive; the peak height, location, both, or neither may respond to differences in trophic complexity.

To test this framework, we simultaneously manipulated tall-grass prairie plant diversity and trophic complexity in 94 tall-grass prairie mesocosms at Cedar Creek, Minnesota, USA. In these mesocosms, we varied plant diversity from 1 to 16 species, following a standard, stratified $\log_2$ randomised design. We simultaneously varied trophic complexity following a factorial design resulting in 4 trophic treatments; above-ground insect-dominated mesofaunal communities only (INS), below-ground litter mesofaunal communities only (LIT), both above- and below-ground mesofauna (BOTH) or all mesofauna excluded (NONE). For comparison, we pooled all the data (POOLED).

We measured four ecosystem functions; aboveground biomass, belowground biomass, soil water retention and biomass recovery after harvest. We analysed the average values of these functions across the different plant diversity and trophic complexity treatments. We calculated a combined multifunctionality metric for each community across a range of thresholds following standard approach for thresholds[4,8]. We then analysed the diversity-multifunctionality effect (the DME) as the slope of the linear fit of the number of functions maintained above the threshold against plant diversity. Finally, we tested whether this relationship was sensitive to trophic complexity.

Results

For every function measured, communities on average had low percent threshold values; most communities failed to maintain functions at high values (Fig 2). The average values of each function remained within a small range of values across different plant diversity treatments (Fig S1). Trophic treatment did not alter the curves substantially for any single function, although there were some

differences at high threshold values for water retention and biomass recovery (Fig 2) and at high diversity treatments for water retention and aboveground biomass (Fig S1).

Ecosystem multifunctionality showed the commonly observed pattern to changes in plant biodiversity (Fig. 3). The plant diversity-multifunctionality relationship for the pooled dataset was positive for a threshold of 25% (slope=0.03, p=0.36) and 50% (slope=0.007, p=0.81) but was negative for 75% (slope=-0.14, p=0.4) and 90% thresholds (slope=-0.11, p=0.36) (Fig 3). Biodiversity had a weak positive effect on multifunctionality at low thresholds while at high thresholds, the effect was negative. At the 90% threshold, most plots had no functions above cutoff, making a biodiversity or trophic effect less discernible, while at 25% most plots had a large number of functions above the threshold even without the effect of plant diversity or trophic structure. This suggests that the biodiversity effect is most observable at moderate thresholds. Similarly, the effect of trophic complexity was significant only at 75% (-0.12, $p<0.05$) and marginally significant at 90% (-0.07, p=0.06) while at a 25% threshold (-0.05, p=0.59) and 50% threshold (-0.14, p=0.16), these differences were not detectable.

The diversity-multifunctionality effect (the DME) is sensitive to trophic complexity. In the pooled dataset, where all treatments were taken together, the DME increased and peaked at moderate thresholds (Fig 4). Trophic complexity had an effect on both the height and location of the peak DME (Fig 4). Comparing the curves of the four different treatments, we found that the addition of either the above-ground or litter trophic level led to higher peak DMEs than the plant-only or full-complexity (i.e., plants, litter, and above-ground fauna) communities, but all treatments peaked at similar intermediate thresholds (~ 35- 40%). The transition from positive to negative DMEs occurred between ~ 40 – 60% thresholds for all trophic treatments. Finally, the full trophic complexity treatment was the most distinct of the four treatments, having the lowest peak (at ~10% threshold) with DME values that were consistently lower in magnitude and largely negative across most threshold values.

Discussion

In agreement with recent literature[7,8] , our results show that biodiversity is necessary for maintaining multiple functions from low to moderate thresholds in grassland communities (i.e., the jack-of-all-trades effect), but at higher thresholds, increases in diversity are inversely associated with multifunctionality (i.e., the master-of-none effect) (Figs 3-4). Our study's findings are therefore consistent with what has been observed across different taxa and habitats[7].

