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# Soil microbial community and functionality in response to degradation in alpine grassland: a meta-analysis

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## Abstract

Alpine grasslands have undergone severe degradation over the past half century. Investigating the changes in soil microbial communities is crucial for developing effective restoration strategies in degraded grassland. In this study, we synthesize data from 36 studies worldwide and show that soil microbial alpha and beta diversity did not show significant response to grassland degradation. However, we found increases in the relative abundance of bacteria related to adaptation to extreme environments, e.g., *Gemmatimonadetes*, but decreases in the relative abundance of sensitive fungi taxa (e.g., *Basidiomycota*) following degradation. Additionally, we observed a decline in soil functions related to nitrogen decomposition and fixation under heavy degradation. Overall, these findings advance our understanding of the impact of grassland degradation on soil microbial communities and their functions across a large scale. This study highlights the importance of restoring soil microbial communities in degraded grasslands to sustain soil function. Future research should develop suitable practices associated with microbial inoculation or regulation to facilitate grassland restoration.

## Introduction

Alpine grasslands are experiencing serious degradation due to the dual impacts of climate change and human activities. Soil microbial communities are of great importance for ecosystem functions, for instance, nutrient cycling. Usually, significant changes in soil microbial diversity and community structure will occur following degradation in temperate grassland. However, previous studies did not observe directional changes in soil microbial community composition in alpine grassland. For example, a previous study suggest that there was a decrease in bacterial richness after degradation (Wang *et al.*, 2021), while another study found bacterial richness did not show significant response to degradation, and the response of fungal richness varied by site (Che *et al.*, 2019). Given that the variance in microbial diversity and community composition induced by grassland degradation can dramatically suppress ecosystem functions (Breidenbach *et al.*, 2022), there is an urgent need to uncover the general response of soil microbes to degradation at a global scale when restoring degraded grasslands. We aimed to systematically assess the response of microbial communities to degradation in alpine grassland through Meta-analysis. This study will provide a scientific basis for formulating ecological protection and restoration strategies.

## Methods

### *Data collection*

We searched Web of Science (WoS, http://apps.webof knowledge.com/) in January 2024, with the following topic search ((grassland OR alpine) AND (micro* OR bacteria OR fungi OR AMF OR arbuscular mycorrhiza* fung* OR AM fung* OR AM symbiosis) AND (degrad* OR degenerat* OR deteriorat* OR deplet* OR restor* OR recover* OR reestablish* OR rehabilitat* OR renew*)). This search yielded more than 14,000 records. To avoid bias, the following criteria were used to screen studies: (1) The study was conducted on alpine grasslands and clearly described whether the studied grasslands were degraded or not. (2) Microbial community structure was determined using high-throughput sequencing, and at least calculated one soil microbial community metric, including alpha diversity (e.g., Chao1 or Simpson), beta diversity, and community structure was reported. (3) The study site was old-growth grassland rather than artificial grassland. (4) Sampling was conducted during the peak growing season. (5) Experiments with factors, such as N deposition, warming, drought, etc., were excluded. (6) Only field studies were selected, and laboratory incubation studies were not included. Finally, a total of 36 publications were remained for meta-analysis. For each study, we extracted means, sample size and standard deviation (SD) or standard error (SE) or 95% confidence interval (CI) if reported. If results were presented graphically, the software WebPlotDigitizer 4.6 (https://automeris.io/WebPlotDigitizer/) was used to digitize the data. Given that the levels of degradation likely affect the results, we captured the levels of degradation for each study. The degraded grasslands were considered to be lightly degraded if vegetation cover has decreased by more than 30%, and heavily degraded grasslands were those with a decrease of more than 50% in vegetation cover, compared to undegraded grasslands.

### *Calculation of the individual response ratios (RRs)*

We used the natural logarithm-transformed (ln) RR to calculate the response of variables for each case study:

$$RR = ln\frac{Xt}{Xc} \qquad (1)$$

where $\bar{X}_t$ and $\bar{X}_c$ are the means of the concerned variable in the degraded and undegraded grasslands, respectively.

Its variance (v) were calculated as:

$$v = \frac{SDt^2}{Xt^2} + \frac{SDc^2}{Xc^2} \qquad (2)$$

where $SD_t$ and $SD_c$ are the standard deviations of the variable in the degraded and undegraded grasslands, respectively.

