ECOLOGY

# Drought soil legacy alters drivers of plant diversity-productivity relationships in oldfield systems

**Nianxun Xi[1]†‡, Dongxia Chen[1]†, Michael Bahn[2], Hangyu Wu[1], Chengjin Chu[1]*, Marc W. Cadotte[3], Juliette M. G. Bloor[4]**



**Ecosystem functions are threatened by both recurrent droughts and declines in biodiversity at a global scale, but the drought dependency of diversity-productivity relationships remains poorly understood. Here, we use a two-phase mesocosm experiment with simulated drought and model oldfield communities (360 experimental mesocosms/plant communities) to examine drought-induced changes in soil microbial communities along a plant species richness gradient and to assess interactions between past drought (soil legacies) and subsequent drought on plant diversity-productivity relationships. We show that (i) drought decreases bacterial and fungal richness and modifies relationships between plant species richness and microbial groups; (ii) drought soil legacy increases net biodiversity effects, but responses of net biodiversity effects to plant species richness are unaffected; and (iii) linkages between plant species richness and complementarity/selection effects vary depending on past and subsequent drought. These results provide mechanistic insight into biodiversity-productivity relationships in a changing environment, with implications for the stability of ecosystem function under climate change.**

## INTRODUCTION

Plant diversity and climate change are key drivers of primary productivity and nutrient cycling (*1*–*3*). Interactions between plant diversity loss and the droughts associated with climate change have faced increasing attention in recent years, and widespread evidence suggests that declines in ecosystem productivity due to diversity loss may be exacerbated by drought events (*4*–*6*). Improved understanding of the mechanisms underlying the linkages between diversity-productivity relationships and drought stress is critical for climate mitigation efforts and the implementation of effective nature-based climate solutions (*5*, *7*).

Over the last 20 years, a large body of literature has emphasized the role of plant resource strategies and plant-plant interactions for plant diversity-productivity relationships (*8*–*10*). However, recent work suggests that plant-soil interactions may be equally important for plant community dynamics and biodiversity effects (BEs) (*11*–*14*). Plants can influence soil microbial community composition and generate soil legacies through the production of biochemically diverse litter and root exudates (*15*–*17*). In return, soil microbes, and fungi in particular, have the potential to modulate plant diversity-productivity relationships by enhancing complementarity effects (CEs; i.e., generating higher relative performance of species in mixtures compared with that expected based on monocultures), because of the dilution of pathogenic effects and the accumulation of diverse soil mutualists in multispecies mixtures (*12*, *13*, *18*–*23*). Soil microbes may also influence plant diversity-productivity relationships via selection effects (SEs), when particular microbial groups reduce the performance of their host plant species in mixtures, thus enhancing the competitive advantage of other plant species (*20*, *24*, *25*).

Environmental conditions affect soil microorganisms (*26*, *27*), and it is therefore likely that mediation of plant diversity-productivity relationships by soil microbes may depend on local abiotic conditions. Many studies have documented shifts in both fungal and bacterial community composition in response to decreases in water availability and subsequent rewetting events (*28*–*32*). At the same time, drought-induced changes in plant physiology and/or plant community structure may contribute to drought soil legacies via indirect drought effects on the microbial community (*33*–*35*). Such drought soil legacies may affect longer-term plant community structure and function through plant-soil feedbacks (*36*–*38*), but the magnitude and persistence of drought legacies in soil microbes are currently subject to debate (*39*). To date, information on the influence of drought soil legacies on plant diversity-productivity relationships is lacking, and responses of plant diversity-productivity relationships to subsequent drought events are unclear.

Here, we conducted a two-phase experiment to investigate whether and how chronic reductions in rainfall (press droughts) modify soil legacies and plant diversity-productivity relationships under subsequent drought, with a particular focus on the role of fungal mutualists [arbuscular mycorrhizal fungi (AMF)] and pathogens. In phase I, 120 model plant communities with five species richness levels were grown in homogeneous soils under either ambient or drought conditions to generate "conditioned" soil samples. In phase II, newly established plant communities were provided with soil inoculums with or without previous drought exposure and subjected to either drought or ambient watering conditions (Fig. 1). Specifically, we hypothesized that (i) drought and plant species richness coinfluence soil microbial communities, (ii) drought soil legacy influences BEs on plant productivity, and (iii) subsequent drought alters effects of drought soil legacy on diversity-productivity relationships.

## RESULTS

### Interactive effects of drought and plant species richness on soil microbial communities in phase I

Analysis of soil samples across the biodiversity experiment yielded on average 594 fungal operational taxonomic units (OTUs) and

[1]State Key Laboratory of Biocontrol, School of Life Sciences/School of Ecology, Sun Yat-sen University, Guangzhou 510275, China. [2]Department of Ecology, University of Innsbruck, 6020 Innsbruck, Austria. [3]Department of Biological Sciences, University of Toronto-Scarborough, 1265 Military Trail, Toronto, ON M1C 1A4, Canada. [4]Université Clermont Auvergne, INRAE, VetAgro-Sup, UREP, 5 Chemin de Beaulieu, F-63100 Clermont-Ferrand, France.
*Corresponding author. Email: chuchjin@mail.sysu.edu.cn
†These authors contributed equally to this work.
‡Present address: College of Forestry, Hainan University, Haikou 570228, China.

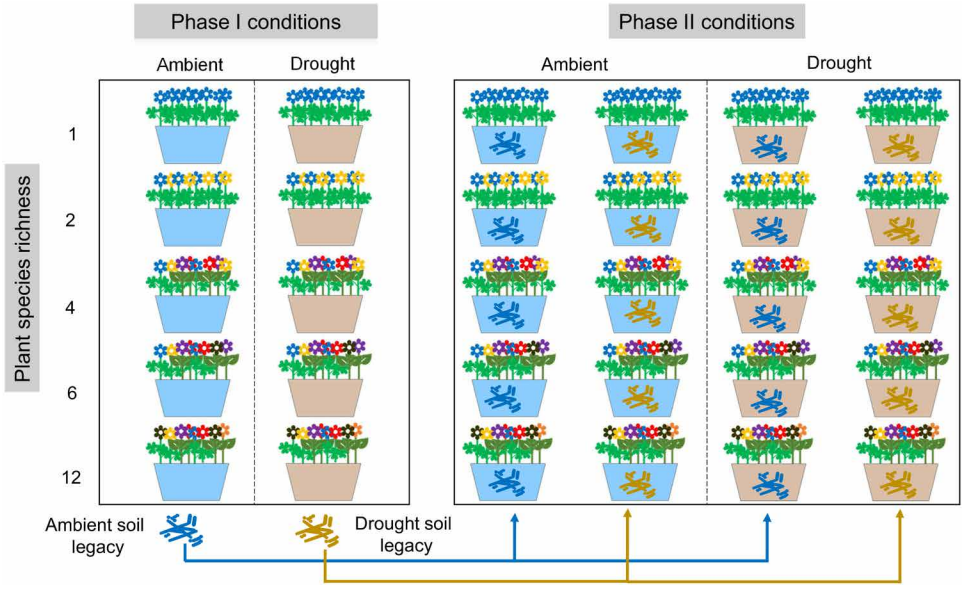


**Fig. 1. Schematic depiction of the experimental design.** The study combined a soil legacy experiment and a plant diversity-productivity relationship experiment. In phase I, plant communities with a range of species-richness levels were grown in homogeneous soils under drought or ambient conditions to generate soil microbial legacies. In phase II, the effects of soil microbial legacies were tested on newly established plant communities grown under drought or ambient conditions, using soil inoculums conditioned by the same plant communities.

4683 bacterial OTUs per mesocosm (fig. S1). Overall, 105 classes of bacteria and 45 classes of fungi were identified (supplementary data S1); soil fungal communities were strongly dominated by Sordariomycetes, which accounted for 51% of the sequences, whereas bacterial communities were dominated by Acidobacteriia, Alphaproteobacteria, Gammaproteobacteria, Verrucomicrobiae, and Deltaproteobacteria, which jointly represented 54% of the sequences. Fungal community structure was significantly influenced by both the level of soil moisture content ($F_1 = 12.94$, $P < 0.001$) and plant species richness ($F_1 = 3.78$, $P < 0.001$; Fig. 2A). These factors explained 15.13% of the variation in soil fungal community composition. Bacterial community structure was also affected by the level of soil moisture content and plant species richness (soil moisture: $F_1 = 23.91$, $P < 0.001$; plant species richness: $F_1 = 4.06$, $P < 0.001$; Fig. 2B), together explaining 23.09% of the variation in bacterial community composition.

