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# Elevated $CO_2$ and N Gradually Weaken the Influence of Diversity on Ecosystem Stability

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## ABSTRACT

Biodiversity promotes ecosystem productivity and stability, positive impacts that often strengthen over time. But ongoing global changes such as rising atmospheric carbon dioxide ($CO_2$) levels and anthropogenic nitrogen (N) deposition may modulate the impact of biodiversity on ecosystem productivity and stability over time. Using a quarter-century grassland biodiversity-global change experiment we show that diversity increasingly enhanced productivity over time irrespective of global change treatments. In contrast, the positive influence of diversity on ecosystem stability strengthened over time under ambient conditions but weakened to varying degrees under global change treatments, largely driven by a greater reduction in species asynchrony under global changes. Thus, over 25 years, $CO_2$ and N enrichment gradually eroded some of the positive effects of biodiversity on ecosystem stability. As elevated $CO_2$, N eutrophication, and biodiversity loss increasingly co-occur in grasslands globally, our results raise concerns about their potential joint detrimental effects on long-term grassland stability.

## 1 | Introduction

Biodiversity loss, rising carbon dioxide ($CO_2$) and nitrogen (N) deposition are ongoing drivers of change. They each alone influence ecosystem stability (Hautier et al. 2014; Hector et al. 2010; Liu et al. 2019; McNaughton 1977; Tilman et al. 2006; Xu et al. 2021, 2022; Zhang et al. 2016), often measured as inter-annual consistency in primary production, relevant for managing both natural and agricultural systems. Ecosystem stability is often promoted by increasing biodiversity (Craven et al. 2018; Hector et al. 2010; Isbell et al. 2015; Tilman et al. 2006; Tilman and Downing 1994; Xu et al. 2021), but the positive effect of diversity on ecosystem stability has been observed to change (albeit inconsistently, i.e., to be stronger or weaker) under global changes that enrich resources such as nitrogen (Hautier et al. 2014; Suonan et al. 2023; Zhang et al. 2016). A strengthening over time of the effect of diversity on ecosystem stability has also been observed, at least under ambient conditions (Wagg et al. 2022), but whether and how global changes alter the impact of diversity on ecosystem stability over a decade or more has not yet been considered. Given the rising levels of $CO_2$ and N deposition globally, and concomitant loss of biodiversity, it is crucial to understand the long-term implications of these global changes on ecosystem services.

The influence of diversity on ecosystem stability may be stronger (i.e., more positive) or weaker (i.e., less positive or even negative) under global change compared to ambient conditions. Ecosystem stability, often quantified as the ratio of mean productivity to its interannual variation, can increase when plant diversity (hereafter diversity) supports higher plant productivity and/or when diversity reduces interannual variation in productivity because of asynchronous and offsetting interannual variation in productivity of different species (i.e., species asynchrony) (Hector et al. 2010; Xu et al. 2021). Global changes that enrich resources may increase the positive effects of biodiversity on ecosystem stability, if resource enrichment elevates productivity relative to its temporal standard deviation at higher diversity (Reich et al. 2001). Alternatively, ecosystem stability may be negatively impacted in diverse communities if global changes such as N-addition cause species loss (Harpole et al. 2016; Harpole and Tilman 2007; Hautier et al. 2009; Reich 2009), thus reducing returns of productivity from fertilisation (Isbell et al. 2013) and/or reducing asynchrony amongst species (Hautier et al. 2014, 2020; Song et al. 2023) and ecosystem stability (Davidson et al. 2025; Hautier et al. 2015). Elevated $CO_2$ may similarly lead to shifts in composition (Reich et al. 2024) that could decrease species asynchrony and can also interact with N enrichment to influence the impact of diversity on ecosystem stability. A recent meta-analysis of relatively short-term studies (Su et al. 2022) suggests that N enrichment has a net negative impact whereas elevated $CO_2$ has no impact on ecosystem stability, but how biodiversity may modify these effects remains to be studied. In theory, biodiversity can also decrease ecosystem stability by decreasing population stability (i.e., the weighted mean of species population variability) (Thibaut and Connolly 2013; Tilman et al. 2006), but evidence for influence of global changes on the relationship between diversity and ecosystem stability via this process is scarce. Lastly, it is possible that global changes do not modify the influence of diversity on ecosystem productivity and stability but instead increase or decrease productivity and its temporal standard deviation (and thereby ecosystem stability) proportionally across all diversity levels (Craven et al. 2016).

The positive impact of diversity on ecosystem stability may also strengthen or weaken over time, even under ambient conditions, but the development of theory applicable to temporal trends in diversity-ecosystem function remains sparse (see (Amyntas et al. 2023) for diversity-productivity trends over time). Evidence from a long-term grassland biodiversity experiment indicates that the impact of diversity on ecosystem stability strengthens over time, at least under ambient conditions (Wagg et al. 2022). This positive temporal trend was attributed to (i) the strengthening impact of diversity on productivity over time (e.g., through compounding of positive feedbacks), a pattern that has growing support from other grassland and forest biodiversity experiments around the world (Guerrero-Ramírez et al. 2017; Reich et al. 2012; Tilman et al. 2001; Zheng et al. 2024); (ii) increasing species asynchrony over time via temporal complementarity effects (Allan et al. 2011; Isbell et al. 2011; Zavaleta et al. 2010) that stabilise productivity from year-to-year; and (iii) increasing stabilising effects of species richness on population stability over time. However, whether these patterns and processes apply to other biodiversity experiments and/or whether ongoing 'chronic press' global changes influence such temporal trends remains to be tested.