We advance our understanding of biodiversity and multifunctionality, however, by also showing that this jack-of-all-trades and master-of-none pattern is sensitive to trophic complexity. Although the bottom-up effect of plant diversity on herbivore diversity and potentially ecosystem functions has been observed in long-term experimental data[17,19–21], our study tests the top-down (aboveground fauna) and donor-controlled (litter fauna) effects of trophic complexity on multifunctionality. The simultaneous presence of aboveground and litter fauna trophic levels, the treatment closest to natural systems where trophic complexity is common, showed a very low-threshold switch to negative diversity-multifunctionality effects (DMEs) and persistent negative DMEs throughout most thresholds values (Fig. 4). Any reduction in trophic complexity altered the diversity-multifunctionality relationship. Thus, both the location and height of peak DMEs are affected by trophic complexity, as suggested in our conceptual framework (Fig. 1).

The nature and direction of the trophic impact on the DME curve depends on the trophic component considered. When examined across thresholds, both aboveground and litter fauna amplified the biodiversity effect at low thresholds but at high thresholds, only aboveground fauna amplified the effect. This suggests that the presence of aboveground arthropods increases the contribution of biodiversity in providing ecosystem multifunctionality at moderate thresholds. This could be because

plant diversity is known to decrease associational effects to herbivory damage[22, 23] leading to an additional advantage of diversity in the presence of aboveground fauna.

In contrast to what we observed for multifunctionality, our analyses did not reveal significant impacts of trophic complexity on any single ecosystem function (Figs. 2-3). Further we did not observe any impacts of the trophic complexity treatments on average ecosystem function value at any level of plant diversity (Fig S1). Although the treatment with plants alone performed better at water retention along all thresholds (Fig. 2) and intermediate diversities (Fig S1), this observation alone cannot explain the differences between the treatments. Our results also show that the presence of multiple trophic components decreases the magnitude of the diversity-multifuntionality effect across all thresholds, consistent with results from a long-term experiment from the same region demonstrating that non-producer trophic components obscure the diversity-productivity relationship due to a loss in complementarity effects with the addition of herbivores.[17] As the plants we used were taxonomically and functionally quite distinct, identity effects[24], or unique contributions of individual species to the ecosystem functions considered, are also likely to be responsible for the nature of the biodiversity-multifunctionality relationship we observed.

Our findings have important implications for our understanding of the relationship between biodiversity and ecosystem functions. Plant diversity is currently understood to be critical to sustaining multifunctionality at or below moderate thresholds, but our results show that such effects are influenced by trophic complexity. Given that declines in trophic complexity are widespread, occurring, for example, where agriculture leads to habitat simplification through the use of biocides and where trophic downgrading is occurring in terrestrial and marine habitats, most ecosystems are experiencing declines in both producer diversity and trophic complexity. Sustaining a broad spectrum of ecosystem

functions and the services they provide will require more plant or producer diversity in the face of widespread trends in trophic simplification.

Methods

Experimental Methods

We used data from a year long experiment on grassland mesocosms in a tall-grass prairie, part of the Cedar Creek Ecosystem Science Reserve, Minnesota. The experimental design was factorial with 100 pots, 1m in diameter, grown inside inside netted insect exclosures. Each pot was maintained at one of 5 levels of plant diversity - 1, 2, 4, 8 or 16 species. The species used in this experiment (Table S1) were native perennial species used in other experimental studies from the site[17,25,26]. Pots with data discrepancies were excluded from this analysis, resulting in a sample size of 94. Crossed with the plant diversity treatments were trophic complexity treatments – plants + aboveground mesofauna (primarily insects and other invertebrates, INS), plants + litter mesofauna (LIT), plants + both aboveground and litter mesofauna (BOTH), or plants only (i.e., no aboveground or litter fauna, NONE). These treatments were achieved by first applying a pesticide treatment on all the pots and removing all aboveground and litter fauna. 25% of the pots in each treatment were allowed to be recolonised by aboveground fauna only, 25% by liter fauna only, 25% by both aboveground and litter fauna, and 25% were plants only, leading to the four community complexity treatments. The experiment was run for a year from July 2000 to July 2001 when final biomass and soil water retention were measured and biomass recovered in each pot after harvest was measured a year later.

Ecosystem function measurements

Four candidate ecosystem functions were analysed: (i) Aboveground biomass - the total aboveground biomass in each pot was measured in 2001, after one year of experiment. (ii) Belowground biomass - the total belowground biomass in each pot was measured after one year of experiment (iii) Water

retention - the time taken for a fixed volume of water to flow into a collection flask at the bottom of the pot was measured at the end of one year of experiment (iv) Biomass recovery - the biomass recovered one year post harvest was measured to quantify biotic control over ecosystem resilience. Recovery was calculated as the ratio of the total recovered biomass to the biomass prior to disturbance (aboveground + belowground biomass). A principal component analysis of functions to test correlations resulted in water retention close to PC1 and aboveground biomass close to PC2 and the vector of biomass post harvest was in a similar direction as the biomass in the previous year.