In general, soil fungal richness decreased in response to experimental drought but was unaffected by plant species richness (Table 1 and Fig. 2C). In contrast, effects of plant species richness on bacterial richness differed between the ambient and drought treatments (species richness × drought interaction; Table 1 and Fig. 2D). Under ambient conditions, bacterial richness showed a negative relationship with plant species richness (adjusted $R^2 = 0.15$ and $P = 0.003$), but bacterial richness was unrelated to plant species richness under drought conditions ($P = 0.245$; Fig. 2D). Under ambient conditions, soil moisture content also decreased with increasing plant species richness (fig. S2), but accounting for relationships between bacterial/fungal richness and soil moisture did not alter the observed relationships between bacterial/fungal richness and plant species richness (Table 1).

Experimental drought also decreased AMF richness, with greatest drought-induced decreases in AMF observed under higher species richness levels (species richness × drought interaction; Table 1 and Fig. 2E). Nevertheless, AMF richness showed a positive relationship with plant species richness in both "drought" treatments (Fig. 2E; adjusted $R^2 = 0.29$, $P < 0.001$ and adjusted $R^2 = 0.14$, $P = 0.003$ for ambient and drought treatments respectively; table S1). Effects of plant species richness on fungal pathogen richness differed between the ambient and drought treatments (species richness × drought interaction; Table 1 and Fig. 2F). Under ambient conditions, fungal pathogen richness showed a negative relationship with plant species richness (adjusted $R^2 = 0.12$, $P = 0.007$), but fungal pathogen richness was unrelated to plant species richness under drought conditions ($P = 0.145$; table S1 and Fig. 2F). Fungal saprotroph richness did not show any responses to plant species richness, drought treatments, or their interaction (table S2 and fig. S2). In parallel, plant productivity showed a positive relationship with plant species richness under ambient conditions in phase I (adjusted $R^2 = 0.12$, $P = 0.006$), but no relationship with plant species richness under drought conditions (adjusted $P = 0.907$; fig. S3).

## Plant biomass and BEs in phase II

In general, plant aboveground biomass increased with plant species richness (Table 2, table S3, and Fig. 3A). However, the positive effects of plant species richness on aboveground biomass were greater in communities with drought soil legacy treatments (i.e., with soil previously exposed to drought) compared with those without drought soil legacy (plant species richness × drought legacy interaction; Table 2 and Fig. 3A). In addition, plant species richness effects were lower in mesocosms experiencing phase II drought compared with those in ambient treatments (plant species richness × phase II drought interaction; Fig. 3A).

Net BEs and CEs increased with plant species richness across all drought treatments (Table 2, table S3, and Fig. 3, B and C). Positive effects of species richness were greater for ambient plant communities compared with droughted-plant communities (species

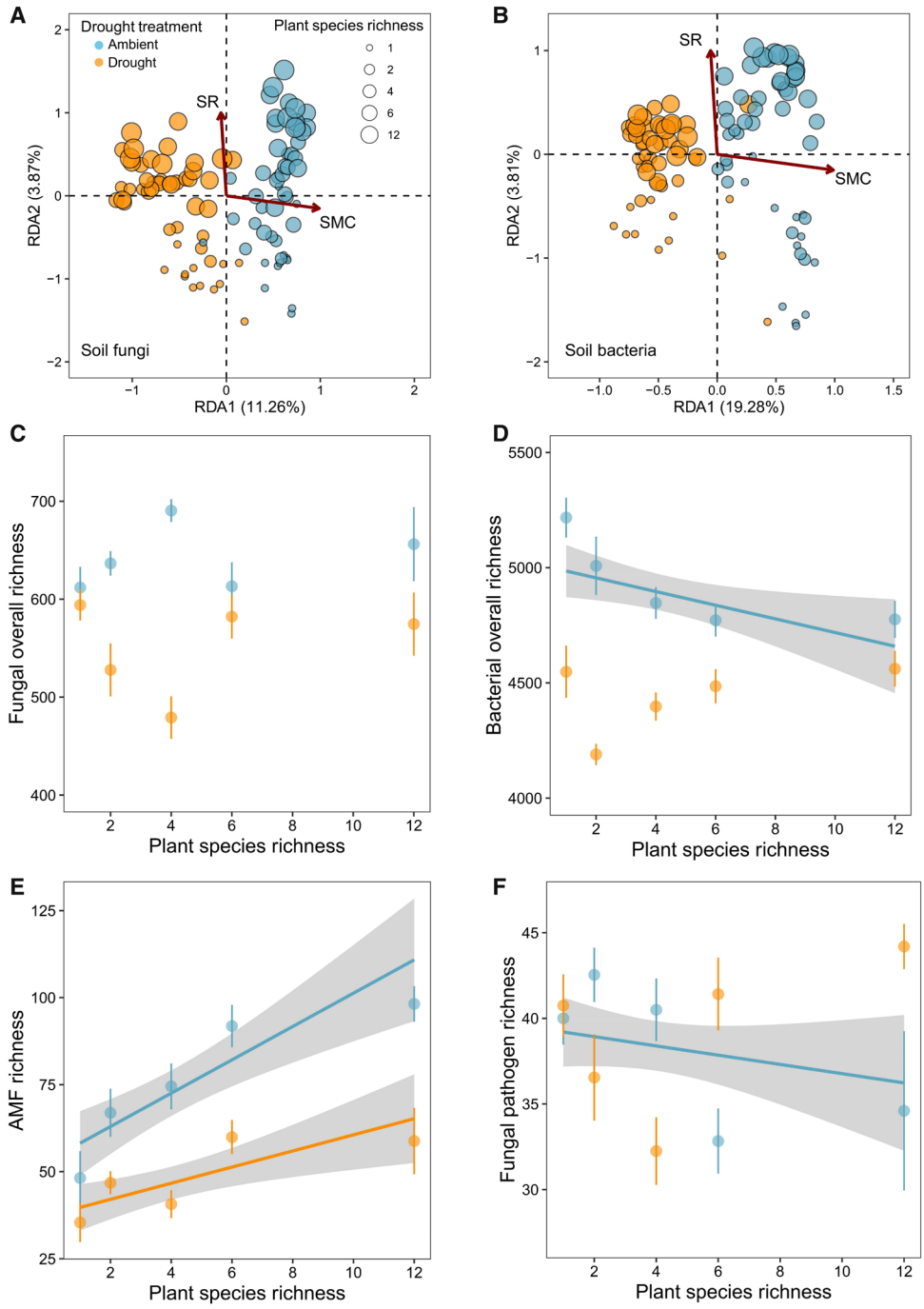


**Fig. 2. Soil microbial communities in phase I.** Redundancy analysis (RDA) showing the influence of soil moisture content and plant species richness on (**A**) soil fungal community composition and (**B**) soil bacterial community composition in phase I (SR, plant species richness; SMC, soil moisture content). Effects of drought treatments and plant species richness on (**C**) total soil fungal richness, (**D**) total bacterial richness, (**E**) AMF richness, and (**F**) fungal pathogen richness. Means and SE are given. Lines indicate significant relationships between plant species richness and microbial variables at the mesocosm level ($P < 0.05$). Shaded areas indicate 95% confidential intervals of fitted lines.

richness × ongoing drought interaction; Table 2 and Fig. 3, B and C). Net BEs and CEs were also greater in communities with soil previously exposed to drought, driven by a particularly large effect of drought history under subsequent ambient conditions (drought legacy × phase II drought interactions; Table 2 and Fig. 3, B and C). Plant species richness effects on net BEs did not vary with drought legacy treatments, nor was there any interaction between plant species richness, drought legacy, and ongoing drought treatments on net BEs (Table 2). However, plant species richness interacted with drought legacy and ongoing drought treatments to influence CEs (species richness × drought legacy × ongoing drought interaction; Table 2). The magnitude of positive plant species richness effects on

**Table 1. Results from linear mixed-effects models for soil microbes in phase I.** The interactive effects of plant species richness and drought treatments on soil fungal richness, bacterial richness, AMF richness, and pathogen richness in phase I, with soil moisture content over phase I (average values) as a covariate. Degrees of freedom (df) and *F* values are given. Bold types are significant effects (**P* < 0.05, ***P* < 0.01, and ****P* < 0.001). SR, species richness.

| Variable | df | *F* |
|---|---|---|
| (a) **Fungal richness** | | |
| SR | 1, 45 | 0.07 |
| Drought treatment | 1, 54 | **34.95***** |
| SR × drought | 1, 54 | 1.37 |
| Soil moisture | 1, 54 | <0.01 |
| (b) **Bacterial richness** | | |
| SR | 1, 45 | **11.36**** |
| Drought treatment | 1, 54 | **143.30***** |
| SR × drought | 1, 54 | **8.65**** |
| Soil moisture | 1, 54 | **80.12***** |
| (c) **AMF richness** | | |
| SR | 1, 45 | **29.18***** |
| Drought treatment | 1, 54 | **55.41***** |
| SR × drought | 1, 54 | **5.88*** |
| Soil moisture | 1, 54 | 1.33 |
| (d) **Fungal pathogen richness** | | |
| SR | 1, 45 | 1.97 |
| Drought treatment | 1, 54 | <0.01 |
| SR × drought | 1, 54 | **4.85*** |
| Soil moisture | 1, 54 | 0.13 |

complementarity was greatest in ambient communities with previously droughted soil (drought legacy), whereas the effects of plant species richness on complementarity were lowest in ambient plant communities with ambient soil inoculum (Fig. 3C).