Herein, using a 25-year dataset from a unique biodiversity-global change experiment (BioCON) (see Materials and Methods), we asked how resource enrichment global changes, specifically elevated $CO_2$ and N-addition, modify the impacts of diversity on ecosystem stability and its components (productivity, species asynchrony and population stability), and how these effects change over time. We found that the global change treatments, especially in combination, did not erase but gradually weakened the effects of diversity on ecosystem stability, likely because global changes decreased asynchrony amongst species over time.

## 2 | Methods

Experiment design: The BioCON experiment at Cedar Creek Ecosystem Science Reserve (East Bethel, MN, USA, 45°40′ N, 93°18′ W) was established in 1997 and manipulates biodiversity, $CO_2$ and N (Reich et al. 2001, 2004). The full-factorial experiment comprises 296 $2\times2$ m plots that vary in their planted diversity (1, 4, 9 or 16 species), N (ambient or $+4\,\mathrm{g\,m^{-2}\,year^{-1}}$) and $CO_2$ (ambient or $+180$ ppm) treatments that are applied in a split-plot design for the $CO_2$ treatment. By 2012, 36 of the total plots, all belonging to 9-species mixtures, had been used for a sub-experiment involving drought and temperature treatments, leaving 260 plots to be a part of the main experiment. The 16 species in the experiment were chosen to represent common or naturalised prairie species in the region and spanned four functional groups: $C_3$ grasses (*Bromus inermis*, *Elymus repens* [formerly *Agropyron repens*], *Koeleria macrantha* [formerly *Koeleria cristata*] and *Poa pratensis*), $C_4$ grasses (*Andropogon gerardii*, *Bouteloua gracilis*, *Schizachyrium scoparium* and *Sorghastrum nutans*), legumes (*Amorpha canescens*, *Lespedeza capitata*, *Lupinus perennis* and *Petalostemum villosum*) and non-legume forbs (*Achillea millefolium*, *Anemone cylindrica*, *Asclepias tuberosa* and *Solidago rigida*). Species found in the plots that were not part of the planted species list were weeded annually and no species have been re-seeded after 1998. Over the years, species have been lost from the plots likely due to species interactions and community dynamics, but also due to global change treatments (Reich 2009; Reich et al. 2024). Therefore, we also estimate a realised richness from plant cover systematically sampled annually. Additionally, the experiment is located on sandy soils generally poor in N (Grigal and Station 1974) and the plots were burned in spring for half of the years between 2000 to 2012 and every fall since 2013 (Adair et al. 2009).

Plant biomass: Peak aboveground plant biomass was estimated from $10\times100$ cm clip strips collected late each growing season in early August. The clip strips were collected from pre-marked locations that were moved annually to reduce effects of repeatedly clipping in one part of the plot. The clipped biomass was sorted to the species level and weighed. Additionally, species-specific cover data were collected from a permanent 50 × 100 cm region (that was never clipped or moved) for each plot in every year (except for a subset of 4 species plots in 2020, representing < 1% of all data). We counted all species present in the cover data to estimate realised species richness. Lastly, to obtain a more representative estimate of species-specific biomass and to impute missing biomass values when cover data were present, we generated species-specific correlations between the clip strip biomass and cover values to then estimate species biomass data for all years based on cover data and whole plot clip strip biomass (Mohanbabu et al. 2024). This method also allowed us to account for some of the variability in biomass-cover associations across different species. We used clipped biomass measures for the whole plot metrics such as ecosystem stability and productivity, and only used estimated biomass for species level metrics such as asynchrony and population stability.

Diversity–stability relationships: We calculated stability as the mean divided by the standard deviation of aboveground biomass (i.e., clipped biomass) for each plot over a moving 5-year window which gave us 21 5-year groups. We then divided all the stability values by the mean stability value of monocultures for each temporal block and plot to get relative stability. This standardised value of stability only changes the value of the intercept and not the slope of the relationship. Thus, it does not influence the interpretation of results and further allows for comparison of stability across diversity levels on the same scale. For each time block and global change treatment combination, we estimated the slope of the relationship between log-transformed variables: planted species richness (or realised species richness) and relative stability. We repeated the above analysis for different time intervals from 3- to 7-year intervals and the results were consistent for the different time intervals (Figure S1). Based on that we focused on 5-year intervals/groups for the rest of the analyses. We then examined temporal trends between log-transformed realised species richness and components of stability, i.e., mean and standard deviation in productivity.

Species asynchrony and population stability: We calculated asynchrony as the ratio of the variance of community biomass (i.e., clipped biomass) across years to the square of the sum of standard deviations across years of the estimated species-specific biomass, i.e., $1 - \frac{\sigma^2_{community}}{\left(\sum \sigma_{species}\right)^2}$ (Loreau and de Mazancourt 2008). Larger values indicate greater asynchrony with 1 being perfect asynchrony amongst species and 0 being perfect synchrony amongst species. We estimated population stability as the ratio of community stability to the sum of standard deviations across the years for species-specific biomass (Thibaut and Connolly 2013). Impact of diversity on asynchrony and population stability were assessed in the same way as for ecosystem stability.

