Plant Soil
https://doi.org/10.1007/s11104-025-07601-5

RESEARCH ARTICLE

Microbial diversity outweighs plant diversity in mediating the response of ecosystem multifunctionality to altered precipitation in a desert steppe

Cui Han · Yi Zhang · Xu Luo · Ying Zhao · Xueqin Yang · Jiali Lian · Jianping Li

Received: 9 December 2024 / Accepted: 27 May 2025

Abstract
Aims Dryland ecosystems are susceptible to variations in precipitation. Biodiversity plays a vital role in regulating ecosystem functions. However, the effects of altered precipitation on plant and soil microbial diversity and their relationship with ecosystem multifunctionality (EMF) in desert steppes remain unclear.
Methods We conducted a three–year precipitation manipulation experiment in a desert steppe in northwestern China to evaluate how altered precipitation influences plant and microbial diversity, EMF, and individual ecosystem functions.
Results R33 significantly decreased the Shannon and Pielou indices of plant, as well as EF–GP and EF–P. R66 significantly decreased EMF, EF–GP, EF–N, and EF–P. In contrast, only R166 significantly decreased the Pielou index of plant. There were significant interannual variations in plant diversity, microbial diversity, EMF, and single functions. The interaction between precipitation and year significantly affected only plant diversity, microbial diversity, and EF–GP. The multi–threshold approach indicated that ecosystem functions were positively correlated with plant and fungal diversity and negatively correlated with bacterial diversity under altered precipitation. Additionally, the structural equation model demonstrated that precipitation regulated EMF by affecting plant and microbial diversity through changes in soil water content. Compared with plant diversity, microbial diversity was more significant in regulating the response of EMF to altered precipitation.
Conclusions This study demonstrates that plant and microbial diversity jointly regulate ecosystem functions under altered precipitation, but microbial diversity plays a more prominent role. Therefore, the importance of microbial communities should be given particular attention when predicting and managing the response of dryland ecosystems to future climate–induced precipitation changes.

Keywords Ecosystem multifunctionality · Plant diversity · Soil microbial diversity · Desert steppe · Precipitation

Responsible Editor: Xinhua He.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s11104-025-07601-5.

C. Han · Y. Zhang · X. Luo · Y. Zhao · X. Yang · J. Lian · J. Li
School of Forestry and Grassland Science, Ningxia University, Yinchuan 750021, China

C. Han · Y. Zhang · X. Luo · Y. Zhao · X. Yang · J. Lian · J. Li (✉)
Northern Yanchi Desert Steppe Observation and Research Station of Ningxia, Yanchi 751500, China
e-mail: lijianpingsas@163.com

Published online: 05 June 2025

Introduction

Based on the IPCC's Sixth Assessment Report findings, the global average temperature has increased significantly over the past 100 years, affecting the global hydrological cycle and changing the spatial and temporal regimes of precipitation (IPCC, 2021). Precipitation directly affects key ecological processes, including plant growth and elemental cycling (Seddon et al. 2016), and drives ecosystem evolution by regulating plant–microbe–soil relationships, thereby profoundly affecting terrestrial biodiversity and ecosystem functioning (Felton et al. 2020; Zhou et al. 2020). Grassland ecosystems, a crucial part of terrestrial ecosystems, account for approximately 40% of the global land area. They are vital in supporting biodiversity and improving various ecosystem services and functions (Gibson 2008; Wang et al. 2022). Grasslands are primarily distributed in arid and semi–arid regions, where water availability is the predominant limiting factor. Altered precipitation regulates the availability of soil resources in ecosystems, thereby affecting plant growth and microbial activity (Beier et al. 2012; Cleland et al. 2006). These alterations are strongly linked to the stability of the vegetation–soil system. Therefore, research on precipitation changes in grassland ecosystems can provide data for an in–depth investigation of the ecological effects of altered precipitation patterns.

Ecosystem functions, which include energy flow, material cycling, and information transfer, arise from interactions between species and their environments (Jax 2005). Ecosystem multifunctionality (EMF) describes an ecosystem's capacity to deliver multiple functions and services simultaneously (Sanderson et al. 2004; Zavaleta et al. 2010). The relationship between biodiversity and EMF has gained significant attention in recent decades, particularly in light of the ongoing global biodiversity loss (Delgado-Baquerizo et al. 2016; Hu et al. 2021; Maestre et al. 2012). For instance, EMF has dramatically improved with increased plant diversity in different ecosystems. However, the observed relationship between plant diversity and EMF in global dryland ecosystems is only weakly positive (Maestre et al. 2012). Moreover, research on the Tibetan Plateau revealed that plant species richness is positively associated with EMF, which largely explains its spatial variation (Jing et al. 2015). Soil microbial diversity is crucial in influencing EMF, but its effects vary among different trophic groups (Bastida et al. 2016; Wagg et al. 2019). Further research has shown that ecosystems comprise multiple trophic levels. Different trophic groups may have opposite or complementary effects on ecosystem functions, with different relative contributions (Soliveres et al. 2016). Therefore, thoroughly considering the interactions between different trophic groups can more accurately indicate the role of biodiversity in EMF regulation.

Precipitation is a crucial factor affecting key processes in grassland ecosystems, and changes in its pattern affect grassland biodiversity, EMF, and their interactions (Beier et al. 2012; Zhou et al. 2020). Recent findings have demonstrated that increased precipitation has a positive (Shi et al. 2024) or insignificant (Wang et al. 2023) effect on EMF, whereas drought reduces EMF (Zhai et al. 2024) in grassland ecosystems. Most studies have found that increased precipitation improves soil moisture and affects plant water status and photosynthetic rates, positively affecting grassland plant diversity. In contrast, decreased precipitation reduces plant species richness, particularly under extreme precipitation reductions (Currie et al. 2004; Li et al. 2018; Wu et al. 2024). Studies have also demonstrated that increased precipitation intensifies aboveground plant competition, increasing competitive exclusion and reducing species richness (DeMalach et al. 2017). Changes in precipitation may have positive (Cregger et al. 2012), negative (Yang et al. 2024), or insignificant (Gutknecht et al. 2012) effects on microbial diversity by regulating plant–soil nutrient dynamics and changing soil temperature and water availability. Additionally, precipitation may indirectly regulate EMF through the cascade effect of changes in the composition and structure of plant and microbial communities (Valencia et al. 2018; Wolters et al. 2000). Nevertheless, our current understanding of how grassland biodiversity reacts to altered precipitation and the subsequent effects on biodiversity–related EMF remains limited, necessitating further experimental exploration.

As a major grassland ecosystem in the arid and semi–arid regions of northwestern China, the desert steppe is an ecologically fragile area with low precipitation and high evaporation. This ecosystem is particularly vulnerable to altered precipitation owing to its low water availability (Na et al. 2019). Although some studies have indicated the effects of changing precipitation on biodiversity and EMF in grassland ecosystems (Wang et al. 2023; Yang et al. 2024; Zhai et al. 2024), extensive conclusions concerning the relationship between aboveground and belowground biodiversity and EMF in the desert steppes of northern China are yet to be drawn. In this study, the effects of precipitation on plant and microbial diversity and 11 ecosystem functions were assessed through a field experiment on precipitation changes in the desert steppe of northwestern China. A structural equation model (SEM) was used to investigate the indirect effects of precipitation on EMF by assessing its impact on plant and microbial diversity in the desert steppe. The following hypotheses were examined in this study: 1) increased precipitation would improve plant diversity and EMF in the desert steppe, while the response of microbial diversity to altered precipitation differs between microbes; 2) the contribution would vary among diverse biodiversity to EMF, but in general microbes are more sensitive to environmental changes, and their diversity may contribute more to regulating EMF.

Material and methods

Site description

The experiment was conducted at the Northern Yanchi Desert Steppe Observation and Research Station of Ningxia, Yanchi County, China (37°48′ N, 107°27′ E, 1367 m a.s.l). The selected site is an ecological transition zone in a semi–arid region with a continental semi-arid monsoon climate vulnerable to climate change and human activity. Over the past 41 years (1981–2021), the average annual temperature was 8.7 ℃, and the average annual precipitation measured 294.3 mm (Fig. S1). Approximately 80% of the annual precipitation occurs during the plant–growing season from May to September. During the experimental period, the annual precipitation was 323.1 mm in 2019 and 227.9 mm in 2021, while the annual mean temperatures were 9.3 ℃ and 10.2 ℃, respectively. These variations highlight the interannual climate fluctuations that may influence plant and microbial community dynamics, as well as ecosystem multifunctionality (Fig. S1). The soil in the experimental field was classified as Sierozem according to the Chinese Classification System. The soil is moderately alkaline, with its pH ranging from approximately 8.46 to 9.08. The vegetation in this region is characterized as desert steppe. The dominant species are *Lespedeza potaninii*, *Agropyron mongolicum*, *Polygala tenuifolia*, *Artemisia scoparia*, and *Astragalus melilotoides*.

Experimental design

In June 2018, we selected a representative area with flat terrain and uniform vegetation for a long–term precipitation change experiment (Fig. 1). Precipitation treatments were determined based on the average annual precipitation and extreme fluctuations in precipitation observed over the past 38 years (1981–2018) in the study area, while considering the practicability of the field experiment (Fig. S2). Five precipitation treatments were established using a completely randomized block design with three replicates, resulting in 15 plots. Each plot measured 6 m ×6 m with a 5–m buffer zone. The five experimental treatments used were R33 (33% natural precipitation), R66 (66% natural precipitation), RCK (control, natural precipitation), R133 (133% natural precipitation), and R166 (166% natural precipitation). Decreased precipitation was achieved using home–made rain shelters (year–round rain protection). Tile–like transparent polyvinyl chloride (PVC) sheets of 15 cm width were uniformly placed on a rain shelter, approximately 1.8 m high, to shade out natural precipitation equivalent to 66% and 33% of the plot, respectively. Increased precipitation was achieved using manual sprinklers. During the experiment, the precipitation was collected using rain barrels. Natural precipitation levels of 33% and 66% were calculated and evenly applied to the plots corresponding to the R133 and R166 treatments through sprinkler irrigation within 48 h of the precipitation event. A 1.2–m–wide plastic sheet was buried around each plot. The burial depth was 1.1 m, and 0.1 m was left aboveground to prevent disturbances such as surface run–off and sub–surface leakage.