### *Calculation of the overall RR*

All statistical analyses were performed in R (R Devel opment Core Team, 2024) using the R package *metafor* (Wolfgang Viechtbauer, 2010). The mixed-effect model was used to calculate the overall RR and the corresponding 95% confidence intervals of target variables. This model was also used to compare the RRs of variables between the light and heavy degradation by the omnibus test (QM). If the 95% confidence intervals for one RR overlapped with zero, then it was considered as an insignificant response to grassland degradation.

## Results

### *Grassland degradation did not alter microbial diversity but reduced microbial function.*

Grassland degradation did not affect alpha diversity (Chao1 and Simpson) and beta diversity of bacteria and fungi, but significantly changed community structure (Fig 1a). For instance, grassland degradation increased *Gemmatimonadetes*, while decreased *Basidiomycota* (Fig 1b). Grassland degradation had no effects on microbe biomass, enzyme activities of carbon (C) and nitrogen (N) decomposition. Grassland degradation did not affect the abundance of denitrification and nitrification genes, but decreased gene abundance related to nitrogen fixation (Fig 1c).

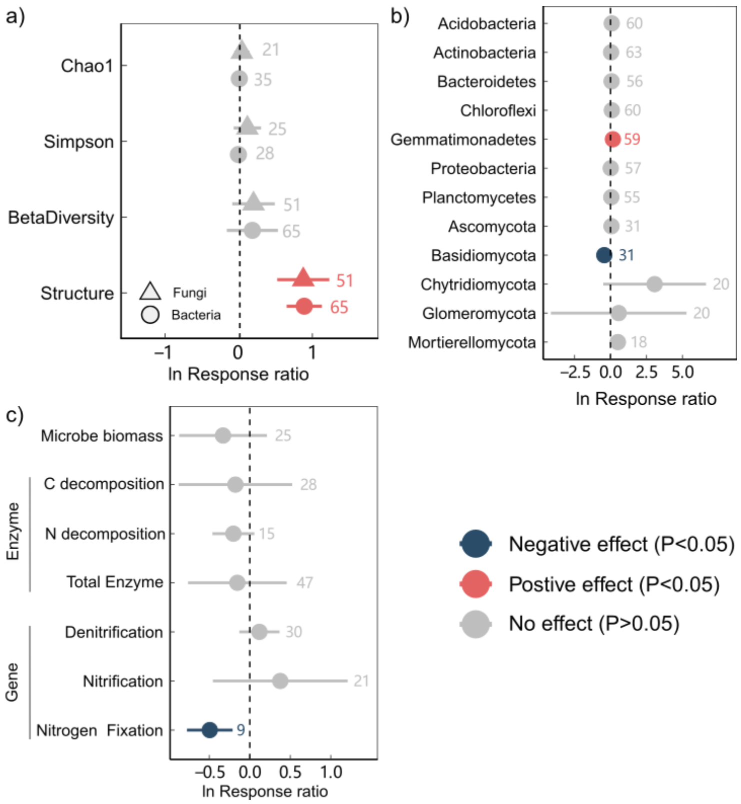

Fig. 1 Response ratios of soil microbial diversity (a), microbial community composition (b) and microbial function (c). Points are weighted means of RRs with 95% confidence intervals. The numbers on the right side of confidence intervals represent sample sizes. Positive mean values indicate increased variables induced by degraded grassland (red dots), while negative mean values indicate decreased variables induced by degraded grassland (blue dots). The intersection of confidence intervals and zero line indicates that there is no significant difference between the degraded and undegraded grassland (grey dots).

The RRs were significantly affected by the levels of degradation (Fig 2). As the levels of grassland degradation increased, we observed stronger RRs of enzyme activities (Fig 2a) and genes related to N cycling (Fig 2b). The overall and light degradation did not significantly affect C decomposition and N decomposition enzyme activities. However, when grasslands experienced heavily degradation, there were dramatic decreases in C and N decomposition enzyme activities (Fig 2a).