For each function, the decay curves for each trophic treatment was plotted along a standardised range of threshold values. These were visually examined for differences.

Multifunctionality

To assess multifunctionality across diversity treatments, measurements of four ecosystem functions – aboveground biomass, belowground biomass, water retention and biomass recovery after harvest - were chosen and analysed using published methodology of the threshold approach[18]. To control for effects of outliers in the dataset, the maximum value of function in each case was set as the mean of the five highest values. The slope of linear models fit to the number of functions maintained above threshold to the species diversity in the community was defined as the biodiversity effect on multifunctionality. Biodiversity effect on multifunctionality was analysed for the pooled dataset as well as the dataset split into the four community complexity treatments. The magnitude of the peak and the point at which the curve of biodiversity effect vs. threshold crosses the x-axis for each of the treatments were examined in comparison with the pooled data for reasons described in the introduction.

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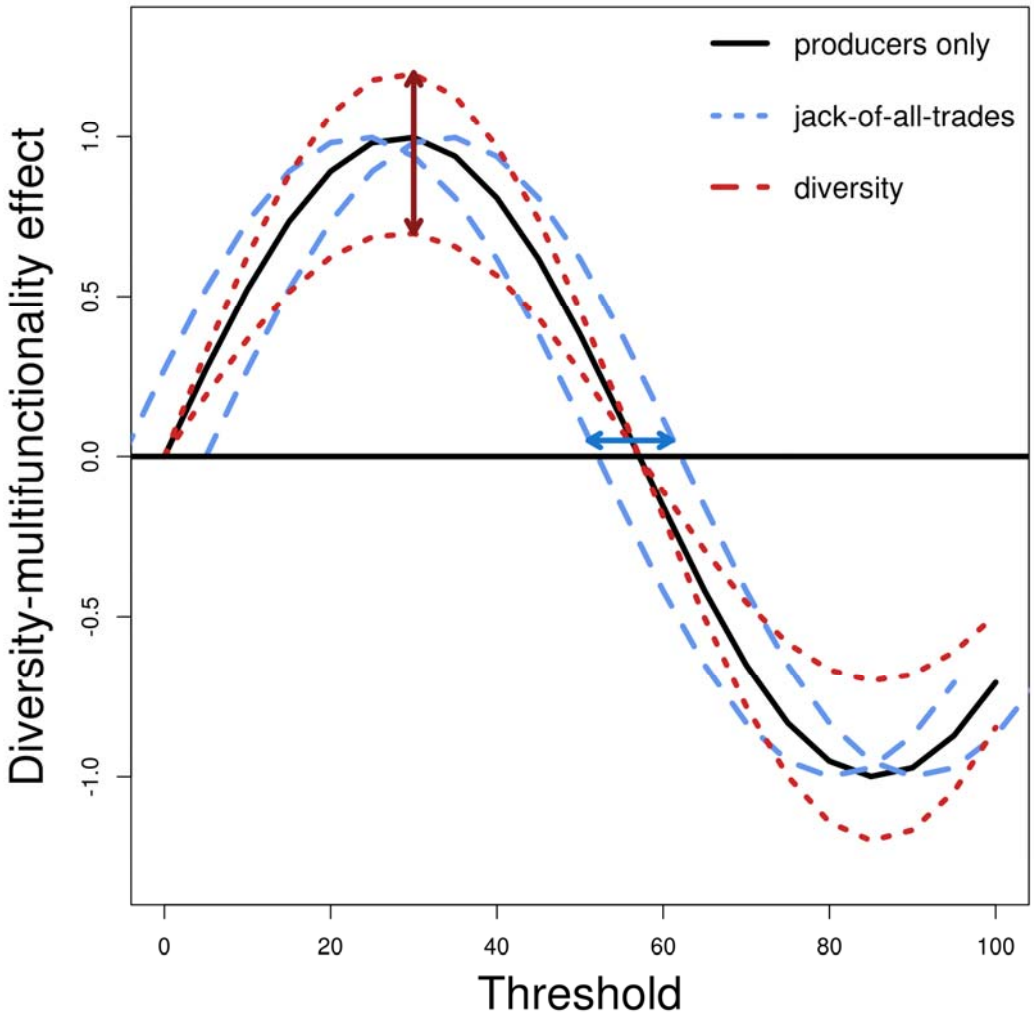
producers only
jack-of-all-trades
diversity
Diversity-multifunctionality effect
1.0
0.5
0.0
-0.5
-1.0
0
20
40
60
80
100
Threshold