SEs were significantly affected by drought, but the magnitude and direction of drought effects varied depending on both drought legacy and plant species richness (plant species richness × drought legacy × ongoing drought interaction; Table 2). SEs showed a positive relationship with plant species richness for ambient communities with ambient soil inoculum (table S3). In contrast, SEs became more negative as plant species richness increased for communities with ongoing drought and/or drought soil legacy (table S3 and Fig. 3D). The magnitude of negative SEs associated with high species richness was particularly pronounced for ambient communities growing with soil previously subjected to drought (Fig. 3D).

## DISCUSSION

Mounting evidence indicates that biodiversity loss and drought are interconnected and that impacts of these drivers cannot be understood in isolation, but to date, combined effects of biodiversity loss and drought on ecosystem function remain poorly explored (*40*). Our two-phase experiment, examining direct and indirect (legacy) effects of drought on plant diversity-productivity relationships, resulted in three key findings: (i) drought modifies the relationships between plant species richness and richness of soil microbial groups (bacteria, AMF, and fungal pathogens), (ii) drought soil legacy increases the magnitude of net BEs but not the response patterns of net BEs to increasing plant species richness, and (iii) drought soil legacy interacts with subsequent drought, modifying the relative contribution of complementarity and selection to net diversity effects. Overall, levels of soil fungi and AMF richness were high, although similar fungal and AMF OTU richness has been reported in some other grassland studies (*41*–*43*).

In line with our first hypothesis, we found that both drought and plant diversity modified the structure of soil fungal and bacterial communities, confirming results from previous studies in herbaceous ecosystems (*11*, *17*, *44*). Soil microorganisms play a crucial role for plant performance and community dynamics via mutualists and pathogens, as well as via decomposition of organic matter and nutrient cycling (*42*, *45*). Within grassland ecosystems, increasing plant diversity has been shown to have a positive effect on microbial diversity in some studies (*16*, *42*) but a negative or no effect in others (*11*, *17*, *46*, *47*), suggesting confounding abiotic and biotic effects (*48*). In the present study, we found that linkages between plant species richness and fungal richness varied depending on both fungal guild and drought treatment. Under ambient conditions, plant species richness increased richness of mutualists but decreased richness of pathogens, in agreement with inoculation experiments (*49*) and data on AMF in herbaceous plant communities (*50*, *51*). Plant species richness also showed a negative relationship with total bacterial diversity under ambient conditions, with decreases in bacterial diversity at least partly driven by concurrent decreases in soil moisture. Under drought conditions, however, the strength of plant species richness effects on AMF was reduced, and the negative relationships between plant species richness and pathogens/bacteria disappeared. Drought has previously been shown to weaken the link between plant and bacterial carbon turnover (*52*), and decreased carbon allocation from plants to microorganisms under drought conditions could at least partly explain the weaker effects of plant species richness on microbial groups observed in our drought treatment.

Soil water stress may promote selection for drought-adapted microbial taxa (*28*, *53*), and recent work on linkages between AMF diversity and genetic diversity in barley has suggested that drought may generate unexpected plant-soil biotic interactions (*54*). Our study demonstrates that drought mediates linkages between multiple fungal guilds and plant species richness in herbaceous communities, with implications for both soil legacy effects and plant diversity-productivity relationships in a changing environment. It is notable that positive plant diversity-productivity relationships in phase I were only apparent under ambient conditions (fig. S3), providing added support to the idea that soil fungal pathogens are an important driver of plant diversity-productivity relationships (*18*). Given that positive plant diversity-productivity relationships in phase I also occurred at the expense of bacterial richness, our results could indicate a role for soil bacterial pathogens in plant diversity-productivity relationships; decreases in the diversity of bacterial pathogens with increasing plant species richness could promote plant productivity. Resolving microbial and, in particular, bacterial diversity into functional groups is a challenge (*55*), and links between soil bacterial pathogens and plant diversity-productivity relationships remain to be explicitly demonstrated.

**Table 2. Results of linear mixed-model analysis for plant data in phase II.** The interactive effects of plant species richness, drought legacy (DL; past drought), and present drought (PD; phase II) treatments on aboveground biomass, net BE, CEs, and SEs in phase II. Degrees of freedom (df) and *F* values are given. Bold types are significant effects (**P* < 0.05, ***P* < 0.01, and ****P* < 0.001).

| | Aboveground biomass | | BE | | CE | | SE | |
|---|---|---|---|---|---|---|---|---|
| | **df** | ***F*** | ***df*** | ***F*** | **df** | ***F*** | **df** | ***F*** |
| Plant SR | 1, 45 | **8.38**** | 1, 33 | **11.02**** | 1, 33 | **37.13***** | 1, 33 | 0.37 |
| DL | 1, 67 | 0.30 | 1, 57 | **57.48***** | 1, 57 | **76.85***** | 1, 57 | **16.11***** |
| PD | 1, 112 | **305.97***** | 1, 90 | **13.48***** | 1, 90 | **5.80*** | 1, 90 | 0.34 |
| SR × DL | 1, 67 | **7.49**** | 1, 57 | 0.19 | 1, 57 | **19.06***** | 1, 57 | **24.75***** |
| SR × PD | 1, 112 | **11.39**** | 1, 90 | **8.38**** | 1, 90 | **3.96*** | 1, 90 | 0.12 |
| DL × PD | 1, 112 | 0.09 | 1, 90 | **31.19***** | 1, 90 | **59.33***** | 1, 90 | **16.23***** |
| SR × DL × PD | 1, 112 | 3.37 | 1, 90 | 0.66 | 1, 90 | **26.5***** | 1, 90 | **30.08***** |

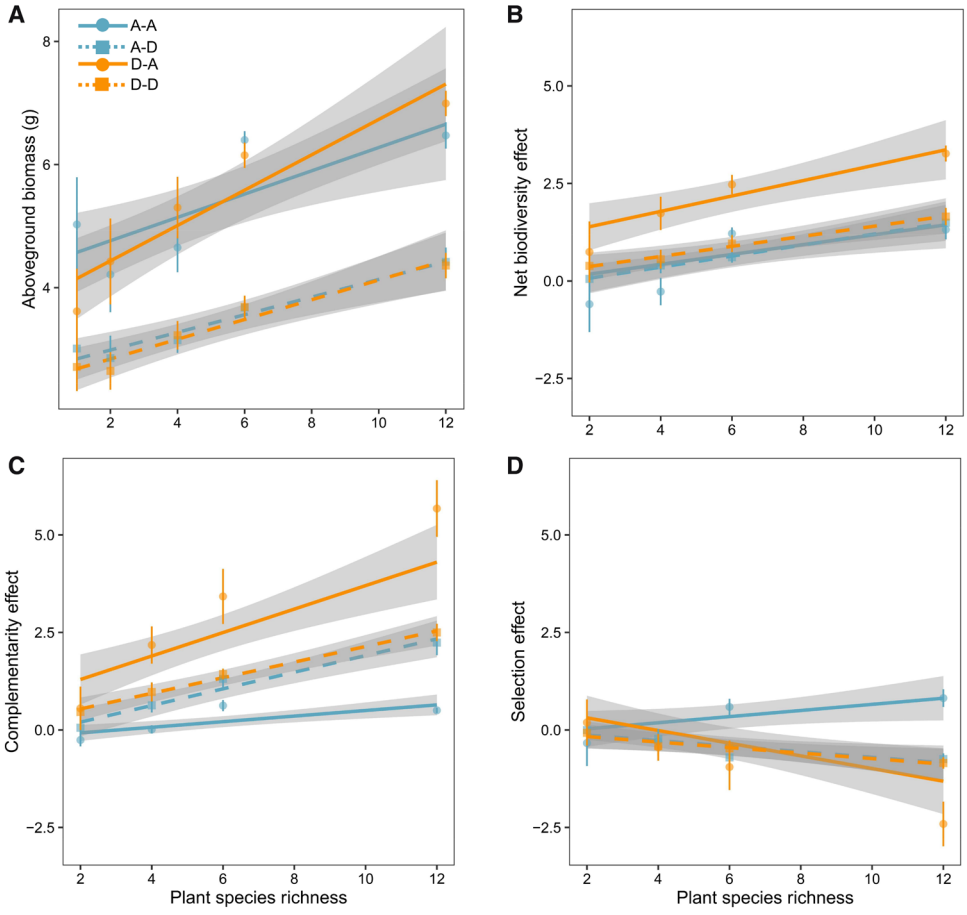


**Fig. 3. Aboveground biomass and BEs in phase II.** Effects of past (drought legacy) and present (phase II) drought treatments on (**A**) aboveground biomass, (**B**) net BEs, (**C**) CEs, and (**D**) SEs at different species richness levels in experimental mesocosms. Treatments are given by the following: A-A, ambient-ambient; D-A, drought-ambient; A-D, ambient-drought; and D-D, drought-drought. Lines indicate significant relationships between plant species richness and plant variables at the mesocosm level (*P* < 0.05). Shaded areas indicate 95% confidential intervals of fitted lines. Means and SE are given for all treatment combinations.