Field survey and sampling

In mid–August 2019 and 2021, three 1 × 1 m quadrats were randomly placed in each plot for plant community surveys and sampling. Plant species, coverage, density, height and frequency were also investigated.

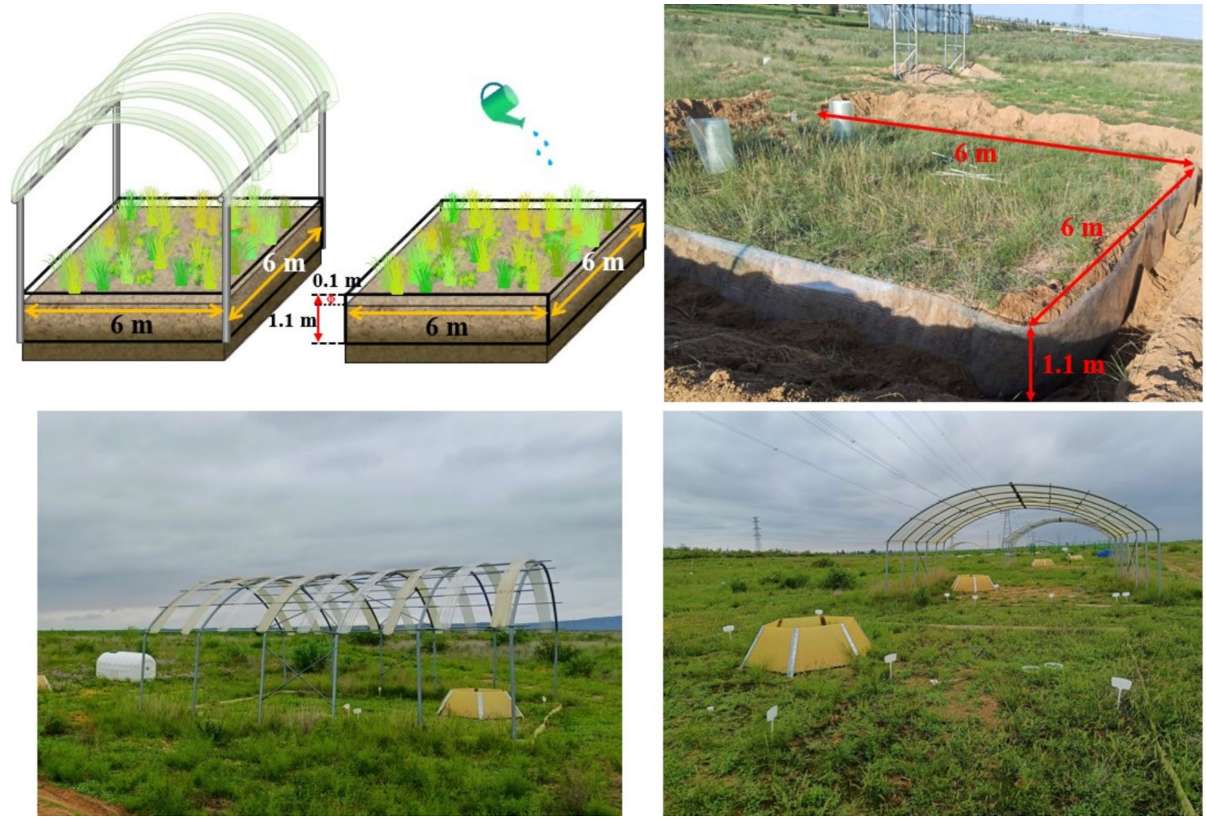

Fig. 1 Experimental plot design

Following the survey, aboveground vegetation was collected by species and placed in Kraft paper bags. The quadrats were cleaned after collecting the aboveground vegetation, and the roots from 0 to 30 cm in the quadrats were collected in mesh bags according to the species, rinsed, and placed in Kraft paper bags. The plants from each quadrat were sorted by species, dried in an oven at 65 ℃ for 48 h, and then weighed to determine the biomass of each plant population. The plant community aboveground biomass (AGB) and belowground biomass (BGB) in each quadrat were calculated by summing the dried biomass of all plant categories. The AGB, BGB, and plant diversity in each plot were calculated as the average biomass and diversity of three quadrats. Plant community diversity was assessed using the Shannon–Wiener diversity (*H*), Pielou evenness (*E*), Margalef (*Ma*) and Simpson dominance (*D*) indices.

$$P_i = (relative\ biomass + relative\ height + relative\ density)/3 \quad (1)$$

$$H = -\sum_{i=1}^{S} P_i \ln P_i \quad (2)$$

$$E = H/\ln S \quad (3)$$

$$Ma = (S-1)/\ln N \quad (4)$$

$$D = 1 - \sum_{i=1}^{S} P_i^2 \quad (5)$$

where S is the number of species in each quadrat, P_i is the importance value of species i, and N indicates the total number of individuals of the species in each quadrat.

A soil sub–sample (0–10 cm) was collected from each quadrat using a 5–cm–diameter soil drill. The subsamples from the three quadrats were combined into a single soil sample. Each soil sample was sieved through a 2 mm mesh and divided into two portions. One portion was placed in a 10 ml centrifuge tube and then transferred to a refrigerator at −80 ℃ to determine soil microbes. The remainder was air–dried for analysis of soil pH, organic C (C_s), total N (N_s), and P (P_s).

Plant and soil property measurements

Leaf and root samples from each category were dried, ground using a ball mill (MM400, Retsch, Germany), and sieved through a 0.15–mm mesh for elemental analysis. The concentrations of total C, N, and P in the leaves and roots of each category were measured using the $K_2Cr_2O_7$ oxidation (Mebius 1960), Kjeldahl method (Bremner 1996), and molybdenum yellow colorimetry, respectively (Bao 2000). The plant community–level leaf total C (C_l), N (N_l), and P (P_l) were calculated as community–weighted means of the traits (CWM) (Lavorel et al. 2008). Total root C (C_r), N (N_r), and P (P_r) at community–level were calculated using the same methods used for C_l, N_l, and P_l. C_s, N_s, and P_s were determined using the $K_2Cr_2O_7$ (Mebius 1960), Kjeldahl (Bremner 1996), and molybdenum antimony resistance colorimetric methods, respectively (Bao 2000). The soil water content (SWC) and pH were measured using a moisture detector carried by LI–8100 A (LI–COR, Nebraska, USA) and a portable acidity meter.

The Illumina MiSeq PE300 platform was used for microbial DNA sequencing, and microbial diversity was assessed. Supplementary Materials (Appendix S1) describe the methods used to quantify microbial diversity.

Ecosystem multifunctionality

The EMF index serves as a comprehensive measure of the ability of a system to maintain multiple functions concurrently. We selected 11 ecosystem function indicators and classified them into five ecosystem function groups (EF): 1) functions related to grassland ecosystem primary productivity (EF–GP: AGB and BGB); 2) functions related to grassland ecosystem C storage (EF–C: C_l, C_r, and C_s); 3) functions related to grassland ecosystem N storage (EF–N: N_l, N_r, and N_s); 4) functions related to grassland ecosystem P storage (EF–P: P_l, P_r, P_s); and 5) grassland ecosystem multifunctionality (EMF, including all 11 ecosystem function indicators). A common averaging approach was used to calculate the EMF index and each function was standardized using the maximum method before averaging. However, the averaging method cannot capture the unique levels of functionality offered by various indicators, such as when one function is delivered at a low level and another at a high level or when both functions are provided at a moderate level (Byrnes et al. 2014). To address this limitation, we supplemented the averaging method with a multi–threshold approach (Byrnes et al. 2014; Delgado-Baquerizo et al. 2016). These calculations were performed using the "*multifunc*" package in R 4.3.1.

Data analysis

One–way analysis of variance (ANOVA) was employed to analyze the annual effects of precipitation on plant diversity, microbial diversity, EMF, single functions and individual functions. All analyses were conducted using SPSS 22.0. The Mantel test was used to test the correlations among plant diversity, microbial diversity, EMF, and single functions. The "*LinkET*" and "*ggplot2*" packages were used for the Mantel test. Partial least-squares path models were used to determine the possible paths for the variables controlling EMF. We screened eight independent variables with large contributions by loading them in the outer models that required VIF > 0.7 and classified them into six categories: precipitation, SWC, pH, plant diversity (the Shannon and Pielou indices), bacterial diversity (the Shannon and ACE indices), and fungal diversity (the ACE and Coverage indices). These analyses were conducted using the "*plspm*" package in R 4.3.1. The results were visualized using Origin 2021 software.

Results

Impacts of precipitation on plant and soil microbial diversity

Precipitation significantly affected the Shannon, Pielou, and Margalef indices ($P < 0.001$), while the sampling year significantly affected the Shannon and Margalef indices ($P < 0.001$). Additionally, the interaction between precipitation and sampling year significantly affected the Pielou and Margalef indices ($P < 0.001$) (Fig. 2). Based on the combined two–year data, compared to the control (RCK), extremely decreased precipitation (R33) significantly decreased the Shannon and Pielou indices ($P < 0.05$), while extremely increased precipitation (R166) significantly decreased the Pielou index ($P < 0.05$) (Fig. 2 and Fig. S3).