Figure 1. Conceptual framework illustrating how changes in trophic complexity can alter the height and location of the peak in the diversity-multifunctionality effect (DME) curve. The DME is a measure of the slope of the relationship between diversity and multifunctionality (e.g., number of functions gained through the addition of species) whose value is dependent on the threshold (i.e., percent ecosystem function obtained) used in estimating multifunctionality. Thus, all parts of the DME curve above the zero line have positive slopes for the number of functions above a given threshold that are gained by an increase in diversity. The continuous black curve represents a hypothetical DME for a plant community in the absence of trophic complexity (i.e., the plant only curve). The coloured lines represent possible deviations from the plant-only curve with the addition of trophic complexity. The blue curves represent trophic-induced changes attributable to the “jack-of-all-trades” effect which shift the location of the peak in the DME curve along the threshold axis. The red curves represent trophic-induced changes attributable to the complementarity effect which shift the height of the DME curve.

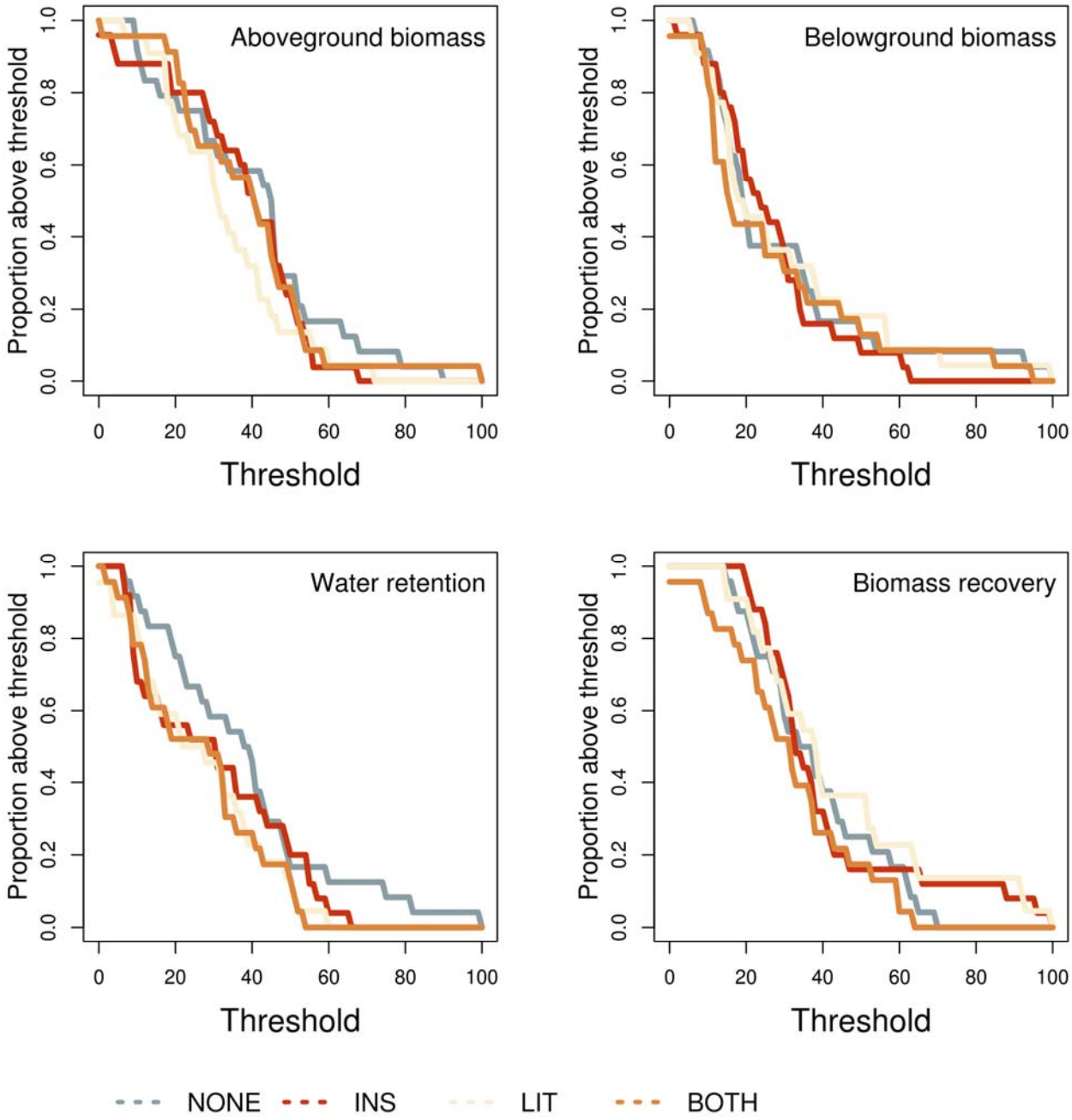

Figure 2: Proportion of communities maintaining single ecosystem functions along a range of percent thresholds. Panels represent the four functions – aboveground biomass, flow time, biomass recovery and belowground biomass. Coloured curves represent the four trophic treatments.