We found that net BEs were more positive in plant communities with previously droughted soil, supporting our second hypothesis that drought soil legacy influences BEs on plant productivity in species mixtures. Drought history and drought soil legacy can have lasting effects on plant community structure and ecosystem processes (*32*, *56*, *57*), but the effects of drought soil legacy on plant diversity-productivity relationships are poorly understood. In our study, positive effects of drought legacy on diversity effects were driven by a higher differential between production of monocultures and mixed communities growing in ambient conditions on previously droughted soil, but drought soil legacy alone did not alter the response patterns of net BEs to plant species richness. Inspection of

plant communities in droughted conditions also revealed a tendency for reduced monoculture performance in previously droughted soil.

Reduced growth of monocultures in previously droughted soil in the present study is consistent with drought legacy effects on conspecific plant-soil feedback documented for herbaceous monocultures elsewhere (*57*) and may reflect changes in AMF abundance and mycorrhizal growth responses due to drought legacy effects (*58*). The large drought-induced reductions in bacterial richness observed for monocultures in phase I could also contribute to the lower performance of monocultures in previously droughted soil observed in phase II. We do not know the precise mechanism explaining the improved performance of high-diversity mixtures on soil with a history of drought subsequently exposed to ambient conditions but propose that it could result from improved nutrient availability via drought legacy effects on the soil microbial community. Drought-tolerant microbial communities in high-diversity plant communities may have promoted the performance of plant communities in ambient conditions through shifts in plant/microbial competition for N (*59*, *60*) or by enhancing organic matter decomposition and C and N fluxes (*61*, *62*). Quantitative measurements of microbial group abundance and related soil processes as well as soil physicochemical properties would provide additional insights into the mechanistic underpinnings of drought soil legacy effects.

Unlike drought legacy, phase II drought conditions interacted with species richness on plant productivity and net BEs; diversity-productivity relationships [biodiversity and ecosystem functioning (BEF)] were stronger in ambient conditions compared with droughted-plant communities. The weaker BEF relationship in response to drought observed here agrees with some results from experimental grassland communities and gradients of species richness (*63*, *64*), and suggests incomplete buffering and/or incomplete compensatory effects of plant diversity under drought (*65*). Our results for net diversity effects imply that current drought events and plant-driven mechanisms have a greater impact on linkages between plant species richness and ecosystem functioning than do drought soil legacy effects. Nevertheless, the additive effects of drought soil legacy on net diversity effects support recent assertions that knowledge of plant-soil interactions can improve predictions of plant community growth and diversity-productivity relationships (*14*, *66*).

Similar overall response patterns of net BEs may mask different combinations of SEs and CEs (*64*). In the present study, we found that the partitioning of net BEs into CEs and SEs varied depending on both drought soil legacy and ongoing drought conditions. These results provide partial support for our third hypothesis; subsequent drought alters the effects of drought soil legacy on the drivers of BEF relationships, rather than on the form of the overall BEF relationship per se. Our study contributes to the growing number of studies, which suggest that regulators of plant diversity-productivity relationships are context dependent (*64*, *67*). Observed responses of CEs and SEs to ongoing drought were not consistent with the stress gradient hypothesis (i.e., increased ratio of facilitative to competitive interactions under more stressful conditions); previous studies along drought gradients have also provided mixed support for this idea (*68*).

Under subsequent well-watered conditions (i.e., ambient conditions in phase II), drought soil legacy generated divergent relationships between selection and species richness and induced significant increases in the strength of CEs. Changes in CEs may be driven by shifts in the relative contribution of processes underlying complementarity (facilitation via microclimate amelioration versus resource partitioning or altered soil-plant feedbacks), although such phenomena remain poorly characterized (*68*). Drought-induced changes in microbial group richness observed in the first phase of our experiment do not appear to provide a biologically intuitive explanation for the shifts in selection and complementarity recorded in this treatment combination. Instead, these drought legacy effects may reflect drought-induced changes in microbial functioning, which affect carbon turnover and loss, with implications for nutrient cycling and plant resource partitioning (*56*, *69*).

In the absence of soil microbial diversity measurements in the second phase of our experiment, we cannot rule out the possibility that drought history interacted with present drought treatments on the soil microbial community. However, recent work indicates that multiple recurrent drought events strengthen shifts in the composition of microbial communities (*32*). Given that CEs are usually considered to promote ecosystem stability whereas SEs are destabilizing (*70*), our findings also suggest that soil-mediated drought legacy effects may have broader-reaching implications for ecosystem functioning. Of course, in real-world systems, drought will induce legacy effects in both the soil and plant communities, and the interplay between drought soil legacy and drought plant legacy will determine plant-soil interactions and ecosystem processes in the more or less long term (*71*). In theory, variation in the timing, duration, and intensity of drought may also mediate the strength and persistence of drought soil legacy effects on plant communities (*72*, *73*), and future studies are required to determine whether effects of soil legacy from pulse and press droughts differ.

Our experimental design allowed us to examine how the effects of chronic water reduction on the soil microbial community can affect plant diversity-productivity relationships and how subsequent water availability mediates such soil drought legacy effects. Our results demonstrate drought-induced changes in the richness of soil microbial groups and suggest that drought-induced changes in the linkages between plant species richness and soil pathogens may drive concomitant changes in the relationship between plant species richness and aboveground productivity. Effects of drought soil legacy on aboveground productivity and diversity effects were expressed most strongly under subsequent nonlimiting water availability, with implications for ecosystem stability under recurrent drought. Moreover, we found that positive plant diversity-productivity relationships were maintained by shifting patterns of CEs and SEs in the presence or absence of drought soil legacy effects, providing evidence for soil microbial mediation of BEF relationships in a changing environment. Exploring the relative contribution of above- and belowground legacy effects on BEF relationships will be a further necessary step toward mechanistic understanding of the dynamic responses of ecosystems to climate change.

## MATERIALS AND METHODS

### Experimental design

We set up a two-phase experiment with a subtropical old field grassland model system to investigate how the plant community and drought influence soil legacy effects and plant-soil feedbacks. In line with previous soil legacy studies [e.g., (*56*, *71*)], soil microbial communities were conditioned by plant communities in phase I, and the soil microbial legacy was tested using newly established plant communities in phase II (Fig. 1). In each phase, five plant

diversity (species richness) treatments were factorially crossed with two rainfall treatments, i.e., ambient and reduced rainfall conditions (Fig. 1). Diversity treatments were constructed from a pool of 12 herbaceous species that co-occur in natural old fields in southern China, including three grasses (*Dactyloctenium aegyptium*, *Digitaria radicosa*, and *Isachne repens*), eight nonlegume forbs (*Celosia argentea*, *Amaranthus viridis*, *Achyranthes aspera*, *Emilia sonchifolia*, *Ageratum conyzoides*, *Eclipta prostrata*, *Ludwigia hyssopifolia*, and *Capsella bursa-pastoris*), and a legume (*Mimosa pudica*). Seeds for all species were collected from January to February 2019 in old fields in Fengkai County, China. The climate is subtropical monsoon with a mean annual temperature of 19.6°C and a mean annual precipitation of 1532.8 mm. The field site is characterized by high interannual variation in monthly rainfall, with cooler, drier months from November to February. Soil was collected in an old field in Fengkai County, China (23.51°N, 111.82°E) in March 2019. The soil is characterized as lateritic red soil (3.53% C and 0.18% N) and has a $pH_{H2O}$ of 5.28. Topsoil (15-cm depth) was collected, sieved to remove large particles and organic debris (1-cm mesh), and homogenized before the experiment.