Precipitation significantly affected the Shannon, ACE, and Coverage indices ($P < 0.05$), while the sampling year significantly affected the four bacteria indices ($P < 0.001$). Their interaction significantly affected the ACE and Coverage indices ($P < 0.05$) (Fig. 3). No significant effects of precipitation on soil bacterial diversity were observed during the two–year sampling period ($P > 0.05$) (Fig. 3 and Fig. S4).

Precipitation significantly affected the Shannon index ($P < 0.05$), while the sampling year significantly affected the ACE and Coverage indices ($P < 0.001$). Their interaction significantly affected the Shannon index ($P < 0.05$) (Fig. 4). No significant effects of precipitation on soil fungal diversity were observed during the two–year sampling period ($P > 0.05$) (Fig. 4 and Fig. S5).

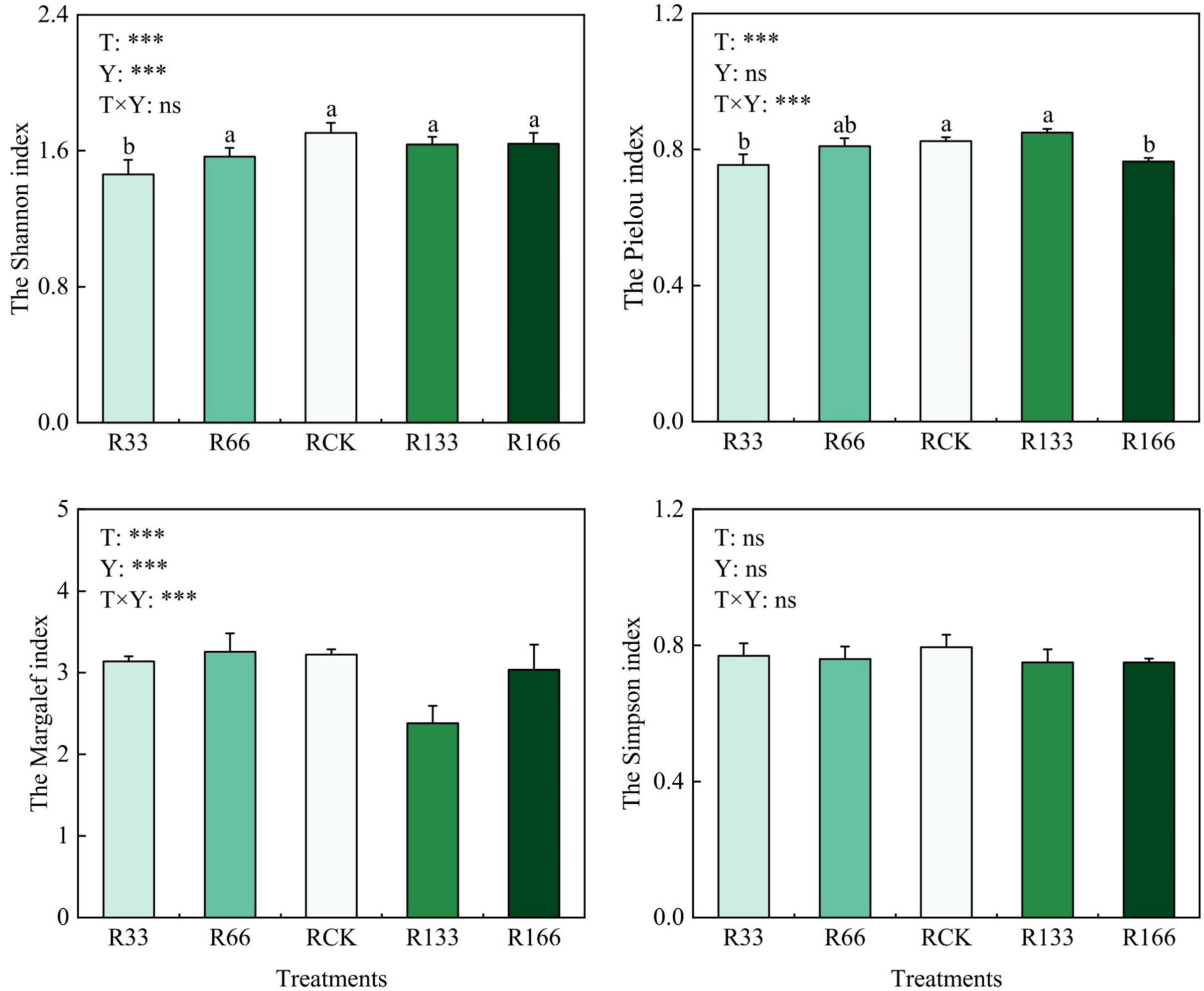

Fig. 2 Effects of precipitation on plant diversity over two years (mean ± SE). Lowercase letters denote significant differences among treatments ($P < 0.05$). T: precipitation treatments; Y: sampling year; T× Y: the interaction between precipitation treatments and sampling year. Not significant (ns): $P > 0.05$, *: $0.01 < P < 0.05$, **: $0.001 < P < 0.01$, ***: $P < 0.001$

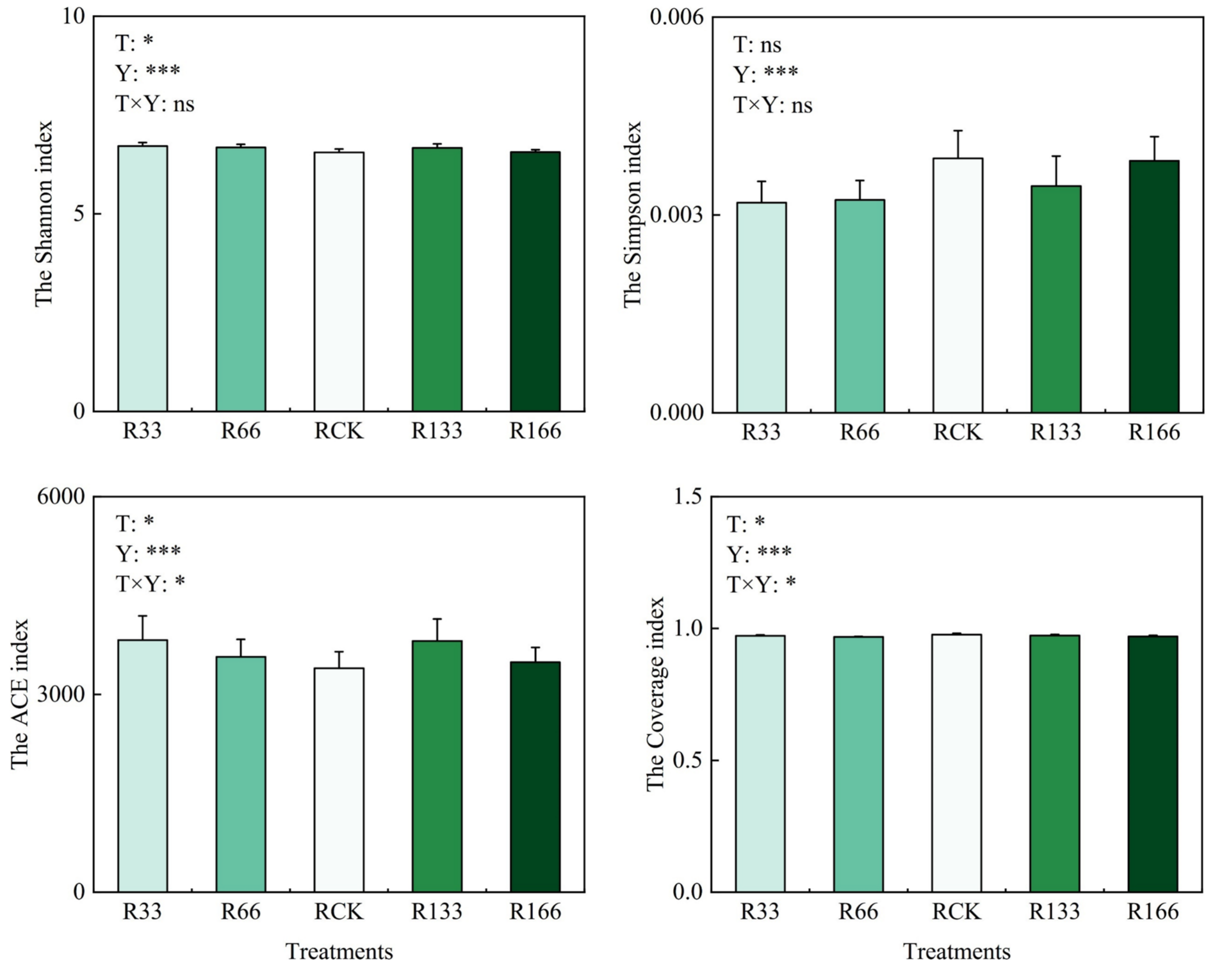

Fig. 3 Effects of precipitation on soil bacterial diversity over two years (mean ± SE). T: precipitation treatments; Y: sampling year; T× Y: the interaction between precipitation treatments and sampling year. Not significant (ns): $P > 0.05$, *: 0.01 < $P < 0.05$, **: 0.001 < $P < 0.01$, ***: $P < 0.001$

Impacts of precipitation on ecosystem multifunctionality and single functions

Precipitation significantly affected EMF, EF–GP, and EF–P ($P < 0.05$), while the sampling year significantly affected EMF, EF–GP, EF–C, EF–N, and EF–P ($P < 0.05$). Their interaction significantly affected EF–GP ($P < 0.001$) (Figs. 5 and 6). Based on the combined two–year data, compared with RCK, R33 significantly decreased EF–GP and EF–P ($P < 0.05$), moderately decreased precipitation (R66) significantly decreased EMF, EF–GP, EF–N, and EF–P ($P < 0.05$), R166 significantly increased EF–GP ($P < 0.05$) (Figs. 5, 6, and Fig. S6).