Community composition shifts in resource-acquisitiveness over time: We calculated community-weighted means of specific leaf area (SLA) for each of the plots to track temporal trends. SLA has been estimated for most of the BioCON species in monocultures over multiple years using standard trait protocols (Pérez-Harguindeguy et al. 2013). We calculated the mean trait value for each species under the four global change treatments and used those values for all the years. Data from BioCON were unavailable for *Poa pratensis* and *Bouteloua gracilis*, so we imputed trait values under ambient conditions for these two species using other available data from the Cedar Creek Ecosystem Science Reserve. As we are interested in community shifts, we use the realised biomass estimates of the species rather than planted abundances. Therefore, trends in the community-weighted means directly reflect shifts in community composition over time (i.e., without estimation of potential plastic responses).

## 2.1 | Statistical Analysis

All statistical analyses were performed in R 4.3 (R Core Team 2023) and R 4.5 (R Core Team 2025).

We ran a linear mixed effects model using lmerTest (Kuznetsova et al. 2017) and glmmTMB (McGillycuddy et al. 2025) packages. All models were structured similarly and had a dependent variable such as relative stability, and planted (or realised) richness [natural log-transformed], N treatment, $CO_2$ treatment, and continuous linear and quadratic year grouping terms (i.e., scaled temporal blocks) and their interactions as independent variables. All models included plot nested within ring as the random effect to account for repeated measures assuming compound symmetry in a split-plot design. We ran similar models for mean productivity, SD productivity, and population stability with lmer(), and used log-transformed dependent variables to meet assumptions of normality and heteroscedasticity. For asynchrony, we used a generalised mixed model to accommodate the large number of 0 values. All monocultures have an asynchrony value of 0, and the other asynchrony values are bounded between 0 and 1. To account for this, we used the ordered beta regression family (Kubinec 2023) with 'link = logit' in the glmmTMB package. We excluded a small number of data points (~0.3%) from the asynchrony model that had -Inf values because of a species becoming locally extinct in monoculture during the later years of the experiment. However, for models with realised richness, we added 0.1 to all richness values to include a small percent (0.3%) of the plot × year group combinations that show local extinction of species, usually in monocultures with *Anemone cylindrica* (but also single instances for *Koeleria cristata*, *Asclepias tuberosa* and *Solidago rigida*) in the later years of the experiment. The model results are summarised in Tables S1 and S3 for planted richness and realised richness, respectively.

The compound symmetry correlation structure was sufficient to account for any temporal autocorrelation in the models for ecosystem stability, mean productivity and SD of productivity. But, for both asynchrony and population stability, we found effects of temporal lag in acf() plots. This is perhaps not surprising given that our models include values calculated on moving windows. However, potentially because of the complex nature of the models (i.e., 4-way interaction terms, split-plot design, etc.), including AR1 structure resulted in convergence issues. Instead, we analysed a subset of the data by excluding 2–3 consecutive year groups to reduce temporal autocorrelation. As this subset has fewer datapoints, we also had to simplify the model by dropping the quadratic year term. The results are presented in Table S5 and the 3- and/or the 4-way interaction terms (with linear year group terms) are still statistically significant for asynchrony and not significant for population stability similar to the full model. Unfortunately, we could not compare the quadratic temporal trends with this smaller subset of data.

Additionally, we ran a simple linear model using lm() with the slope of the log diversity-log ecosystem stability (or mean/SD productivity, asynchrony and population stability) relationship as the dependent variable and year groups (including a quadratic term), N and $CO_2$ treatments, and their interactions as independent variables to better understand the influence of global change treatment on the slope of the relationships over time. The model results are summarised in Tables S2 and S4 for planted richness and realised richness, respectively.

## 3 | Results

The global changes, elevated $CO_2$ and enriched N, influenced ecosystem stability and productivity to varying extents across the diversity gradient (Table S1), consequently modifying diversity impact on ecosystem functions. Elevated $CO_2$ and N enrichment gradually decreased the positive effect of species richness on ecosystem stability over time (Figure 1, Table S1). Under ambient conditions, the impact of diversity on ecosystem stability (i.e., slope of log diversity- log ecosystem stability) increased almost monotonically over time for 18 years such that the diversity impact on ecosystem stability in the 2015–2019 period (i.e., year group 18 based on 5-year moving window intervals) (see Materials and Methods) was 2.6 times that of the impact in 1998–2002 (year group 1), before starting to decline in the 2016–2020, 2017–2021 and 2018–2022 year groups (Figure 1a). When either N or $CO_2$ was enriched, diversity had a larger positive impact on ecosystem stability compared to ambient conditions in the initial years as indicated by steeper slope values (inset plots), and such positive impacts of diversity under global changes also increased for a few years (up to a decade). But this initial strengthening effect was short-lived and declined over the rest of the experiment such that the impact of diversity on ecosystem stability in 2018–2022 was only two-thirds that in 1998–2002, for N or $CO_2$ enrichment (Figure 1b,c). This negative effect over time on the diversity impact on ecosystem stability occurred faster and was even more pronounced when both N and $CO_2$ were enriched, as the slope of diversity effects on ecosystem stability declined continuously and by 2018–2022 was almost half its value in 1998–2002 (Figure 1d, Table S2).