### Phase I: Conditioning soil microbes under different plant communities and rainfall conditions

We performed the soil conditioning phase of the experiment at the Heishiding nature reserve in Fengkai (23.46°N, 111.90°E) using a total of 120 mesocosms (five plant diversity levels × two drought treatments × 12 replicates). Mesocosms were created with single plant species (monocultures) or with mixtures of 2, 4, 6, and 12 species. There were 12 monocultures (one for each plant species) and 12 combinations for each of the two, four, or six species mixtures; species composition was determined using 12 separate random draws from the complete species pool (table S4). The 12-species mixture had constant species composition (12 replicates). Each mesocosm consisted of 12 individual plants, and all plant species had equal density in the mixtures.

Seeds were first surface sterilized by rinsing in 75% ethanol for 2 min and then germinated in sterilized vermiculite. On 28 April 2019, 12 two-week-old seedlings were transplanted into each mesocosm (24 cm in diameter and 19 cm in height), and plants that failed during the first week were replanted. All mesocosms were regularly watered for the first 6 weeks to avoid drought stress. On 15 June, mesocosms in each diversity treatment were assigned to either a drought or ambient watering treatment. Half of the mesocosms continued to be watered regularly (i.e., ambient treatment, 600 ml applied slowly every 4 days), and the remainder received one-third of the water applied in the ambient treatment (i.e., drought treatment, 200 ml provided every 4 days). The drought treatments were performed for 12 weeks. Mesocosm location was randomized monthly to avoid position effects.

Volumetric soil moisture content at 5-cm depth was monitored in all mesocosms by a portable GS-3 soil moisture probe (Decagon Devices Inc., WA) monthly. Over the course of phase I, the reduced watering regime in the drought treatment caused significantly different soil moisture contents between the "drought" and "ambient" mesocosms (table S5 and fig. S4A). Plants were grown for 18 weeks. At the start of September 2019, when all species were in their reproductive stage, all plants were removed from the experimental mesocosms. A subsample of 20-g soil was removed in each mesocosm and stored at −80°C for soil microbial DNA sequencing. For the 12-species mixtures, we randomly selected five mesocosms for microbial analyses in each drought treatment. A subsample of 30% (by volume) of the soil from each mesocosm was stored at −20°C as soil inoculum before phase II.

### Phase II: Plant community responses to soil microbial legacy and a subsequent drought

Bulk soil was collected at the Heishiding nature reserve and sterilized by γ-irradiation at 25 kGy. Gamma irradiation has been shown to effectively sterilize soil with minimal impacts on other soil properties (*74*). Mesocosms (21 cm in diameter and 17 cm in height) were filled with 90% (by volume) sterilized bulk soil and 10% conditioned soil. This approach allowed us to minimize the impacts of differences in soil abiotic properties on plant growth and explore the role of soil microorganisms (*75*). Two-week-old seedlings were transplanted into mesocosms on 13 May 2020 using the same diversity treatments and plant community composition as in in phase I, i.e., all plant communities were grown in "own" soil. Plants that failed during the first week were replaced. During the first 3 weeks, all mesocosms were watered regularly to promote seedling establishment.

On 2 June 2020, drought and ambient watering treatments were established (same watering regime as in phase I), and the watering regime in phase I was crossed with the watering regime in phase II. Half the mesocosms with soil from the ambient treatment in phase I were assigned to the ambient watering treatment in phase II, whereas the remainder were assigned to drought (Fig. 1). Similarly, half of the mesocosms with soil from the drought treatment in phase I were assigned to the ambient watering treatment in phase II, whereas the remainder were assigned to drought (Fig. 1). This generated four drought treatments in phase II, namely, ambient treatments in both phases (A-A), drought treatments in both phases (D-D), ambient in phase I + drought in phase II (A-D), and drought in phase I + ambient in phase II (D-A). In total, there were 240 mesocosms (five diversity levels × four drought treatments × 12 replicates). During the phase II period of the experiment, volumetric soil moisture content at 5-cm depth was monitored every 2 weeks using a GS-3 soil moisture probe (Decagon Devices Inc. WA). Over the period of phase II, the drought treatments caused significantly different soil moisture contents among mesocosms (table S5 and fig. S4B).

Mesocosm locations were randomized every 2 weeks, and plants were left to grow until early August 2020 when all species were in their reproductive stages. At final harvest, aboveground biomass was sorted by species in each mesocosm. Belowground biomass was harvested at the mesocosm level because the root systems could not be sorted out to individual species. Plant materials were oven dried at 60°C for 48 hours and weighted to determine biomass.

### Partitioning BEs

The net BE of each multispecies mesocosm was determined as the difference between the observed yield and its expected yield, based on species' proportional abundances multiplied by their biomass in monoculture (*76*). The CE was calculated as the deviation in average relative yield in the mixture, i.e., $N\,\overline{\Delta RY_i}\,\overline{M_i}$, where $i$ is the component species in the mixture, $N$ is the number of component species, $\Delta RY_i$ is the deviation from the expected relative yield of species $i$ in the mixture, and $M_i$ is the aboveground biomass of species $i$ in its monoculture. The SE was calculated as the covariance between the monoculture yield of species and the deviation in relative yield in

the mixture, i.e., $N$ cov ($\Delta RY_i$, $M_i$). These various effects can be related by an additive partition: BE = CE + SE. Positive CE indicates that species yields in a mixture are on average higher than expected values of the component species (the weighted average monoculture yields), and positive SE indicates that species with higher-than-average monoculture yields dominate the mixture.

### Soil microbial DNA sequencing

DNA was extracted from soil samples using the MOBIO PowerSoil DNA Isolation Kit (MOBIO Laboratories, Carlsbad, CA, USA). DNA quality, concentration, and purification were checked on 1% agarose gel electrophoresis and NanoDrop One UV-Vis Spectrophotometer (Thermo Fisher Scientific, Wilmington, USA). The V4 hypervariable region of the 16*S* ribosomal RNA (rRNA) for bacteria and second internal transcribed spacer (ITS2) region of the rRNA operon for fungi were amplified using primer combinations 515F/806R (*77*) and ITS3F/ITS4R (*78*), respectively. Polymerase chain reaction (PCR) was conducted using Bio-Rad S1000 (Bio-Rad Laboratory, CA), with a 50-μl mixture containing 25 μl of 2× Premix Taq, 10 mM of each primer, 60 ng of DNA, and nuclease-free water. The PCR amplification had an initial denaturation step at 94°C for 5 min, followed by 30 cycles of denaturation at 94°C (30 s), annealing at 52°C (30 s), extension at 72°C (30 s), and a final extension at 72°C (10 min). PCR products were extracted and purified using E.Z.N.A. Gel Extraction Kit (Omega, USA). Amplicon libraries were prepared using NEBNext Ultra DNA Library Prep Kit for Illumina (New England Biolabs, USA) following the manufacturer's protocol, and PE250 sequencing was performed on a NovaSeq 6000 Sequencing System (Illumina, San Diego, USA).

OTUs were categorized at 97% sequence similarity using the UPARSE package, and taxonomic assignment for bacteria and fungi was performed using the Silva and Unite databases, respectively. Singleton sequences, chloroplast or mitochondria (16*S* amplicon) sequences, and sequences that could not be assigned to the kingdom level were removed from the final OTU table. Each sample was rarefied to the same number of reads as the smallest sample (52,190 and 29,940 reads for bacteria and fungi, respectively) in subsequent analyses. Fungal OTUs were assigned to AMF, plant pathogens and saprotrophs using FUNGuild (*79*) and further checked based on literature.

### Statistical analyses

Effects of plant species richness and drought on soil microbial community structure in phase I were examined using distance-based redundancy analysis (db-RDA). Fungal and bacterial OTU compositional data were Hellinger transformed, and Bray-Curtis dissimilarity was used to measure community distances. Plant species richness and soil moisture content (the average values of mesocosms over the phase I, natural log transformed) were used as explanatory variables. The db-RDAs were performed in R using the "capscale" function in package "vegan." In addition, we performed general linear mixed-effects models using the "lme" function in R package "nlme" to evaluate interactive effects of plant species richness and drought on soil fungal richness, bacterial richness, AMF richness, fungal pathogen richness, and saprotroph richness. Plant species richness (natural log transformed) and drought treatments in phase I were included as fixed factors, and plant community composition was used as a random factor. Average soil moisture content over phase I was included as a covariate. Separate regression analyses were then performed to test whether there were linear relationships between plant species richness and these response variables in each drought treatment.