Relationships of plant and soil microbial diversity with ecosystem multifunctionality and single functions under altered precipitation

As shown in the Mantel test results (Fig. 7), EMF was strongly correlated with the Margalef index of plant and the Shannon index of bacteria ($P < 0.05$). EF–GP had strong correlations with the Margalef index of plant, the Simpson index of bacteria, and the Shannon index of fungi ($P < 0.05$). EF–C was strongly correlated with the Shannon and ACE indices of bacteria ($P < 0.05$). EF–N was strongly correlated with the Margalef index of plant and the Shannon index of bacteria ($P < 0.05$). EF–P was strongly correlated

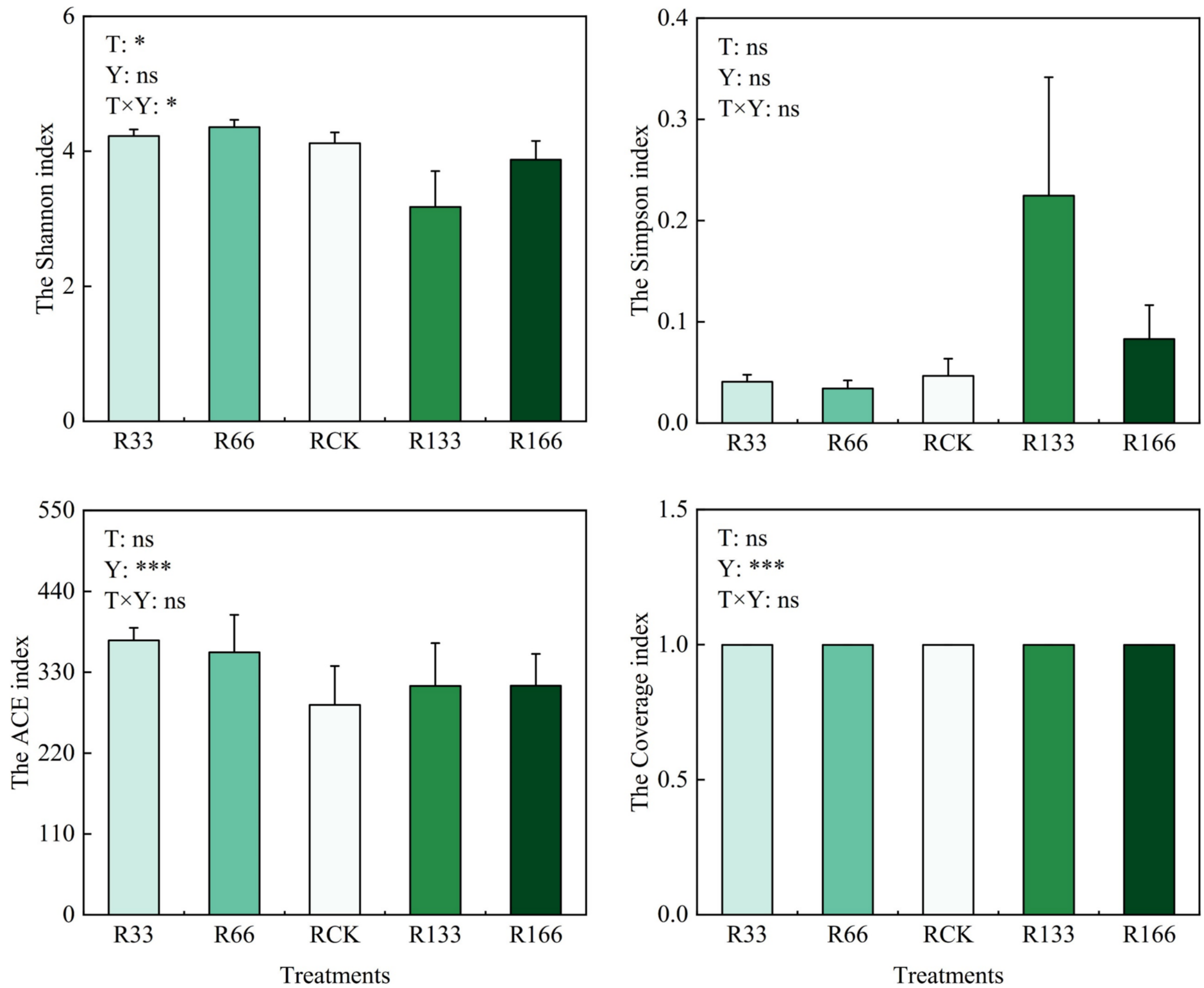

Fig. 4 Effects of precipitation on soil fungal diversity over two years (mean ± SE). T: precipitation treatments; Y: sampling year; T× Y: the interaction between precipitation treatments and sampling year. Not significant (ns): $P > 0.05$, *: $0.01 < P < 0.05$, **: $0.001 < P < 0.01$, ***: $P < 0.001$

with the Shannon and Pielou indices of plant ($P <$ 0.05).

This study also applied the multi–threshold approach to further explore the effects of plant and microbial diversity on ecosystem functions at different threshold levels. At a threshold of 83%, adding one plant species had the greatest positive effect on EMF, achieving a maximum realized diversity effect (Rmde) of 6.31 (Fig. 8a, d). This indicates that adding a single plant species resulted in a 6.31 increase in the ecosystem function. The threshold range for the effects of bacterial diversity on EMF was 57–70%, indicating that bacterial diversity began to affect EMF at the 57% threshold and that the effects were not significant at the 70% threshold (Fig. 8b, e). The inclusion of one fungal species at the 76% threshold increased 4.00 ecosystem function (Fig. 8c, f). Moreover, plant, bacterial, and fungal diversity influenced EF–GP, EF–C, and EF–N within different threshold ranges (Figs. S7–S9). Plant diversity significantly enhanced EF–P over a broad threshold range (66–99%). However, bacterial and fungal diversity showed no significant relationships with EF–P across all threshold levels (Fig. S10).

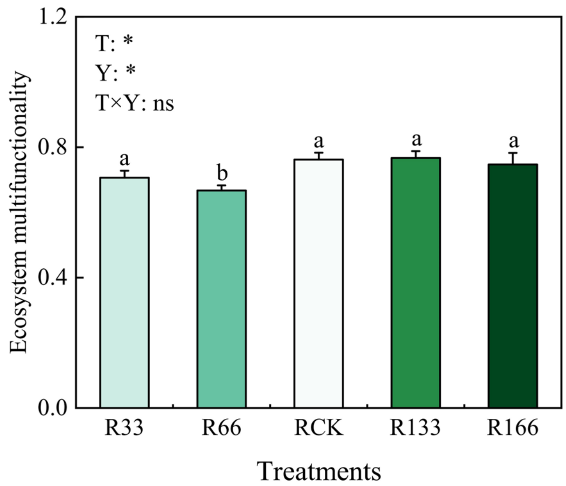

Fig. 5 Effects of precipitation on ecosystem multifunctionality (EMF) over two years (mean ± SE). Lowercase letters denote significant differences among treatments ($P < 0.05$). T: precipitation treatments; Y: sampling year; T× Y: the interaction between precipitation treatments and sampling year. Not significant (ns): $P > 0.05$, *: 0.01 < P < 0.05, **: 0.001 < P < 0.01, ***: $P < 0.001$

Critical factors affecting ecosystem multifunctionality under altered precipitation

Precipitation, SWC, soil pH, plant diversity, and microbial diversity collectively explained 30% of the variation in EMF (Fig. 9). Our SEM model revealed indirect positive effects of precipitation on EMF via plant diversity (+) and SWC (+). Plant diversity had a positive indirect effect on EMF through bacterial diversity. Bacterial diversity regulated negatively by SWC, played negative a central role in EMF. Fungal diversity had an indirect negative effect on EMF through bacterial diversity (Fig. 9a, b).

Discussion

Plant and microbial diversity responses to altered precipitation

Our study revealed that precipitation had a significant impact on plant diversity, but its effects on microbial diversity were relatively weak. Over the two–year period, decreased precipitation, especially extremely decreased precipitation, decreased plant diversity in the desert steppe (Fig. 2). These findings are consistent with those of the meta–analysis by Li et al. (2018b). Water is the most important ecological factor controlling plant growth in desert steppes. Decreased precipitation reduces SWC, which in turn causes drought stress on plant growth and development. Consequently, species less tolerant to drought are gradually excluded from the community, ultimately causing a decline in community diversity (Korell et al. 2021; Zuo et al. 2023). Increased precipitation is generally thought to provide sufficient water, improve plant habitats, and promote complementary ecological niche effects between species, thereby increasing plant diversity (Bunting et al. 2017). However, the results of the present study demonstrated that a 66% increase in precipitation decreased plant diversity. This may be because increased precipitation aggravates aboveground competition between plants, enhances competitive exclusion between species, and reduces community diversity (DeMalach et al. 2017). In contrast to plant diversity, bacterial and fungal diversity responded minimally to changing precipitation in the desert steppe (Figs. 3 and 4). Soil microbes have possibly adapted to the arid environment and low soil water utilization in the study area through long–term natural selection, resulting in a short–term decrease in precipitation that did not significantly alter microbial diversity (Na et al. 2019). Conversely, the response of microbial diversity to increased precipitation is influenced by variations in the soil microenvironment (e.g., soil temperature, moisture, pH, and nutrient contents) and plant communities, such that there may be a lag between the onset of the treatment and the emergence of significant effects (Gutknecht et al. 2012; Sherry et al. 2008). We observed that the sampling year and its interaction with precipitation affected bacterial and fungal diversity. The finding closely resembled the previous studies suggesting that soil microbial diversity depends on interannual variations in precipitation (Shi et al. 2020; Yang et al. 2021). This may be because interannual climatic conditions variability, such as temperature fluctuations and seasonal soil moisture dynamics, directly or indirectly affects the shaping of microbial communities (Shi et al. 2020).