These temporal trends held and were similar when we considered different interval periods ranging from three to seven

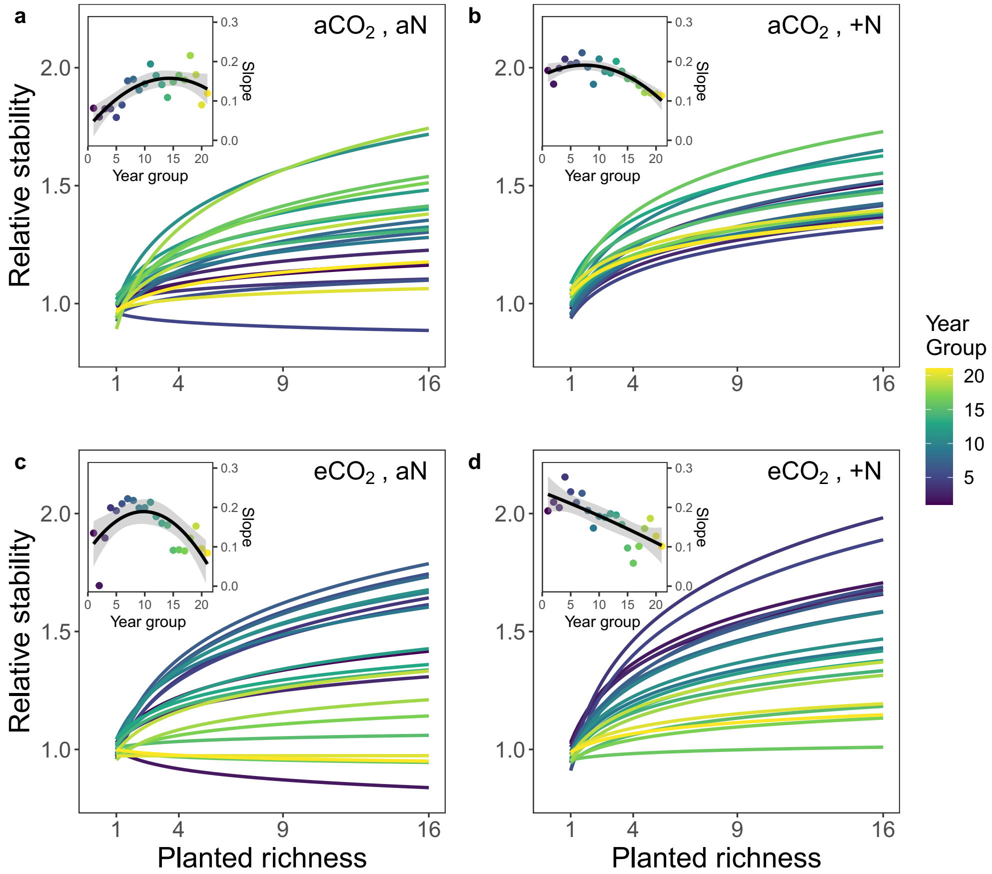

**FIGURE 1** | Temporal trends in the impact of diversity on relative ecosystem stability under global change treatments. The main graphs show the diversity–relative stability relationship for each 5-year group and the inset graphs show the slope of log–log relationship over time. The ecosystem stability values are relative to the mean monoculture ecosystem stability in each year group. Ambient treatment conditions are denoted by 'a' and enriched treatments are denoted by 'e' or '+'.

years, with 3-year groups showing slightly weaker temporal trends and 7-year groups showing slightly stronger temporal trends, especially under ambient $CO_2$ treatments (Figure S1). The temporal trends for 5-year groups were also very similar when we replaced planted richness with realised richness as a predictor of ecosystem stability (Figure S2a–d). Thus, regulation by enriched $CO_2$ and N of diversity impacts was consistent regardless of methodological choices or diversity measures used.

The 25-year dynamics of diversity effects on ecosystem stability could not be explained by the dynamics of diversity effects on productivity, as diversity–productivity relationships strengthened over time in all treatments. Planted richness (and realised richness) markedly increased productivity in all time groups in all treatment combinations and with increasing enhancement over time such that the impact of planted diversity on productivity more than doubled (or tripled for N enrichment under both $CO_2$ levels) in 2018–2022 compared to 1998–2002 (Figure 2; Figure S2e–h; Table S1). These effects of N and/or $CO_2$ enrichment were similar and hence these drivers had relatively less influence on the temporal trend of the diversity effect on productivity compared to their effects on the temporal trends of diversity effects on ecosystem stability (Table S1; Figure 2b–d). However, the significant 4-way interaction term indicates that global change treatments influenced the diversity–productivity relationships in complex ways (Tables S1 and S3).