To test for interactive effects of plant species richness, drought legacy from phase I, and ongoing drought treatments on plant variables in phase II (i.e., plant community biomass, net BEs, CEs, and SEs), we performed general linear mixed-effects models. Plant species richness (natural log transformed) and drought treatments from phases I and II were included as fixed factors, whereas plant community composition and mesocosms that provided soil inoculums were used as random factors. Between-group comparisons were performed using the Tukey's post hoc tests. All analyses were conducted in R (*80*).

## SUPPLEMENTARY MATERIALS

Supplementary material for this article is available at https://science.org/doi/10.1126/sciadv.abn3368

## REFERENCES AND NOTES


1. M. Loreau, S. Naeem, P. Inchausti, J. Bengtsson, J. P. Grime, A. Hector, D. U. Hooper, M. A. Huston, D. Raffaelli, B. Schmid, D. Tilman, D. A. Wardle, Biodiversity and ecosystem functioning: Current knowledge and future challenges. *Science* **294**, 804–808 (2001).
2. D. U. Hooper, F. S. Chapin, J. J. Ewel, A. Hector, P. Inchausti, S. Lavorel, J. H. Lawton, D. M. Lodge, M. Loreau, S. Naeem, B. Schmid, H. Setälä, A. J. Symstad, J. Vandermeer, D. A. Wardle, Effects of biodiversity on ecosystem functioning: A consensus of current knowledge. *Ecol. Monogr.* **75**, 3–35 (2005).
3. D. Tilman, F. Isbell, J. M. Cowles, Biodiversity and ecosystem functioning. *Annu. Rev. Ecol. Evol. Syst.* **45**, 471–493 (2014).
4. F. Isbell, D. Craven, J. Connolly, M. Loreau, B. Schmid, C. Beierkuhnlein, T. M. Bezemer, C. Bonin, H. Bruelheide, E. de Luca, A. Ebeling, J. N. Griffin, Q. Guo, Y. Hautier, A. Hector, A. Jentsch, J. Kreyling, V. Lanta, P. Manning, S. T. Meyer, A. S. Mori, S. Naeem, P. A. Niklaus, H. W. Polley, P. B. Reich, C. Roscher, E. W. Seabloom, M. D. Smith, M. P. Thakur, D. Tilman, B. F. Tracy, W. H. van der Putten, J. van Ruijven, A. Weigelt, W. W. Weisser, B. Wilsey, N. Eisenhauer, Biodiversity increases the resistance of ecosystem productivity to climate extremes. *Nature* **526**, 574–577 (2015).
5. H. J. De Boeck, J. M. G. Bloor, J. Kreyling, J. C. G. Ransijn, I. Nijs, A. Jentsch, M. Zeiter, Patterns and drivers of biodiversity-stability relationships under climate extremes. *J. Ecol.* **106**, 890–902 (2018).
6. D. Craven, N. Eisenhauer, W. D. Pearse, Y. Hautier, F. Isbell, C. Roscher, M. Bahn, C. Beierkuhnlein, G. Bönisch, N. Buchmann, C. Byun, J. A. Catford, B. E. L. Cerabolini, J. H. C. Cornelissen, J. M. Craine, E. De Luca, A. Ebeling, J. N. Griffin, A. Hector, J. Hines, A. Jentsch, J. Kattge, J. Kreyling, V. Lanta, N. Lemoine, S. T. Meyer, V. Minden, V. Onipchenko, H. W. Polley, P. B. Reich, J. van Ruijven, B. Schamp, M. D. Smith, N. A. Soudzilovskaia, D. Tilman, A. Weigelt, B. Wilsey, P. Manning, Multiple facets of biodiversity drive the diversity–stability relationship. *Nat. Ecol. Evol.* **2**, 1579–1587 (2018).
7. A. S. Mori, L. E. Dee, A. Gonzalez, H. Ohashi, J. Cowles, A. J. Wright, M. Loreau, Y. Hautier, T. Newbold, P. B. Reich, T. Matsui, W. Takeuchi, K. Okada, R. Seidl, F. Isbell, Biodiversity–productivity relationships are key to nature-based climate solutions. *Nat. Clim. Chang.* **11**, 543–550 (2021).
8. J. van Ruijven, F. Berendse, Diversity-productivity relationships: Initial effects, long-term patterns, and underlying mechanisms. *Proc. Natl. Acad. Sci. U.S.A.* **102**, 695–700 (2005).
9. D. F. B. Flynn, N. Mirotchnick, M. Jain, M. I. Palmer, S. Naeem, Functional and phylogenetic diversity as predictors of biodiversity–ecosystem-function relationships. *Ecology* **92**, 1573–1581 (2011).
10. X. Morin, L. Fahse, M. Scherer-Lorenzen, H. Bugmann, Tree species richness promotes productivity in temperate forests through strong complementarity between species. *Ecol. Lett.* **14**, 1211–1219 (2011).
11. L. Mommer, T. E. A. Cotton, J. M. Raaijmakers, A. J. Termorshuizen, J. van Ruijven, M. Hendriks, S. Q. van Rijssel, J. E. van de Mortel, J. W. van der Paauw, E. G. W. M. Schijlen, A. E. Smit-Tiekstra, F. Berendse, H. de Kroon, A. J. Dumbrell, Lost in diversity: The interactions between soil-borne fungi, biodiversity and plant productivity. *New Phytol.* **218**, 542–553 (2018).
12. M. Liang, X. Liu, I. M. Parker, D. Johnson, Y. Zheng, S. Luo, G. S. Gilbert, S. Yu, Soil microbes drive phylogenetic diversity-productivity relationships in a subtropical forest. *Sci. Adv.* **5**, eaax5088 (2019).
13. J. van Ruijven, E. Ampt, D. Francioli, L. Mommer, Do soil-borne fungal pathogens mediate plant diversity–productivity relationships? Evidence and future opportunities. *J. Ecol.* **108**, 1810–1821 (2020).