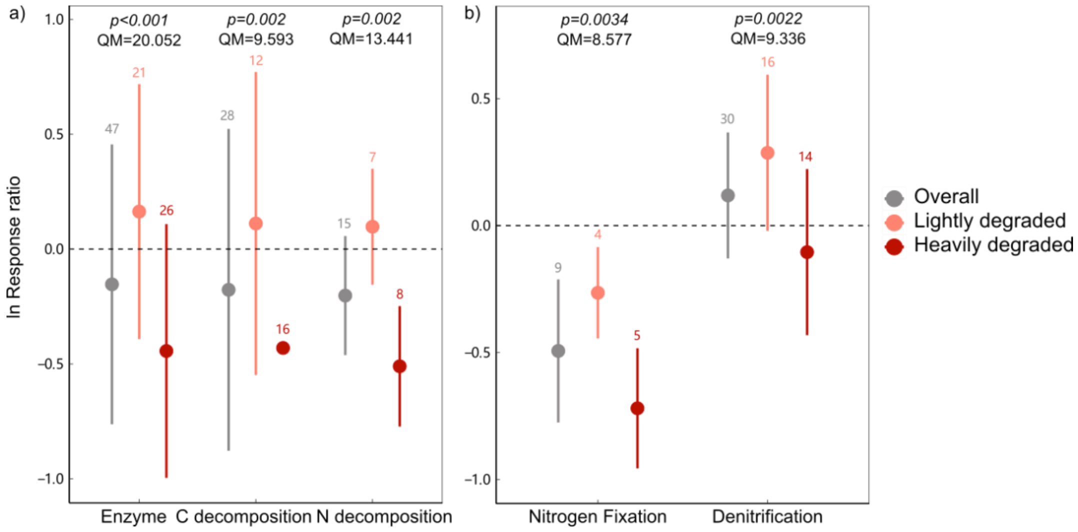

Fig. 2 Response ratios of soil enzyme activities (a) and functional genes (b) across grassland degradation levels. Points are weighted means of RRs with 95% confidence intervals. The numbers on the right side of confidence intervals represent sample sizes. Positive mean values indicate increased variables induced by degraded grassland (red dots), while negative mean values indicate decreased variables induced by degraded grassland (blue dots). The intersection of confidence intervals and zero line indicates that there is no significant difference between the degraded and undegraded grassland (grey dots).

## Discussion

Grassland degradation altered soil microbial community structure. Degraded grasslands had lower soil nutrient content and poorer physical soil properties, such as reduced soil organic carbon content and lower aggregate stability, compared to undegraded grasslands (Li *et al.*, 2021; Luo *et al.*, 2023). These changes may explain the increase in the relative abundance of *Gemmatimonadetes*, which can adapt to extreme environments, and the decrease in *Basidiomycota*, which are particularly sensitive to environmental disturbances. Grassland degradation did not significantly affect microbial diversity. Microbial communities can sustain biodiversity through species turnover. For instance, following grassland degradation, certain microbial taxa, such as drought-tolerant bacteria, can replace less resilient species, thus preserving overall species diversity within the community (Liu *et al.*, 2023).

Grassland degradation decreased nitrogen fixation genes. The abundance of efficient nitrogen-fixing bacteria, such as *Rhizobium* spp., decreased, while microorganisms with lower nitrogen-fixing efficiency became more dominant following grassland degradation (Zhang *et al.*, 2022). Heavy degradation suppressed C and N cycling indicated by a decrease in C and N decomposition genes. Key microbial groups, such as efficient N-fixing bacteria and decomposers, diminished and cannot be replaced by other microorganisms, leading to a decline in critical soil ecological functions, such as organic matter decomposition and nitrogen cycling (Breidenbach *et al.*, 2022). This suggest that practices stimulating C and N cycling or increasing nutrient content should be developed in further studies. Light degradation did not significantly affect other functions of soil microbes, suggesting that these functions may be redundant (Louca *et al.*, 2018).

Grassland degradation shifted soil microbial community composition, increasing stress-tolerant taxa while decreasing those that are sensitive. Additionally, heavy degradation weakened soil functions, suggesting the loss of functional redundancy. These findings highlight the importance of managing microbial communities in grassland restoration. Future restoration efforts should prioritize the recovery of microbial community and functional redundancy. Moreover, the severity of grassland degradation influenced the maintenance of soil functions, highlighting the importance of early intervention and effective land management to prevent irreversible loss of microbial functions.

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