The impact of diversity (both planted and realised richness) on interannual variation in productivity (i.e., standard deviation or SD) also increased over time and sometimes non-monotonically (Figure S3, Tables S1–S4). Thus, diversity boosted both productivity and its variability and increasingly so over time. However, the increase in diversity impact on SD over time was less steep for ambient conditions and steeper for global change treatments relative to the corresponding trends for productivity (Figure S4). This divergence or convergence in how diversity changed mean and variability of productivity over time across treatments explains the strengthening or weakening effect, respectively, of diversity on ecosystem stability.

Ecosystem stability can also increase with increasing species asynchrony or population stability. Early in the experiment, the impact of diversity on asynchrony was positive and similar across all $CO_2 \times N$ treatment combinations. These impacts of diversity on asynchrony remained positive but declined over the duration of the experiment in all $CO_2 \times N$ combinations. However, the decline in the positive impact of diversity on asynchrony over time was relatively modest for ambient conditions, with a 22% decline in 2018–2022 compared to 1998–2002 (Figure 3a), whereas it declined by 35% under either enriched N or $CO_2$ treatments over the same timeframe (Figure 3b–d). Plots with both elevated $CO_2$ and N inputs showed the steepest declines, such that the impact of diversity

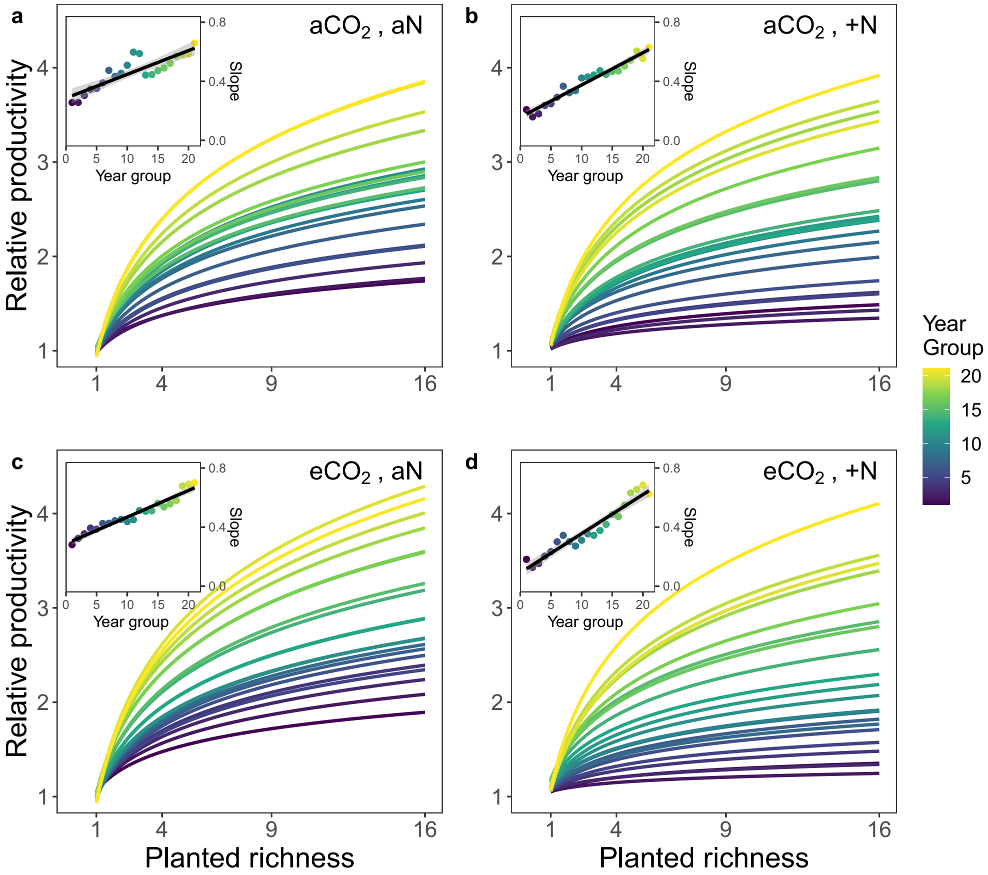

**FIGURE 2** | Temporal trends in the impact of diversity on relative mean productivity under different global change treatments. The main graphs show the diversity–mean productivity relationship for 5-year intervals whereas the inset graphs show the slope of the log–log relationship changing over time. The productivity values are relative to the mean monoculture productivity in each year group. Ambient treatment conditions are denoted by ‘a’ and enriched treatments are denoted by ‘e’ or ‘+’.