14. M. P. Thakur, W. H. van der Putten, R. A. Wilschut, G. F. C. Veen, P. Kardol, J. van Ruijven, E. Allan, C. Roscher, M. van Kleunen, T. M. Bezemer, Plant–soil feedbacks and temporal dynamics of plant diversity–productivity relationships. *Trends Ecol. Evol.* **36**, 651–661 (2021).
15. G. A. Kowalchuk, D. S. Buma, W. de Boer, P. G. L. Klinkhamer, J. A. van Veen, Effects of above-ground plant species composition and diversity on the diversity of soil-borne microorganisms. *Antonie Van Leeuwenhoek* **81**, 509 (2002).
16. M. Lange, N. Eisenhauer, C. A. Sierra, H. Bessler, C. Engels, R. I. Griffiths, P. G. Mellado-Vázquez, A. A. Malik, J. Roy, S. Scheu, S. Steinbeiss, B. C. Thomson, S. E. Trumbore, G. Gleixner, Plant diversity increases soil microbial activity and soil carbon storage. *Nat. Commun.* **6**, 6707 (2015).
17. S. Dassen, R. Cortois, H. Martens, M. de Hollander, G. A. Kowalchuk, W. H. van der Putten, G. B. D. Deyn, Differential responses of soil bacteria, fungi, archaea and protists to plant species richness and plant functional group identity. *Mol. Ecol.* **26**, 4085–4098 (2017).
18. J. L. Maron, M. Marler, J. N. Klironomos, C. C. Cleveland, Soil fungal pathogens and the relationship between plant diversity and productivity. *Ecol. Lett.* **14**, 36–41 (2011).
19. S. A. Schnitzer, J. N. Klironomos, J. HilleRisLambers, L. L. Kinkel, P. B. Reich, K. Xiao, M. C. Rillig, B. A. Sikes, R. M. Callaway, S. A. Mangan, E. H. van Nes, M. Scheffer, Soil microbes drive the classic plant diversity–productivity pattern. *Ecology* **92**, 296–303 (2011).
20. K. M. Vogelsang, H. L. Reynolds, J. D. Bever, Mycorrhizal fungal identity and richness determine the diversity and productivity of a tallgrass prairie system. *New Phytol.* **172**, 554–562 (2006).
21. K. O. Reinhart, B. L. Anacker, More closely related plants have more distinct mycorrhizal communities. *AoB Plants* **6**, plu051 (2014).
22. H. Maherali, J. N. Klironomos, Influence of phylogeny on fungal community assembly and ecosystem functioning. *Science* **316**, 1746–1748 (2007).
23. G. Wang, P. Schultz, A. Tipton, J. Zhang, F. Zhang, J. D. Bever, Soil microbiome mediates positive plant diversity-productivity relationships in late successional grassland species. *Ecol. Lett.* **22**, 1221–1232 (2019).
24. C. Wagg, J. Jansa, B. Schmid, M. G. A. van der Heijden, Belowground biodiversity effects of plant symbionts support aboveground productivity. *Ecol. Lett.* **14**, 1001–1009 (2011).
25. F. Walder, M. G. A. van der Heijden, Regulation of resource exchange in the arbuscular mycorrhizal symbiosis. *Nat. Plants* **1**, 15159 (2015).
26. J. K. Jansson, K. S. Hofmockel, Soil microbiomes and climate change. *Nat. Rev. Microbiol.* **18**, 35–46 (2020).
27. Z. Zhou, C. Wang, Y. Luo, Meta-analysis of the impacts of global change factors on soil microbial diversity and functionality. *Nat. Commun.* **11**, 3072 (2020).
28. A. Meisner, S. Jacquiod, B. L. Snoek, F. C. ten Hooven, W. H. van der Putten, Drought legacy effects on the composition of soil fungal and prokaryote communities. *Front. Microbiol.* **9**, 294 (2018).
29. A. Meisner, B. L. Snoek, J. Nesme, E. Dent, S. Jacquiod, A. T. Classen, A. Priemé, Soil microbial legacies differ following drying-rewetting and freezing-thawing cycles. *ISME J.* **15**, 1207–1221 (2021).
30. F. T. de Vries, R. I. Griffiths, M. Bailey, H. Craig, M. Girlanda, H. S. Gweon, S. Hallin, A. Kaisermann, A. M. Keith, M. Kretzschmar, P. Lemanceau, E. Lumini, K. E. Mason, A. Oliver, N. Ostle, J. I. Prosser, C. Thion, B. Thomson, R. D. Bardgett, Soil bacterial networks are less stable under drought than fungal networks. *Nat. Commun.* **9**, 3033 (2018).
31. J. P. Schimel, Life in dry soils: Effects of drought on soil microbial communities and processes. *Annu. Rev. Ecol. Evol. Syst.* **49**, 409–432 (2018).
32. A. Canarini, H. Schmidt, L. Fuchslueger, V. Martin, C. W. Herbold, D. Zezula, P. Gündler, R. Hasibeder, M. Jecmenica, M. Bahn, A. Richter, Ecological memory of recurrent drought modifies soil processes via changes in soil microbial community. *Nat. Commun.* **12**, 5308 (2021).
33. A. K. Knapp, Rainfall variability, carbon cycling, and plant species diversity in a mesic grassland. *Science* **298**, 2202–2205 (2002).
34. R. J. Griffin-Nolan, D. M. Blumenthal, S. L. Collins, T. E. Farkas, A. M. Hoffman, K. E. Mueller, T. W. Ocheltree, M. D. Smith, K. D. Whitney, A. K. Knapp, Shifts in plant functional composition following long-term drought in grasslands. *J. Ecol.* **107**, 2133–2148 (2019).
35. L. W. Ploughe, E. M. Jacobs, G. S. Frank, S. M. Greenler, M. D. Smith, J. S. Dukes, Community response to extreme drought (CRED): A framework for drought-induced shifts in plant–plant interactions. *New Phytol.* **222**, 52–69 (2019).
36. J. D. Bever, Feeback between plants and their soil communities in an old field community. *Ecology* **75**, 1965–1977 (1994).
37. W. H. van der Putten, R. D. Bardgett, J. D. Bever, T. M. Bezemer, B. B. Casper, T. Fukami, P. Kardol, J. N. Klironomos, A. Kulmatiski, J. A. Schweitzer, K. N. Suding, T. F. J. Van de Voorde, D. A. Wardle, Plant-soil feedbacks: The past, the present and future challenges. *J. Ecol.* **101**, 265–276 (2013).
38. F. I. Pugnaire, J. A. Morillo, J. Peñuelas, P. B. Reich, R. D. Bardgett, A. Gaxiola, D. A. Wardle, W. H. van der Putten, Climate change effects on plant-soil feedbacks and consequences for biodiversity and functioning of terrestrial ecosystems. *Sci. Adv.* **5**, eaaz1834 (2019).
39. B. Wang, S. D. Allison, Drought legacies mediated by trait trade-offs in soil microbiomes. *Ecosphere* **12**, e03562 (2021).
40. A. P. F. Pires, D. S. Srivastava, N. A. C. Marino, A. A. M. MacDonald, M. P. Figueiredo-Barros, V. F. Farjalla, Interactive effects of climate change and biodiversity loss on ecosystem functioning. *Ecology* **99**, 1203–1213 (2018).
41. A. Fox, F. Widmer, A. Barreiro, M. Jongen, M. Musyoki, Â. Vieira, J. Zimmermann, C. Cruz, L.-M. Dimitrova-Mårtensson, F. Rasche, L. Silva, A. Lüscher, Small-scale agricultural grassland management can affect soil fungal community structure as much as continental scale geographic patterns. *FEMS Microbiol. Ecol.* **97**, fiab148 (2021).
42. T. Yang, J. M. Adams, Y. Shi, J. He, X. Jing, L. Chen, L. Tedersoo, H. Chu, Soil fungal diversity in natural grasslands of the Tibetan Plateau: Associations with plant diversity and productivity. *New Phytol.* **215**, 756–765 (2017).
43. M. A. Muneer, M. W. K. Tarin, X. Chen, M. S. Afridi, A. Iqbal, M. Z. Munir, C. Zheng, J. Zhang, B. Ji, Differential response of mycorrhizal fungi linked with two dominant plant species of temperate grassland under varying levels of N-addition. *Appl. Soil Ecol.* **170**, 104272 (2022).
44. R. Ochoa-Hueso, S. L. Collins, M. Delgado-Baquerizo, K. Hamonts, W. T. Pockman, R. L. Sinsabaugh, M. D. Smith, A. K. Knapp, S. A. Power, Drought consistently alters the composition of soil fungal and bacterial communities in grasslands from two continents. *Glob. Chang. Biol.* **24**, 2818–2827 (2018).
45. M. G. A. van der Heijden, R. D. Bardgett, N. M. V. Straalen, The unseen majority: Soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. *Ecol. Lett.* **11**, 296–310 (2008).
46. S. M. Prober, J. W. Leff, S. T. Bates, E. T. Borer, J. Firn, W. S. Harpole, E. M. Lind, E. W. Seabloom, P. B. Adler, J. D. Bakker, E. E. Cleland, N. M. DeCrappeo, E. DeLorenze, N. Hagenah, Y. Hautier, K. S. Hofmockel, K. P. Kirkman, J. M. H. Knops, K. J. L. Pierre, A. S. MacDougall, R. L. McCulley, C. E. Mitchell, A. C. Risch, M. Schuetz, C. J. Stevens, R. J. Williams, N. Fierer, Plant diversity predicts beta but not alpha diversity of soil microbes across grasslands worldwide. *Ecol. Lett.* **18**, 85–95 (2015).
47. D. C. Schlatter, M. G. Bakker, J. M. Bradeen, L. L. Kinkel, Plant community richness and microbial interactions structure bacterial communities in soil. *Ecology* **96**, 134–142 (2015).
48. L. Liu, K. Zhu, N. Wurzburger, J. Zhang, Relationships between plant diversity and soil microbial diversity vary across taxonomic groups and spatial scales. *Ecosphere* **11**, e02999 (2020).
49. M. G. A. van der Heijden, J. N. Klironomos, M. Ursic, P. Moutoglis, R. Streitwolf-Engel, T. Boller, A. Wiemken, I. R. Sanders, Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* **396**, 69–72 (1998).
50. S. König, T. Wubet, C. F. Dormann, S. Hempel, C. Renker, F. Buscot, TaqMan real-time PCR assays to assess arbuscular mycorrhizal responses to field manipulation of grassland biodiversity: Effects of soil characteristics, plant species richness, and functional traits. *Appl. Environ. Microbiol.* **76**, 3765–3775 (2010).
51. T. Rottstock, J. Joshi, V. Kummer, M. Fischer, Higher plant diversity promotes higher diversity of fungal pathogens, while it decreases pathogen infection per plant. *Ecology* **95**, 1907–1917 (2014).
52. L. Fuchslueger, M. Bahn, K. Fritz, R. Hasibeder, A. Richter, Experimental drought reduces the transfer of recently fixed plant carbon to soil microbes and alters the bacterial community composition in a mountain meadow. *New Phytol.* **201**, 916–927 (2014).
53. P. Mariotte, A. Canarini, F. A. Dijkstra, Stoichiometric N:P flexibility and mycorrhizal symbiosis favour plant resistance against drought. *J. Ecol.* **105**, 958–967 (2017).
54. A. Sendek, C. Karakoç, C. Wagg, J. Domínguez-Begines, G. M. do Couto, M. G. A. van der Heijden, A. A. Naz, A. Lochner, A. Chatzinotas, S. Klotz, L. Gómez-Aparicio, N. Eisenhauer, Drought modulates interactions between arbuscular mycorrhizal fungal diversity and barley genotype diversity. *Sci. Rep.* **9**, 9650 (2019).
55. A. Escalas, L. Hale, J. W. Voordeckers, Y. Yang, M. K. Firestone, L. Alvarez-Cohen, J. Zhou, Microbial functional diversity: From concepts to applications. *Ecol. Evol.* **9**, 12000–12016 (2019).
56. L. Fuchslueger, M. Bahn, R. Hasibeder, S. Kienzl, K. Fritz, M. Schmitt, M. Watzka, A. Richter, Drought history affects grassland plant and microbial carbon turnover during and after a subsequent drought event. *J. Ecol.* **104**, 1453–1465 (2016).
57. A. Kaisermann, F. T. de Vries, R. I. Griffiths, R. D. Bardgett, Legacy effects of drought on plant-soil feedbacks and plant-plant interactions. *New Phytol.* **215**, 1413–1424 (2017).
58. T. R. Cavagnaro, Soil moisture legacy effects: Impacts on soil nutrients, plants and mycorrhizal responsiveness. *Soil Biol. Biochem.* **95**, 173–179 (2016).
59. J. M. G. Bloor, R. D. Bardgett, Stability of above-ground and below-ground processes to extreme drought in model grassland ecosystems: Interactions with plant species diversity and soil nitrogen availability. *Perspect. Plant Ecol. Evol. Syst.* **14**, 193–204 (2012).
60. N. Legay, G. Piton, C. Arnoldi, L. Bernard, M.-N. Binet, B. Mouhamadou, T. Pommier, S. Lavorel, A. Foulquier, J.-C. Clément, Soil legacy effects of climatic stress, management and plant functional composition on microbial communities influence the response of *Lolium perenne* to a new drought event. *Plant and Soil* **424**, 233–254 (2018).