Ecosystem multifunctionality and single functions response to altered precipitation

This study investigated the impacts of altered precipitation on ecosystem multifunctionality and single functions (functions related to grassland ecosystem

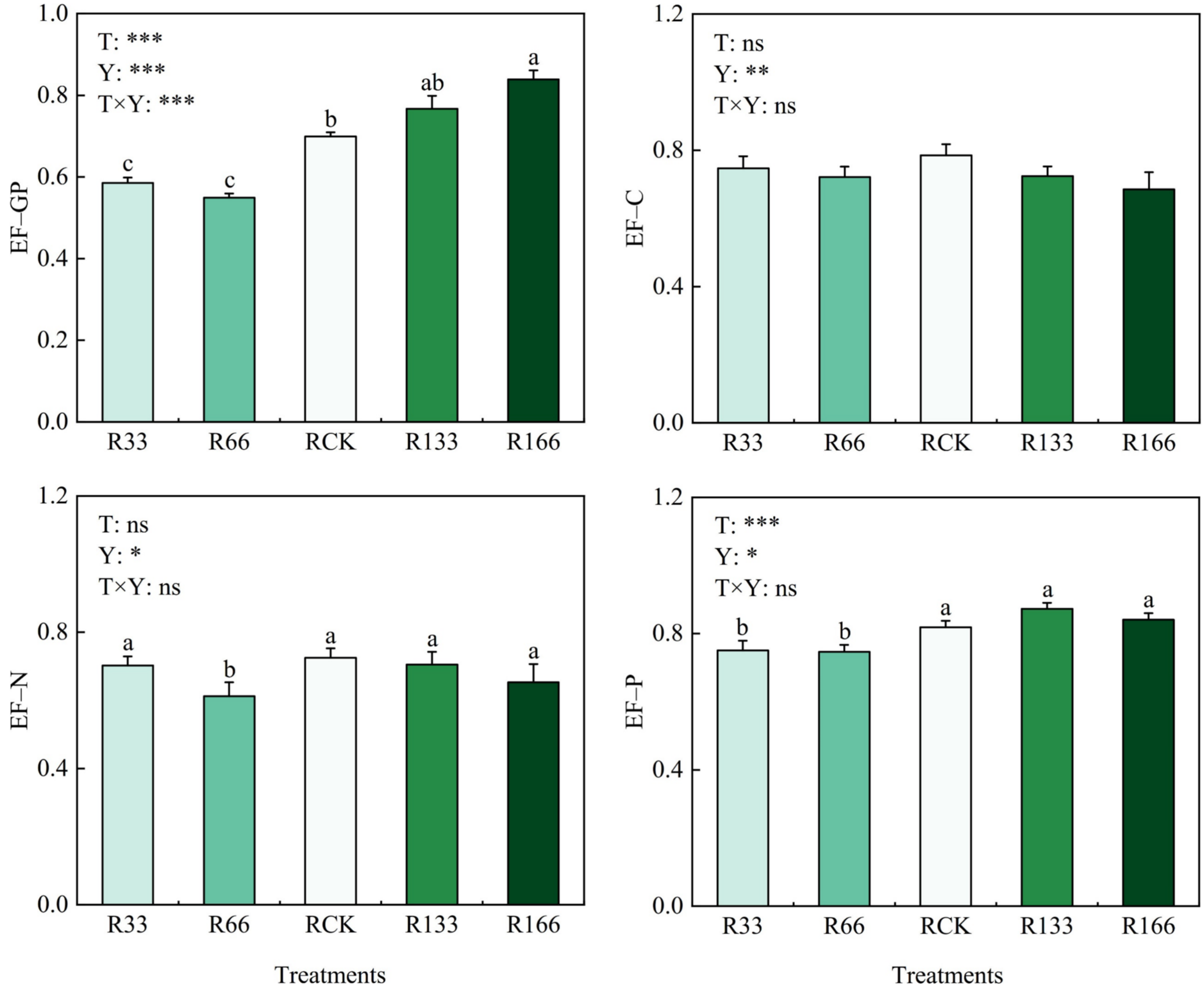

Fig. 6 Effects of precipitation on ecosystem single functions over two years (mean ± SE). EF–GP, grassland productivity; EF–C, plant–soil C contents; EF–N, plant–soil N contents; EF–P, plant–soil P contents

primary productivity, C storage, N storage, and P storage) within the desert steppe. The findings revealed that decreased precipitation decreased EMF, consistent with previous studies (Yang et al. 2023; Zhai et al. 2024). This effect was primarily attributed to the significant decrease in several single ecosystem functions (e.g., EF–GP, EF–N, and EF–P) that contributed to the overall EMF under decreased precipitation (Figs. 5 and 6). The study area is characterized by long–term low precipitation and high evapotranspiration (Fig. S1), rendering both plant and soil processes highly sensitive to changes in water availability. On the one hand, decreased precipitation lowers SWC, inhibiting plant growth and development and causing a decline in aboveground and belowground plant biomass (Yang et al. 2024; Zhang et al. 2024). On the other hand, drought aggravates water restriction, adversely impacting microbially mediated nutrient cycling processes (such as organic matter decomposition and nutrient mineralization), which reduces soil nutrient availability and suppresses key soil functions (Felton et al. 2020; Wang et al. 2020a). Furthermore, water shortages lead to a decrease in the rate plants absorb and utilize nutrients. This hinders the accumulation and metabolism of essential nutrients, resulting in lower N and P contents in plant leaves and roots (Sardans et al. 2008; Zeng et al. 2016). In contrast, increased precipitation had smaller effects on ecosystem functions, with only a 66% increase in precipitation significantly enhancing EF–GP (Fig. 6).

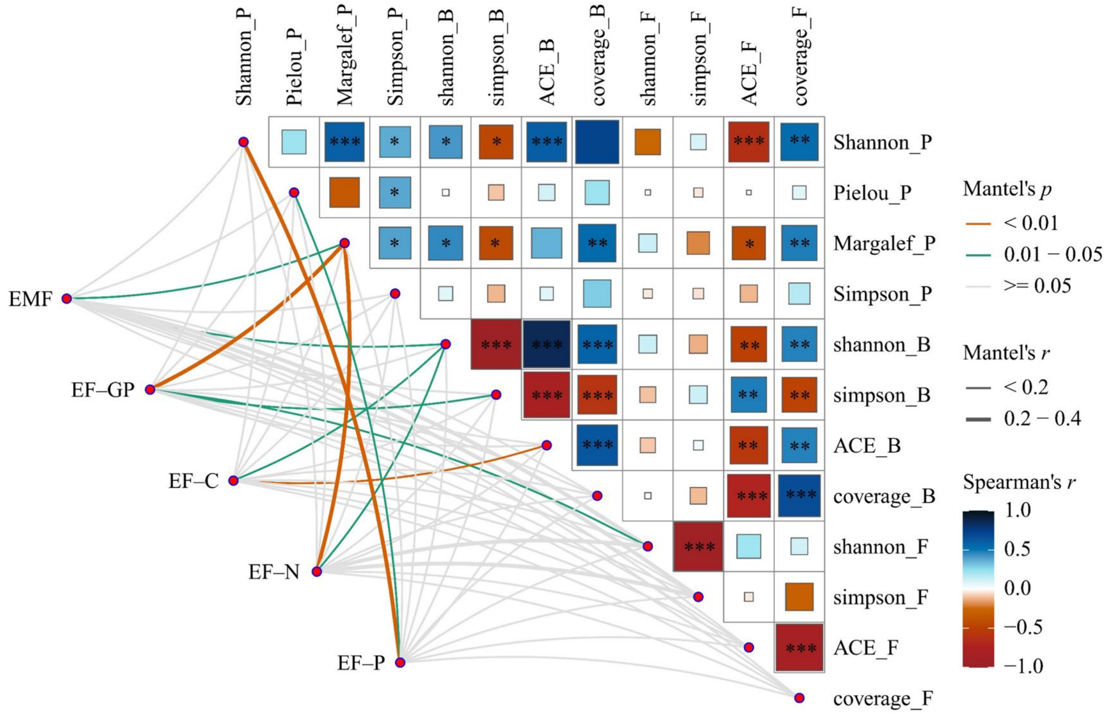

Fig. 7 Relationships among plant and soil microbial diversity with ecosystem multifunctionality (EMF) and single functions. EF–GP, grassland productivity; EF–C, plant–soil C contents; EF–N, plant–soil N contents; EF–P, plant–soil P contents; P, plant; B, bacteria; F, fungi

This may be because soil moisture increases under extremely increased precipitation, which alleviated water stress on plant growth, promoted biomass accumulation, and improved plant productivity in the desert steppe (Li et al. 2018). However, moderately increased precipitation did not significantly improve EMF or most single ecosystem functions. A modest water supplementation is insufficient to substantially alleviate the long–standing water restrictions in this region, which may have been responsible for these results. Given the high evapotranspiration and the low water retention capacity of the sandy loam in the study area, small increases in precipitation may quickly dissipate, failing to generate sustained ecological effects (Hao et al. 2017; Zhang et al. 2022).