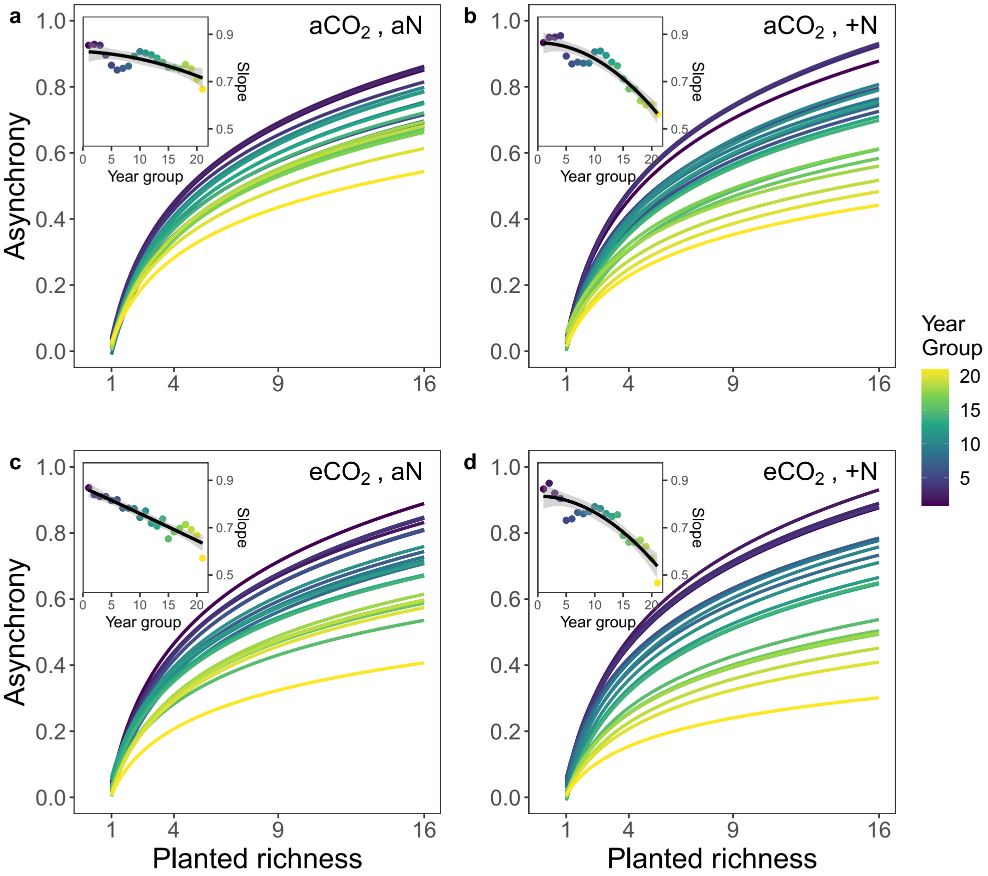

**FIGURE 3** | Temporal trends in the impact of diversity on species asynchrony under global change treatments. The main graphs show the planted diversity–asynchrony relationship for 5-year intervals whereas the inset graphs show the slope of the log–log relationship changing over time. Asynchrony is unitless and bounded between 0 and 1. Ambient treatment conditions are denoted by 'a' and enriched treatments are denoted by 'e' or '+'.

on asynchrony was only half as strong in 2018–2022 compared to 1998–2002 (Figure 3d). These interactive effects of N and $CO_2$ on asynchrony over time were also found in data which excluded overlapping intermediate years to reduce temporal autocorrelation (Table S5).

The weakening over time of the diversity-induced enhancement of species asynchrony was likely due at least in part to species loss in diverse plots, as corroborated by the less negative trends in the impact of diversity on asynchrony over time when we used realised richness as opposed to planted richness as a predictor of asynchrony (Figure S5), except perhaps for the +*N* alone treatment. The impact of diversity on asynchrony showed a slight increase with time under ambient conditions (Figure S5a) and a consistent decline under N enrichment at ambient $CO_2$ in the later years of the experiment (Figure S5b). In contrast, for plots experiencing elevated $CO_2$, at both N treatment levels, the positive impact of diversity on asynchrony showed non-monotonic change with planted richness and negative temporal trend with realised richness (Figure S5c,d; Tables S1 and S3). However, the presence of a significant 4-way interaction term in these models implies that the temporal change in diversity–asynchrony relationship under elevated $CO_2$ also depends on N levels. Nonetheless, these varying temporal trends in the impact of diversity on asynchrony suggest that global changes also reduced asynchrony in high diversity plots over time through other mechanisms besides just species loss.

On the other hand, the impact of diversity on population stability was consistently negative across all treatments, i.e., population stability was lower in more diverse plots, consistent with theoretical predictions (Tilman 1996). Over the entire 25-year period, the impact of diversity on population stability became less negative under ambient conditions (Figure 4a) and N enrichment (at both $CO_2$ levels) (Figure 4b,d). In contrast, under elevated $CO_2$ at ambient N, the impact of diversity on population stability became markedly less negative for most of the first decade, but then declined in subsequent years (i.e., became more negative). The non-monotonic responses are supported by the significant interaction terms when time was treated as a quadratic variable (Tables S1–S4). The temporal trends in the impact of realised richness (Figure S5e–h) were similar to those of the impact of planted richness on population stability.