61. J. C. Yuste, J. Peñuelas, M. Estiarte, J. Garcia-Mas, S. Mattana, R. Ogaya, M. Pujol, J. Sardans, Drought-resistant fungi control soil organic matter decomposition and its response to temperature. *Glob. Chang. Biol.* **17**, 1475–1486 (2011).
62. G. Piton, N. Legay, C. Arnoldi, S. Lavorel, J.-C. Clément, A. Foulquier, Using proxies of microbial community-weighted means traits to explain the cascading effect of management intensity, soil and plant traits on ecosystem resilience in mountain grasslands. *J. Ecol.* **108**, 876–893 (2020).
63. A. Vogel, M. Scherer-Lorenzen, A. Weigelt, Grassland resistance and resilience after drought depends on management intensity and species richness. *PLOS ONE* **7**, e36992 (2012).
64. D. Craven, F. Isbell, P. Manning, J. Connolly, H. Bruelheide, A. Ebeling, C. Roscher, J. van Ruijven, A. Weigelt, B. Wilsey, C. Beierkuhnlein, E. de Luca, J. N. Griffin, Y. Hautier, A. Hector, A. Jentsch, J. Kreyling, V. Lanta, M. Loreau, S. T. Meyer, A. S. Mori, S. Naeem, C. Palmborg, H. W. Polley, P. B. Reich, B. Schmid, A. Siebenkäs, E. Seabloom, M. P. Thakur, D. Tilman, A. Vogel, N. Eisenhauer, Plant diversity effects on grassland productivity are robust to both nutrient enrichment and drought. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* **371**, 20150277 (2016).
65. E. Haughey, M. Suter, D. Hofer, N. J. Hoekstra, J. C. McElwain, A. Lüscher, J. A. Finn, Higher species richness enhances yield stability in intensively managed grasslands with experimental disturbance. *Sci. Rep.* **8**, 15047 (2018).
66. L. E. Forero, A. Kulmatiski, J. Grenzer, J. M. Norton, Plant-soil feedbacks help explain biodiversity-productivity relationships. *Commun. Biol.* **4**, 789 (2021).
67. J. Jing, T. M. Bezemer, W. H. van der Putten, Complementarity and selection effects in early and mid-successional plant communities are differentially affected by plant-soil feedback. *J. Ecol.* **103**, 641–647 (2015).
68. R. Michalet, F. Delerue, P. Liancourt, F. I. Pugnaire, Are complementarity effects of species richness on productivity the strongest in species-rich communities? *J. Ecol.* **109**, 2038–2046 (2021).
69. A. M. Veach, L. H. Zeglin, Historical drought affects microbial population dynamics and activity during soil drying and re-wet. *Microb. Ecol.* **79**, 662–674 (2020).
70. S. Wang, F. Isbell, W. Deng, P. Hong, L. E. Dee, P. Thompson, M. Loreau, How complementarity and selection affect the relationship between ecosystem functioning and stability. *Ecology* **102**, e03347 (2021).
71. S. Wurst, T. Ohgushi, Do plant- and soil-mediated legacy effects impact future biotic interactions? *Funct. Ecol.* **29**, 1373–1382 (2015).
72. D. L. Hoover, B. M. Rogers, Not all droughts are created equal: The impacts of interannual drought pattern and magnitude on grassland carbon cycling. *Glob. Chang. Biol.* **22**, 1809–1820 (2016).
73. F. T. de Vries, M. E. Liiri, L. Bjørnlund, H. M. Setälä, S. Christensen, R. D. Bardgett, Legacy effects of drought on plant growth and the soil food web. *Oecologia* **170**, 821–833 (2012).
74. A. E. Berns, H. Philipp, H.-D. Narres, P. Burauel, H. Vereecken, W. Tappe, Effect of gamma-sterilization and autoclaving on soil organic matter structure as studied by solid state NMR, UV and fluorescence spectroscopy. *Eur. J. Soil Sci.* **59**, 540–550 (2008).
75. N. Xi, J. M. G. Bloor, C. Chu, Soil microbes alter seedling performance and biotic interactions under plant competition and contrasting light conditions. *Ann. Bot.* **126**, 1089–1098 (2020).
76. M. Loreau, A. Hector, Partitioning selection and complementarity in biodiversity experiments. *Nature* **412**, 72–76 (2001).
77. J. G. Caporaso, C. L. Lauber, W. A. Walters, D. Berg-Lyons, C. A. Lozupone, P. J. Turnbaugh, N. Fierer, R. Knight, Global patterns of 16S rRNA diversity at a depth of millions of sequences per sample. *Proc. Natl. Acad. Sci. U.S.A.* **108**, 4516–4522 (2011).
78. T. J. White, T. Bruns, S. Lee, J. Taylor, Amplification and direct sequencing of fungal ribosomal RNA genes for phylogeentics, in *PCR Protocols*, M. A. Innis, D. H. Gelfand, J. J. Sninsky, T. J. White, Eds. (Academic, San Diego, 1990), pp. 315–322.
79. N. H. Nguyen, Z. Song, S. T. Bates, S. Branco, L. Tedersoo, J. Menke, J. S. Schilling, P. G. Kennedy, FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecol.* **20**, 241–248 (2016).
80. R Development Core Team, R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. (2021); https://www.r-project.org/.


**Acknowledgments:** We thank Y.-Y. Zhang (Xiamen University) for the constructive comments on our initial manuscript. **Funding:** This work was supported by the National Natural Science Foundation of China grant 31600342 (N.X.), National Natural Science Foundation of China grant 31925027 (C.C.), and Agence National de la Recherche grant 16-IDEX-0001 (J.M.G.B.).
**Author contributions:** Conceptualization: N.X. and J.M.G.B. Methodology: N.X. and D.C. Investigation: N.X., D.C., and H.W. Visualization: D.C. and N.X. Supervision: N.X. and C.C. Writing, original draft: N.X., D.C., and J.M.G.B. Writing, review and editing: N.X., J.M.G.B., M.B., M.W.C., and C.C. **Competing interests:** The authors declare that they have no competing interests. **Data and materials availability:** All data needed to evaluate the conclusions in the paper are present in the paper and/or the Supplementary Materials.

Submitted 19 November 2021
Accepted 16 March 2022
Published 4 May 2022
10.1126/sciadv.abn3368

ScienceAdvances

# Drought soil legacy alters drivers of plant diversity-productivity relationships in oldfield systems

Nianxun XiDongxia ChenMichael BahnHangyu WuChengjin ChuMarc W. CadotteJuliette M. G. Bloor

*Sci. Adv.*, 8 (18), eabn3368. • DOI: 10.1126/sciadv.abn3368



*Science Advances* (ISSN ) is published by the American Association for the Advancement of Science. 1200 New York Avenue NW, Washington, DC 20005. The title *Science Advances* is a registered trademark of AAAS.