Role of plant and microbial diversity in mediating ecosystem multifunctionality and single functions under altered precipitation

Our findings revealed a close connection between biodiversity and ecosystem functions (Fig. 7). Specifically, we observed a significant positive correlation between plant diversity and ecosystem function under changing precipitation (Fig. 8). This relationship supports the continued maintenance of ecosystem functions, consistent with the results of previous studies (Guo et al. 2023; Hu et al. 2021). This may be attributed to efficient resource utilization by plant communities within a constrained range and the synergistic interactions between species that enhance ecosystem functions (Soliveres et al. 2016; van der Plas 2019). Interestingly, our findings also revealed a differential role of microbial diversity in ecosystem functions. Bacterial diversity was negatively correlated with ecosystem functions, whereas fungal diversity was positively correlated with ecosystem functions (Fig. 8). Previous researches have indicated that the impacts of bacterial diversity on ecosystem functions are not always positive, possibly because bacteria reduce ecosystem functions by competing with plant roots for limited nutrients, especially in desert steppes subjected to prolonged water and nutrient limitation (Van Der Heijden et al. 2008;

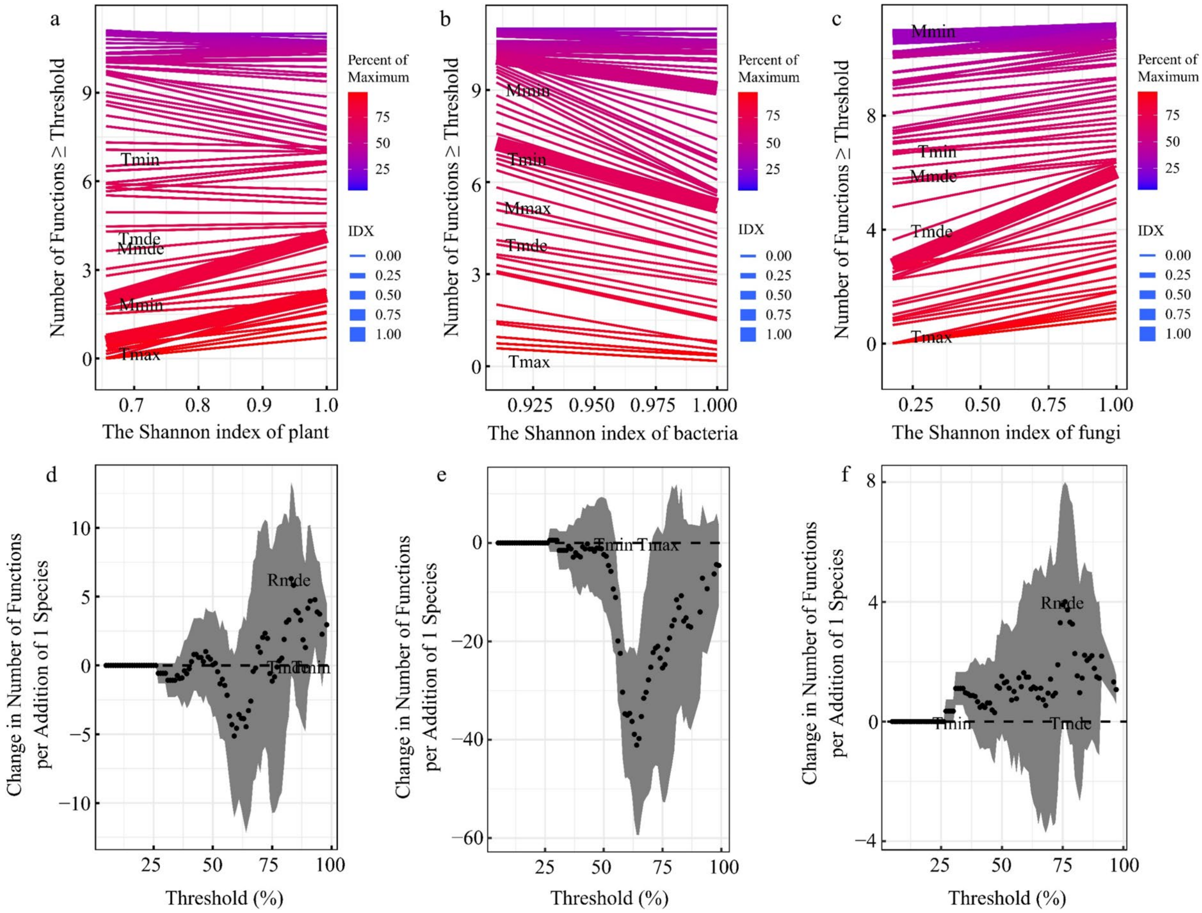

Fig. 8 Results of multi–threshold showing relationships among plant and soil microbial diversity with ecosystem function

Wang et al. 2020b). Furthermore, our results support previous observations and hypotheses that increased fungal diversity improves ecosystem function (Delgado-Baquerizo et al. 2016; Wagg et al. 2014). To our knowledge, the mycelial structure of fungi is essential for energy flow and nutrient cycling in soil ecosystems. Fungal communities can indirectly maintain ecosystem functions by altering resource allocation rates and improving plant nutrient use efficiency (Bhattacharyya and Furtak 2023; Holden et al. 2013).

In this study, we expanded our analysis of the factors and possible pathways affecting EMF by incorporating SWC and pH into the SEM. The results indicated that precipitation exerted a substantial direct positive impact on EMF and indirectly regulated EMF by influencing plant and microbial diversity: plant diversity had indirect effects on EMF, which was positively regulated by precipitation. Bacterial diversity was negatively regulated by SWC, and played a central negative role in EMF. Fungal diversity was influenced by soil moisture and pH, and it indirectly affected EMF through bacterial diversity (Fig. 9). This result confirms our previous results. Water is consistently a major limiting factor in desert steppes. Increased precipitation mitigates water constraints on ecosystems, enhancing plant growth, accelerating litter decomposition and mineralization, and improving soil nutrient effectiveness. This, in turn, contributes to an increase in EMF (Felton et al. 2020; Hu et al. 2022). However, variations in soil moisture resulting from altered precipitation affect the composition and structure of biological communities, and the cascade effects between multi-trophic levels, thereby regulating EMF (Bellard et al. 2012; Soliveres et al. 2016; Wagg et al. 2014). Furthermore, our findings indicated that when combining direct and indirect effects,

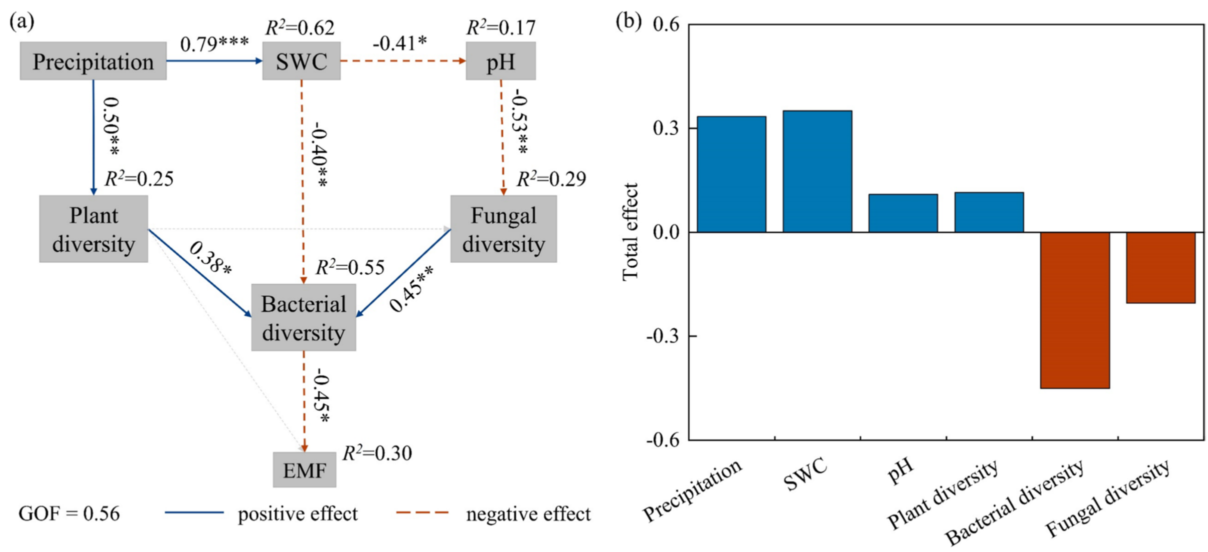

Fig. 9 Structural equation model (SEM) illustrating the impacts of treatment on ecosystem multifunctionality (EMF). SWC, soil water content; pH, soil pH; *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$. Solid blue arrows represent significant positive correlations, while solid orange arrows indicate significant negative correlations. Gray dashed arrows denote insignificant paths. The path coefficients are listed beside the arrows. R^2 values indicate the percentage of variance explained by the model

bacterial and fungal diversity exerted stronger effects on EMF than plant diversity (Fig. 9b). In summary, these findings show that, while plant and microbial diversity are essential for sustaining EMF and individual ecosystem functions, microbial diversity plays a more significant role in mediating the response of EMF to altered precipitation within the desert steppe.

Conclusions

Based on a two–year data analysis, we found that decreased precipitation decreased plant diversity, ecosystem multifunctionality, and single functions, but did not significantly affect microbial diversity. Increased precipitation had a weak effect on biodiversity, with only extremely increasing precipitation decreasing plant diversity and increasing plant productivity. There were interannual differences in plant diversity, microbial diversity, ecosystem multifunctionality, and single functions, whereas the interactions between precipitation and sample year significantly affected only plant diversity, microbial diversity, and plant productivity. Furthermore, plant and microbial diversity collaboratively regulate ecosystem functions in response to altered precipitation. Microbial diversity plays a greater role in mediating ecosystem functions under altered precipitation conditions than plant diversity.

Acknowledgements This research was funded by the Natural Science Foundation of Ningxia (2023AAC02029), the National Natural Science Foundation of China (32160336), the Top Discipline Construction Project of Pratacultural Science of Ningxia University (NXYLXK2017A01), and the Key Research and Development Project of Yinchuan City (2023NYZC01).