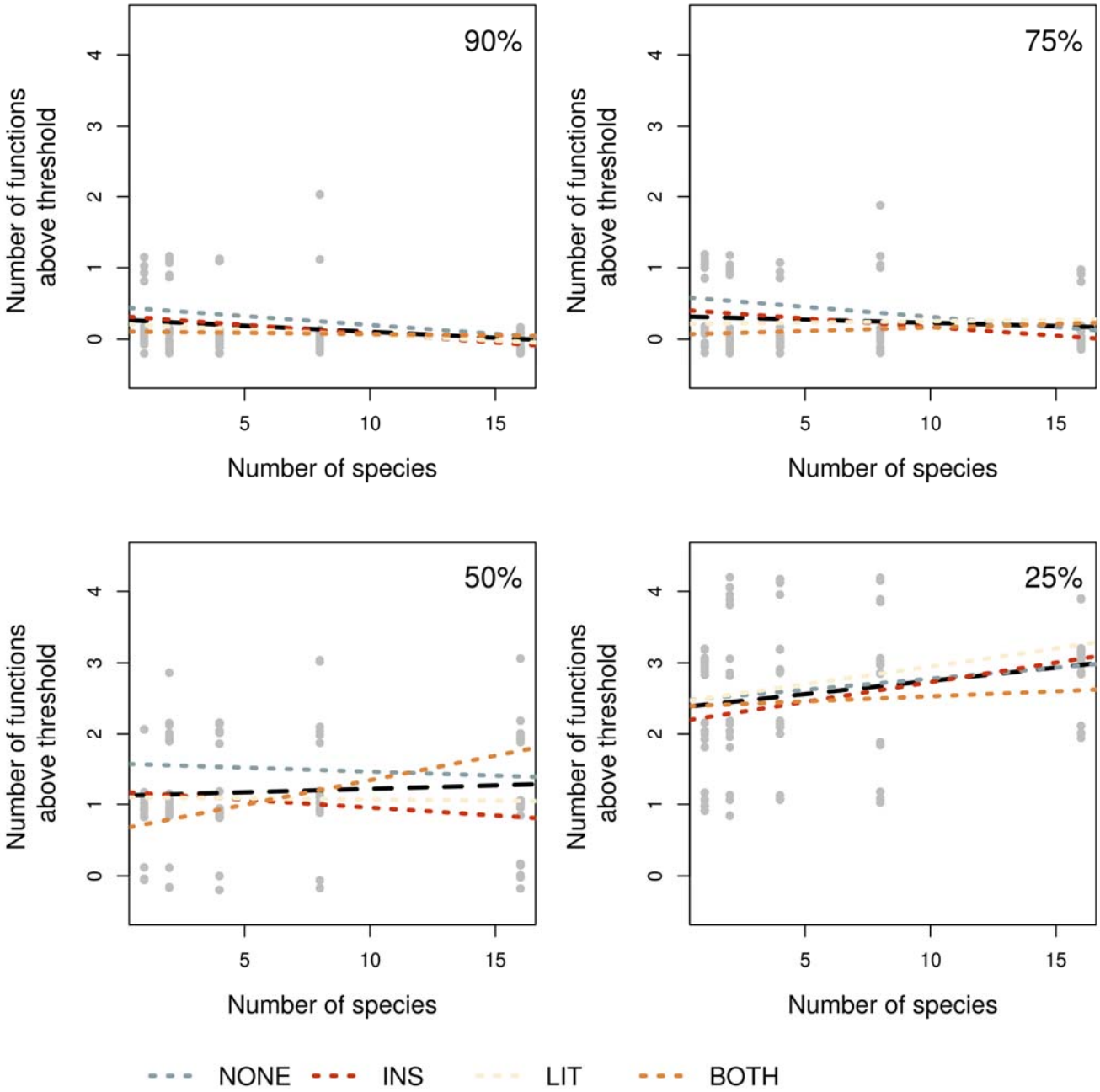

Figure 3. Number of functions above four different thresholds, indicated in the top right corner of each panel, against the number of species in the plot. Lines represent linear model fits for pooled data (black) as well as each treatment (colours). Legend shows colour codes for treatments. Actual data points for each plot represented as grey dots.

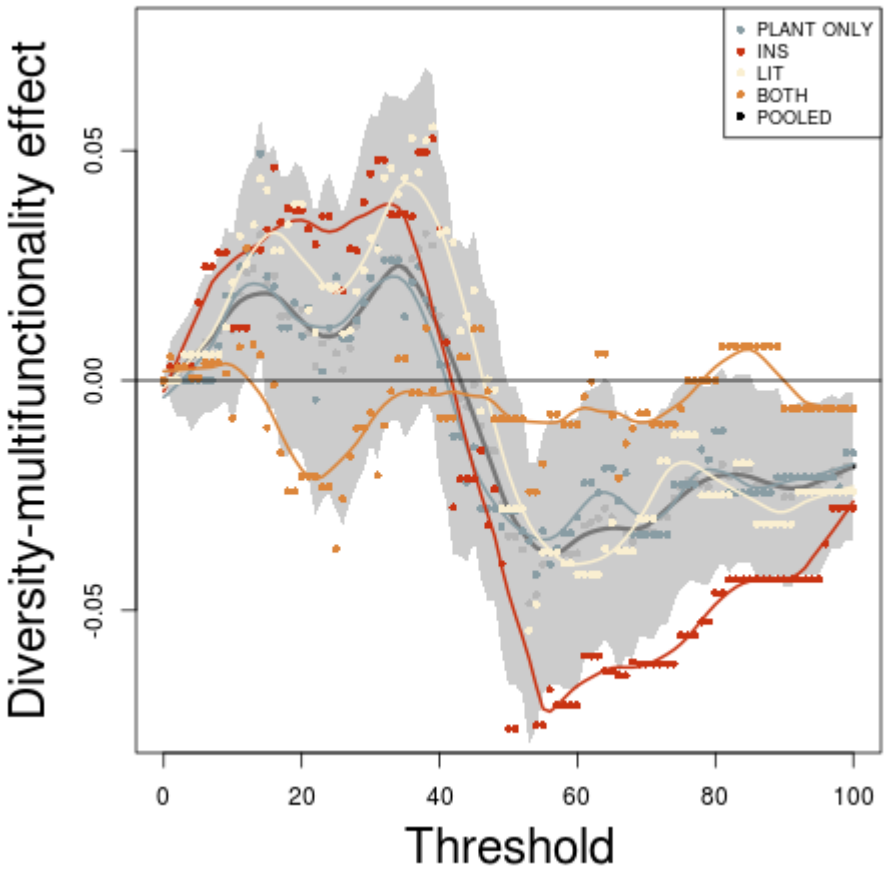

Figure 4. Effect of threshold on the biodiversity-multifunctionality effect (the DME). Each point represents the slope of the linear model between plant species richness and number of functions above the threshold. Each DME curve for each level of trophic complexity is plotted using a different colour, as presented in the key (top right), with the curve for the pooled dataset presented in black. The curves are smooth-spline interpolations. The grey polygon represents the bounds of the standard error of the slope in the pooled dataset.