These global change treatment-specific impacts of diversity on population stability and asynchrony may have modified the influence of diversity on ecosystem stability over time. For ambient and N enrichment treatments, the reduced negative effect of planted diversity on population stability over time likely compensated for the negative effect of declining asynchrony in high diversity plots (Figure 3a,b,d). However, for elevated $CO_2$ and

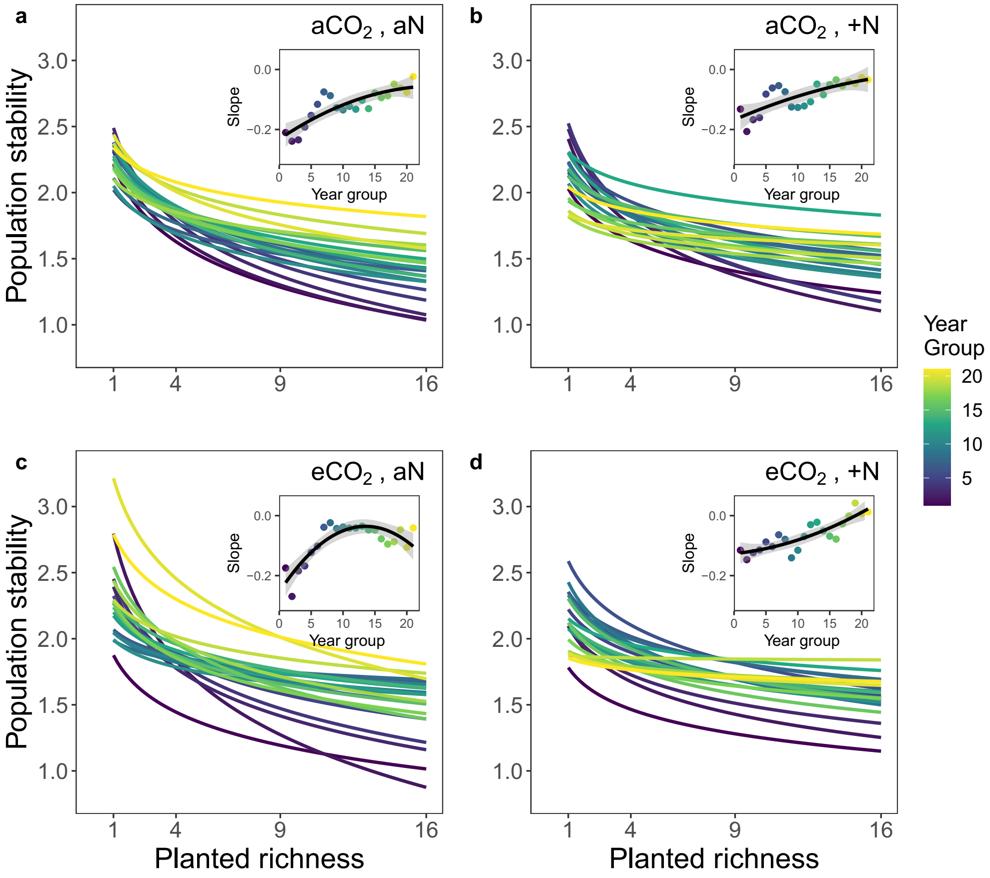

**FIGURE 4** | Temporal trends in the impact of diversity on population stability under global change treatments. The main graphs show the planted diversity-population stability relationship for 5-year intervals whereas the inset graphs show the slope of the log–log relationship changing over time. Ambient treatment conditions are denoted by 'a' and enriched treatments are denoted by 'e' or '+'.

ambient N treatments, the reduced negative impact of diversity on population stability may have stabilised the diverse plots early in the experiment but the declining impact of diversity on asynchrony (Figure 3c) and population stability (Figure 4c) in the later years may have reduced the impact of diversity on ecosystem stability (Figure 1c). This potentially synergistic effect of both reduced population stability and asynchrony likely explains some of the lowest slope values for the diversity effect on ecosystem stability (Figure 1c).

## 4 | Discussion

Using a unique quarter-century experiment that manipulated both biodiversity and global change treatments, we show for the first time that two global changes can gradually weaken—but not eliminate—the positive effect of diversity on ecosystem stability (Figure 1). The temporal decline in the strength of the impact of diversity on ecosystem stability under global changes was due to larger increases in the impact of diversity on interannual variability in productivity compared to its mean, likely driven by decreasing asynchrony amongst species in diverse plots over time (Figure 3). We also discovered a remarkably consistent increase in the enhancement of productivity by diversity over time under all global change conditions, providing evidence that this ecosystem service of biodiversity not only persists but continues to gain strength for longer periods than demonstrated previously. However, surprisingly, this strengthening of diversity impact on productivity over time was not sufficient to offset the weakening impact of diversity on species asynchrony, leading to a weakening of the impact of diversity on ecosystem stability under global changes. Given the ongoing anthropogenic changes in N deposition and CO$_2$ levels globally along with simplification of communities via biodiversity loss, our results suggest concern about potential long-term negative consequences for ecosystem stability in the following decades.

Although it remained positive, a decreasing impact over time of diversity on species asynchrony emerged as a likely explanation for declining impact of diversity on ecosystem stability over time under global change conditions. Such a decline in the impact of diversity on asynchrony over time could be, in part, a result of species loss driven by stochastic processes or N and CO$_2$ addition (Harpole et al. 2016; Isbell et al. 2013; Reich 2009; Reich et al. 2024). Additionally, species may become less asynchronous (i.e., more synchronous) under resource addition (Figure 3) (Hautier et al. 2014, 2020; Zhang et al. 2016), as novel environments may modify population dynamics of species in previously unobserved ways. Alternatively, asynchrony may also decline as more resource-acquisitive species with similar traits, function, and sensitivity to temporal changes may be favoured (Lepš et al. 2018; Polley et al. 2013). There is evidence for both species' loss (Reich et al. 2024) and community shifts towards more resource-acquisitive species in response to treatments over time in this experiment (Figure S6), which likely explains decreases in asynchrony over time in diverse plots under resource addition, similar to other eutrophication studies (Hautier et al. 2014, 2020). Moreover, the fact that we find similar trends with both planted and realised richness also suggests that the weakening effect of diversity on ecosystem stability was not just driven by species loss under global changes. Therefore, as communities tend towards fewer species that are more likely to be functionally similar, they can lose some of the positive effects of temporal complementarity that may increase interannual variation, effectively making communities less stable.