Funding the Natural Science Foundation of Ningxia, 2023AAC02029, jianping li, the National Natural Science Foundation of China, 32160336, jianping li, the Top Discipline Construction Project of Pratacultural Science of Ningxia University, NXYLXK2017A01, jianping li, the Key Research and Development Project of Yinchuan City, 2023NYZC01, jianping li. This work was also supported by the Key Research and Development Project of Yinchuan City (2023NYZC01).

Data availability The data will be provided upon reasonable request.

Declarations

Competing interest The authors of this manuscript affirm that they have no conflicts of interest to report.

References

Bao SD (2000) Soil and agricultural chemistry analysis Chap. 5: determination of phosphorus in soil. China Agriculture Press, Beijing, pp 70–90 (in Chinese with English abstract).

Bastida F, Torres IF, Moreno JL, Baldrian P, Ondoño S, Ruiz-Navarro A, Hernández T, Richnow HH, Starke R, García C, Jehmlich N (2016) The active microbial diversity drives ecosystem multifunctionality and is physiologically related to carbon availability in Mediterranean semi-arid soils. Mol Ecol 25:4660–4673. https://doi.org/10.1111/mec.13783

Beier C, Beierkuhnlein C, Wohlgemuth T, Penuelas J, Emmett B, Körner C, De Boeck H, Christensen JH, Leuzinger S, Janssens IA, Hansen K (2012) Precipitation manipulation experiments – challenges and recommendations for the future. Ecol Lett 15:899–911. https://doi.org/10.1111/j.1461-0248.2012.01793.x

Bellard C, Bertelsmeier C, Leadley P, Thuiller W, Courchamp F (2012) Impacts of climate change on the future of biodiversity. Ecol Lett 15:365–377. https://doi.org/10.1111/j.1461-0248.2011.01736.x

Bhattacharyya SS, Furtak K (2023) Soil–Plant–Microbe Interactions Determine Soil Biological Fertility by Altering Rhizospheric Nutrient Cycling and Biocrust Formation. Sustainability 15:625. https://doi.org/10.3390/su15010625

Bremner JM (1996) Nitrogen-Total. In: Sparks DL, Page AL, Helmke PA, Loeppert RH, Soltanpour PN, Tabatabai MA, Johnston CT, Sumner ME (eds) Methods of Soil Analysis Part 3: Chemical methods. Soil Science Society of America, Madison, pp 1085–1121. https://doi.org/10.2136/sssabookser5.3.c37

Bunting EL, Munson SM, Villarreal ML (2017) Climate legacy and lag effects on dryland plant communities in the southwestern U.S. Ecol Indic 74:216–229. https://doi.org/10.1016/j.ecolind.2016.10.024

Byrnes JEK, Gamfeldt L, Isbell F, Lefcheck JS, Griffin JN, Hector A, Cardinale BJ, Hooper DU, Dee LE, Emmett Duffy J (2014) Investigating the relationship between biodiversity and ecosystem multifunctionality: challenges and solutions. Methods Ecol Evol 5:111–124. https://doi.org/10.1111/2041-210X.12143

Cleland EE, Chiariello NR, Loarie SR, Mooney HA, Field CB (2006) Diverse responses of phenology to global changes in a grassland ecosystem. Proc Natl Acad Sci 103:13740–13744. https://doi.org/10.1073/pnas.0600815103

Cregger MA, Schadt CW, McDowell NG, Pockman WT, Classen AT (2012) Response of the Soil Microbial Community to Changes in Precipitation in a Semiarid Ecosystem. Appl Environ Microbiol 78:8587–8594. https://doi.org/10.1128/AEM.02050-12

Currie DJ, Mittelbach GG, Cornell HV, Field R, Guégan JF, Hawkins BA, Kaufman DM, Kerr JT, Oberdorff T, O'Brien E, Turner JRG (2004) Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. Ecol Lett 7:1121–1134. https://doi.org/10.1111/j.1461-0248.2004.00671.x

Delgado-Baquerizo M, Maestre FT, Reich PB, Jeffries TC, Gaitan JJ, Encinar D, Berdugo M, Campbell CD, Singh BK (2016) Microbial diversity drives multifunctionality in terrestrial ecosystems. Nat Commun 7:10541. https://doi.org/10.1038/ncomms10541

DeMalach N, Zaady E, Kadmon R (2017) Light asymmetry explains the effect of nutrient enrichment on grassland diversity. Ecol Lett 20:60–69. https://doi.org/10.1111/ele.12706

Felton AJ, Slette IJ, Smith MD, Knapp AK (2020) Precipitation amount and event size interact to reduce ecosystem functioning during dry years in a mesic grassland. Glob Change Biol 26:658–668. https://doi.org/10.1111/gcb.14789

Gibson DJ (2008) Grasses and Grassland Ecology. Oxford University Press. https://doi.org/10.1093/oso/9780198529187.001.0001

Guo H, Zhou X, Tao Y, Yin J, Zhang L, Guo X, Liu C, Lin Y, Zhang Y (2023) Precipitation preferences alter the relative importance of herbaceous plant diversity for multifunctionality in the drylands of China. Front Ecol Evol 11:1084949. https://doi.org/10.3389/fevo.2023.1084949

Gutknecht JLM, Field CB, Balser TC (2012) Microbial communities and their responses to simulated global change fluctuate greatly over multiple years. Glob Change Biol 18:2256–2269. https://doi.org/10.1111/j.1365-2486.2012.02686.x

Hao YB, Zhou CT, Liu WJ, Li LF, Kang XM, Jiang LL, Cui XY, Wang YF, Zhou XQ, Xu CY (2017) Aboveground net primary productivity and carbon balance remain stable under extreme precipitation events in a semiarid steppe ecosystem. Agric for Meteorol 240–241:1–9. https://doi.org/10.1016/j.agrformet.2017.03.006

Holden SR, Gutierrez A, Treseder KK (2013) Changes in Soil Fungal Communities, Extracellular Enzyme Activities, and Litter Decomposition Across a Fire Chronosequence in Alaskan Boreal Forests. Ecosystems 16:34–46. https://doi.org/10.1007/s10021-012-9594-3

Hu W, Ran J, Dong L, Du Q, Ji M, Yao S, Sun Y, Gong C, Hou Q, Gong H, Chen R, Lu J, Xie S, Wang Z, Huang H, Li X, Xiong J, Xia R, Wei M, Zhao D, Zhang Y, Li J, Yang H, Wang X, Deng Y, Sun Y, Li H, Zhang L, Chu Q, Li X, Aqeel M, Manan A, Akram MA, Liu X, Li R, Li F, Hou C, Liu J, He JS, An L, Bardgett RD, Schmid B, Deng J (2021) Aridity-driven shift in biodiversity–soil multifunctionality relationships. Nat Commun 12:5350. https://doi.org/10.1038/s41467-021-25641-0

Hu Y, Li X, Guo A, Yue P, Guo X, Lv P, Zhao S, Zuo X (2022) Species diversity is a strong predictor of ecosystem multifunctionality under altered precipitation in desert steppes. Ecol Indic 137:108762. https://doi.org/10.1016/j.ecolind.2022.108762

Jax K (2005) Function and "functioning" in ecology: what does it mean? Oikos 111:641–648. https://doi.org/10.1111/j.1600-0706.2005.13851.x

Jing X, Sanders NJ, Shi Yu, Chu H, Classen AT, Zhao K, Chen L, Shi Y, Jiang Y, He JS (2015) The links between ecosystem multifunctionality and above- and belowground biodiversity are mediated by climate. Nat Commun 6:8159. https://doi.org/10.1038/ncomms9159

Korell L, Auge H, Chase JM, Harpole WS, Knight TM (2021) Responses of plant diversity to precipitation change are strongest at local spatial scales and in drylands. Nat Commun 12:2489. https://doi.org/10.1038/s41467-021-22766-0

Lavorel S, Grigulis K, McIntyre S, Williams NSG, Garden D, Dorrough J, Berman S, Quétier F, Thébault A, Bonis A (2008) Assessing functional diversity in the field – methodology matters! Funct Ecol 22:134–147. https://doi.org/10.1111/j.1365-2435.2007.01339.x

Li W, Li X, Zhao Y, Zheng S, Bai Y (2018) Ecosystem structure, functioning and stability under climate change and grazing in grasslands: current status and future prospects. Curr Opin Env Sust 33:124–135. https://doi.org/10.1016/j.cosust.2018.05.008

Maestre FT, Quero JL, Gotelli NJ, Escudero A, Ochoa V, Delgado-Baquerizo M, García-Gómez M, Bowker MA, Soliveres S, Escolar C, García-Palacios P, Berdugo M, Valencia E, Gozalo B, Gallardo A, Aguilera L, Arredondo T, Blones J, Boeken B, Bran D, Conceição AA, Cabrera O, Chaieb M, Derak M, Eldridge DJ, Espinosa CI, Florentino A, Gaitán J, Gatica MG, Ghiloufi W, Gómez-González S, Gutiérrez JR, Hernández RM, Huang X, Huber-Sannwald E, Jankju M, Miriti M, Monerris J, Mau RL, Morici E, Naseri K, Ospina A, Polo V, Prina A, Pucheta E, Ramírez-Collantes DA, Romão R, Tighe M, Torres-Díaz C, Val J, Veiga JP, Wang D, Zaady E (2012) Plant Species Richness and Ecosystem Multifunctionality in Global Drylands. Science 335:214–218. https://doi.org/10.1126/science.1215442

Mebius LJ (1960) A rapid method for the determination of organic carbon in soil. Anal Chim Acta 22:120–124. https://doi.org/10.1016/S0003-2670(00)88254-9

Na X, Yu H, Wang P, Zhu W, Niu Y, Huang J (2019) Vegetation biomass and soil moisture coregulate bacterial community succession under altered precipitation regimes in a desert steppe in northwestern China. Soil Biol and Biochem 136:107520. https://doi.org/10.1016/j.soilbio.2019.107520

Sanderson MA, Skinner RH, Barker DJ, Edwards GR, Tracy BF, Wedin DA (2004) Plant Species Diversity and Management of Temperate Forage and Grazing Land Ecosystems. Crop Sci 44:1132–1144. https://doi.org/10.2135/cropsci2004.1132

Sardans J, Peñuuelas J, Estiarte M, Prieto P (2008) Warming and drought alter C and N concentration, allocation and accumulation in a Mediterranean shrubland. Glob Change Biol 14:2304–2316. https://doi.org/10.1111/j.1365-2486.2008.01656.x

Seddon AWR, Macias-Fauria M, Long PR, Benz D, Willis KJ (2016) Sensitivity of global terrestrial ecosystems to climate variability. Nature 531:229–232. https://doi.org/10.1038/nature16986

Sherry RA, Weng E, Arnone Iii JA, Johnson DW, Schimel DS, Verburg PS, Wallace LL, Luo Y (2008) Lagged effects of experimental warming and doubled precipitation on annual and seasonal aboveground biomass production in a tallgrass prairie. Glob Change Biol 14:2923–2936. https://doi.org/10.1111/j.1365-2486.2008.01703.x

Shi Y, Zhang K, Li Q, Liu X, He JS, Chu H (2020) Interannual climate variability and altered precipitation influence the soil microbial community structure in a Tibetan Plateau grassland. Sci Total Environ 714:136794. https://doi.org/10.1016/j.scitotenv.2020.136794

Shi L, Lin Z, Yao Z, Peng C, Hu M, Yin N, Lu X, Zhou H, Liu K, Shao X (2024) Increased precipitation rather than warming increases ecosystem multifunctionality in an alpine meadow. Plant Soil 498:357–370. https://doi.org/10.1007/s11104-023-06441-5

Soliveres S, van der Plas F, Manning P, Prati D, Gossner MM, Renner SC, Alt F, Arndt H, Baumgartner V, Binkenstein J, Birkhofer K, Blaser S, Blüthgen N, Boch S, Böhm S, Börschig C, Buscot F, Diekötter T, Heinze J, Hölzel N, Jung K, Klaus VH, Kleinebecker T, Klemmer S, Krauss J, Lange M, Morris EK, Müller J, Oelmann Y, Overmann J, Pašalić E, Rillig MC, Schaefer HM, Schloter M, Schmitt B, Schöning I, Schrumpf M, Sikorski J, Socher SA, Solly EF, Sonnemann I, Sorkau E, Steckel J, Steffan-Dewenter I, Stempfhuber B, Tschapka M, Türke M, Venter PC, Weiner CN, Weisser WW, Werner M, Westphal C, Wilcke W, Wolters V, Wubet T, Wurst S, Fischer M, Allan E (2016) Biodiversity at multiple trophic levels is needed for ecosystem multifunctionality. Nature 536:456–459. https://doi.org/10.1038/nature19092

Valencia E, Gross N, Quero JL, Carmona CP, Ochoa V, Gozalo B, Delgado-Baquerizo M, Dumack K, Hamonts K, Singh BK, Bonkowski M, Maestre FT (2018) Cascading effects from plants to soil microorganisms explain how plant species richness and simulated climate change affect soil multifunctionality. Glob Change Biol 24:5642–5654. https://doi.org/10.1111/gcb.14440

Van Der Heijden MGA, Bardgett RD, Van Straalen NM (2008) The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems. Ecol Lett 11:296–310. https://doi.org/10.1111/j.1461-0248.2007.01139.x

van der Plas F (2019) Biodiversity and ecosystem functioning in naturally assembled communities. Biol Rev 94:1220–1245. https://doi.org/10.1111/brv.12499

Wagg C, Bender SF, Widmer F, van der Heijden MGA (2014) Soil biodiversity and soil community composition determine ecosystem multifunctionality. Proc Natl Acad Sci 111:5266–5270. https://doi.org/10.1073/pnas.1320054111

Wagg C, Schlaeppi K, Banerjee S, Kuramae EE, van der Heijden MGA (2019) Fungal-bacterial diversity and microbiome complexity predict ecosystem functioning. Nat Commun 10:4841. https://doi.org/10.1038/s41467-019-12798-y

Wang X, Li FY, Wang Y, Liu X, Cheng J, Zhang J, Baoyin T, Bardgett RD (2020a) High ecosystem multifunctionality under moderate grazing is associated with high plant but low bacterial diversity in a semi-arid steppe grassland. Plant Soil 448:265–276. https://doi.org/10.1007/s11104-020-04430-6

Wang XG, Lü XT, Zhang HY, Dijkstra FA, Jiang YG, Wang XB, Lu JY, Wu Y, Wang ZW, Han XG (2020b) Changes in soil C:N: P stoichiometry along an aridity gradient in drylands of northern China. Geoderma 361:114087. https://doi.org/10.1016/j.geoderma.2019.114087

Wang C, Vera-Vélez R, Lamb EG, Wu J, Ren F (2022) Global pattern and associated drivers of grassland productivity sensitivity to precipitation change. Sci Total Environ 806:151224. https://doi.org/10.1016/j.scitotenv.2021.151224

Wang X, Zhang Q, Zhang Z, Li W, Liu W, Xiao N, Liu H, Wang L, Li Z, Ma J, Liu Q, Ren C, Yang G, Zhong Z, Han X (2023) Decreased soil multifunctionality is associated with altered microbial network properties under precipitation reduction in a semiarid grassland. iMeta 2: e106. https://doi.org/10.1002/imt2.106

Wolters V, Silver WL, Bignell DE, Coleman DC, Lavelle P, Van Der Putten WH, De Ruiter P, Rusek J, Wall DH, Wardle DA, Brussard L, Dangerfield JM, Brown VK, Giller KE, Hooper DU, Sala O, Tiedje J, Van Veen JA (2000) Effects of Global Changes on Above- and Belowground Biodiversity in Terrestrial Ecosystems: Implications for Ecosystem Functioning: We identify the basic types of interaction between vascular plants and soil biota; describe the sensitivity of each type to changes in species composition; and within this framework, evaluate the potential consequences of global change drivers on ecosystem processes. Bioscience 50:1089–1098. https://doi.org/10.1641/0006-3568(2000)050[1089:EOGCOA]2.0.CO;2

Wu Y, Li H, Cui J, Han Y, Li H, Miao B, Tang Y, Li Z, Zhang J, Wang L, Liang C (2024) Precipitation variation: a key factor regulating plant diversity in semi-arid livestock grazing lands. Front Plant Sci 15:1294895. https://doi.org/10.3389/fpls.2024.1294895

Yang X, Zhu K, Loik ME, Sun W (2021) Differential responses of soil bacteria and fungi to altered precipitation in a meadow steppe. Geoderma 384:114812. https://doi.org/10.1016/j.geoderma.2020.114812

Yang T, Zhong X, Chen J, Nielsen UN, Ochoa-Hueso R, Qu Y, Sui Y, Gao W, Sun W (2023) Ecosystem-level decoupling in response to reduced precipitation frequency and degradation in steppe grassland. Funct Ecol 37:2910–2926. https://doi.org/10.1111/1365-2435.14425

Yang X, Song W, Yang X, Yang T, Bao W, Wang C, Li J, Zhong S, Jiang Q, Li LJ, Sun W (2024) Microbial network structure, not plant and microbial community diversity, regulates multifunctionality under increased precipitation in a cold steppe. Front Microbiol 14:1349747. https://doi.org/10.3389/fmicb.2023.1349747

Zavaleta ES, Pasari JR, Hulvey KB, Tilman GD (2010) Sustaining multiple ecosystem functions in grassland communities requires higher biodiversity. Proc Natl Acad Sci 107:1443–1446. https://doi.org/10.1073/pnas.0906829107

Zeng Q, Li X, Dong Y, An S, Darboux F (2016) Soil and plant components ecological stoichiometry in four steppe communities in the Loess Plateau of China. CATENA 147:481–488. https://doi.org/10.1016/j.catena.2016.07.047

Zhai C, Han L, Xiong C, Ge A, Yue X, Li Y, Zhou Z, Feng J, Ru J, Song J, Jiang L, Yang Y, Zhang L, Wan S (2024) Soil microbial diversity and network complexity drive the ecosystem multifunctionality of temperate grasslands under changing precipitation. Sci Total Environ 906:167217. https://doi.org/10.1016/j.scitotenv.2023.167217

Zhang L, Wang J, Zhao R, Guo Y, Hao L (2022) Aboveground net primary productivity and soil respiration display different responses to precipitation changes in desert grassland. J Plant Ecol 15:57. https://doi.org/10.1093/jpe/rtab067

Zhang S, Chen Y, Zhou X, Zhu B (2024) Spatial patterns and drivers of ecosystem multifunctionality in China: Arid vs. humid regions. Sci Total Environ 920: 170868. https://doi.org/10.1016/j.scitotenv.2024.170868

Zhou Z, Wang C, Luo Y (2020) Meta-analysis of the impacts of global change factors on soil microbial diversity and functionality. Nat Commun 11:3072. https://doi.org/10.1038/s41467-020-16881-7

Zuo X, Sun S, Wang S, Yue P, Hu Y, Zhao S, Guo X, Li X, Chen M, Ma X, Qu H, Hu W, Zhao Y, Allington GRH (2023) Contrasting relationships between plant-soil microbial diversity are driven by geographic and experimental precipitation changes. Sci Total Environ 861:160654. https://doi.org/10.1016/j.scitotenv.2022.160654