Surprisingly, the strengthening effect of diversity on mean productivity over the years under both ambient and global change conditions was not a strong predictor of changes in the impact of diversity on ecosystem stability over time. Although our results are consistent with other studies that have shown a positive effect of diversity on both ecosystem productivity and stability (Wang et al. 2021; Yan et al. 2021), the diverging effects of diversity on ecosystem productivity and stability over 25 years in our study indicate potential for diversity effects on ecosystem stability to be decoupled from diversity effects on mean productivity as has been suggested in previous studies (Cardinale et al. 2013; Su et al. 2022). Such decoupling may be more likely if the high productivity in diverse plots under elevated $CO_2$ and enriched N treatments makes plants more susceptible to limitation by other resources such as water, ultimately causing these plots to respond strongly and synchronously to interannual variation in water. Nonetheless, this result emphasises that positive effects of global changes on the effects of diversity on productivity need not imply strengthening effects of global change on diversity impacts on ecosystem stability.

The combined effect of N and $CO_2$ addition on the influence of planted diversity on ecosystem stability was only modestly lower than would be expected by considering the effects of enrichment of the two global changes individually (i.e., by adding their independent effects together). This result is in agreement with early results from the same experiment, which showed that there were no three-way interactions between N, $CO_2$ and diversity levels on biomass production (Reich et al. 2001). However, a more recent analysis shows that elevated $CO_2$ first dampened and then amplified species loss due to N addition in this experiment (Reich 2009; Reich et al. 2024). This interactive effect of N and $CO_2$ on species loss may have indirectly influenced species composition and asynchrony in these plots but was not strong enough to impact the influence of diversity on ecosystem stability.

The nature of our experiment likely imposes limitations on extrapolating the results to larger scales. At larger scales, dispersal and other metacommunity processes can alter effects of biodiversity on productivity and ecosystem stability (Isbell et al. 2017; Loreau et al. 2003). Our experiment was designed to consider local biodiversity effects and thus, through weeding, eliminates effects of dispersal. Although it would be intractable to conduct an experiment like ours at large spatial extent (e.g., with plots of 1 ha) with elevated $CO_2$, N enrichment, and plant diversity treatments, additional studies in naturally assembled ecosystems will be needed to build a multiscale understanding of these effects. Additionally, ecosystem stability may be influenced by biotic agents such as herbivory or diseases that may have had varying impacts at different timepoints in the experiment, but we lack the data to explore those mechanisms in depth. Nonetheless, this is by far the longest running biodiversity experiment in the world that also manipulates other important global changes, in this case N deposition and $CO_2$, thereby offering a framework to think about effects of global change and diversity loss at longer temporal and larger spatial scales. Thus, our study complements other published (Davidson et al. 2025; Su et al. 2022) and ongoing work on understanding effects of global changes on temporal stability in natural ecosystems.

In conclusion, using a quarter-century experiment, we showed that over time N and $CO_2$ enrichment can weaken (but not eliminate) the positive effect of diversity on ecosystem stability due to decreasing asynchrony amongst species and species loss driven by these global changes in diverse plots. Additionally, we showed that increasingly positive effects of diversity on productivity persisted for the full 25 years of the study and were not altered by global change treatments, providing further evidence for the ubiquity of these effects. However, despite such strong trends, diversity enhancement of productivity over time may not routinely offset the negative effects of decreasing asynchrony at high diversity on ecosystem stability over time. Thus, our results indicate that, over two and a half decades, elevated $CO_2$ and N eutrophication can gradually erode the positive effects of biodiversity on ecosystem stability. This is particularly important as it suggests that increasing biodiversity alone might not be sufficient to maintain stable ecosystems and any action leading to both biodiversity loss and global change may only worsen their impact on ecosystem stability.

### Author Contributions

N.M. conceptualised this study and led the formal analyses, both with input from the other authors, and wrote the first draft of the manuscript. P.B.R. designed the long-term experiment; P.B.R. and S.E.H. implemented it over the 25 years; and P.B.R., S.E.H. and F.I. acquired funding for it. All authors contributed to revising the manuscript.

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### Conflicts of Interest

The authors declare no conflicts of interest.

### Data Availability Statement

All data and code are available on EDI at https://doi.org/10.6073/pasta/55fd0ed9756dc98de2e84ae6ddf543b7.

### Peer Review

The peer review history for this article is available at https://www.webofscience.com/api/gateway/wos/peer-review/10.1111/ele.70170.

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## Supporting Information

Additional supporting information can be found online in the Supporting Information section.