Posted on 26 Sep 2023 | The copyright holder is the author/funder. All rights reserved. No reuse without permission. | https://doi.org/10.22541/au.169572701.10640197/v1 | This a preprint and has not been peer reviewed. Data may be preliminary.

Above- and belowground plant pathogens along elevational gradients: patterns and potential mechanisms

Ziyuan Lin[1], Fletcher Halliday[2], Peng Zhang[1], Xingxing Wang[1], Fei Chen[1], Anya Shi[1], Juanjuan Shi[1], Yao Xiao[1], and Xiang Liu[1]

[1]Lanzhou University
[2]University of Zurich

September 26, 2023

Abstract

Plant pathogens are important for ecosystem functioning and community assembly and respond to a variety of biotic and abiotic factors, which change along elevation gradients. Thus elevational gradients are a valuable model system for exploring how plant community, soil properties, and environmental factors influence pathogens. Yet, how these factors influence pathogens in nature remains poorly understood. We tested patterns and potential mechanisms of plant fungal pathogens along elevational gradients by combining a field survey in the Tibetan Plateau with a global meta-analysis. We found that increasing elevation was associated with a decrease in soil fungal pathogen richness but not foliar fungal disease symptoms. Elevation mainly related to soil fungal pathogen richness through abiotic factors. Whereas no evidence supported association between elevation and foliar fungal disease. The meta-analysis suggests some generality in the results of the field survey: elevation was associated with a decrease in soil fungal pathogen richness, but had no consistent relationship with foliar fungal disease or pathogens. Our study reveals distinct patterns of above- and belowground plant pathogen along elevation gradients and provides new insight into the potential mechanisms in shaping these patterns.

Title: Above- and belowground plant pathogens along elevational gradients: patterns and potential mechanisms

Abstract

Plant pathogens are important for ecosystem functioning and community assembly and respond to a variety of biotic and abiotic factors, which change along elevation gradients. Thus elevational gradients are a valuable model system for exploring how plant community, soil properties, and environmental factors influence pathogens. Yet, how these factors influence pathogens in nature remains poorly understood. We tested patterns and potential mechanisms of plant fungal pathogens along elevational gradients by combining a field survey in the Tibetan Plateau with a global meta-analysis. We found that increasing elevation was associated with a decrease in soil fungal pathogen richness but not foliar fungal disease symptoms. Elevation mainly related to soil fungal pathogen richness through abiotic factors. Whereas no evidence supported association between elevation and foliar fungal disease. The meta-analysis suggests some generality in the results of the field survey: elevation was associated with a decrease in soil fungal pathogen richness, but had no consistent relationship with foliar fungal disease or pathogens. Our study reveals distinct patterns of above- and belowground plant pathogen along elevation gradients and provides new insight into the potential mechanisms in shaping these patterns.

Key-words: Biodiversity-disease Relationship, Community Disease Proneness, Community Pathogen Load, Foliar Fungal Pathogen, Meta-analysis, Soil Fungal Pathogen

Posted on 26 Sep 2023 | The copyright holder is the author/funder. All rights reserved. No reuse without permission. | https://doi.org/10.22541/au.169572701.10640197/v1 | This a preprint and has not been peer reviewed. Data may be preliminary.

Introduction

Pathogens that cause plant diseases can maintain plant diversity by inducing both negative density dependence and life-history trade-offs (Allan et al., 2010; Cappelli et al., 2020), yet the factors that drive pathogen abundance and diversity remain poorly understood. Previous studies in agroecosystems suggest that environmental heterogeneity (shaped by multiple abiotic and biotic factors) can regulate plant pathogens (Stukenbrock and McDonald, 2008). However, whether variation in such factors drive geographic patterns of plant pathogens in natural ecosystem remains an open question.

Abiotic factors such as temperature and precipitation can affect plant pathogens both directly and indirectly through changes in host plant communities and soil properties (Liu et al., 2019). High temperatures often promote both foliar fungal diseases (e.g. Roy et al., 2004; Liu et al., 2019) and soil pathogens (Delgado-Baquerizo et al., 2020). Warming can benefit pathogen fitness by increasing pathogen survival, growth and transmission (Siebold and Tiedemann, 2013), extending the favorable time for pathogen growth (Roy et al., 2004), or narrowing the generation gap of pathogens (Bebber, 2015). Additionally, humidity can increase plant disease by promoting pathogens' spore germination and growth (Romero et al., 2021).

Abiotic factors are also predicted to influence the diversity, phylogenetic structure, and composition of host plant communities (Zhu et al., 2020), and could thereby indirectly affect plant pathogens (Halliday et al., 2021). In disease ecology, pathogens commonly increase in prevalence and severity with decreasing host diversity and corresponding changes in host density (e.g. via changes in host richness and evenness; Keesing et al., 2010; Halliday and Rohr, 2019). This negative biodiversity-disease relationship is potentially caused by reduced encounter rates, susceptible host regulation, and non-random host species loss (Keesing et al., 2006; Halliday, Rohr, et al., 2020). Accumulated empirical evidence from grasslands and temperate and subtropical forests supports the existence of negative biodiversity-disease relationships in natural communities (e.g. Mitchell et al., 2002; Rottstock et al., 2014; and Liu, Chen, et al., 2020 for a meta-analysis; but see Halliday et al 2017, 2020). Furthermore, changes in diversity are often accompanied by compensatory shifts in the density of component host species in grasslands (Mitchell et al., 2002). The density-dependent transmission of foliar fungal diseases causes communities with higher host density to suffer more seriously from disease at the host population level (Burdon and Chilvers, 1982), and soil pathogens may rely on plant biomass since more biomass will provide more nutrient substance and greater chance for pathogenicity (Liu et al., 2021). Furthermore, disease proneness (i.e. expected community pathogen load based on constituent host plant species) can explain why host diversity loss is strongly associated with increased foliar fungal disease in alpine meadows (Liu et al., 2017). A plant species with high disease proneness (i.e. possess good growth but weak defensive abilities) can harbor more pathogens, and thus a host community with a higher proportion of more disease-prone species is expected to have a higher community pathogen load (Liu et al., 2017).

In addition to plant community characteristics, soil properties can also potentially regulate plant pathogens. Both foliar and soil pathogens are likely to benefit from high soil nutrients in both agro- and natural ecosystems (Huber and Watson, 1974; Liu et al., 2017). Soil nutrients can promote pathogens by increasing tissue nitrogen concentration (Veresoglou et al., 2013), which is one of the most important limiting factors for many pathogens, especially for those that extract nutrients only from living plant tissues (i.e. biotrophic pathogens; Liu, Lu, et al., 2020).

Despite the well-characterized impacts of single abiotic or biotic factors on plant pathogens, how these soil-, plant community-, and environment mediated effects combine to generate patterns of above- and belowground plant pathogens across biogeographic gradients remains poorly understood. For instance, a previous study found that fungal diseases on *Phragmites australis* increased with latitude in North America, indicating that geographical gradients were associated with the distribution of pathogens (Allen et al., 2020). However, studies along latitude are massive logistical undertakings, and their results are easily affected by the potential confounding factors of geology and biogeographic history (Halbritter et al., 2018). Compared to other biogeographical gradients, elevational change generates highly variable ecological conditions (including soil, plant community, environment) at a relatively small spatial scale (Rowe, 2009), providing an excellent 'natural laboratory' to study how biotic and abiotic factors affect plant pathogens (Halliday et al., 2021).

Posted on 26 Sep 2023 | The copyright holder is the author/funder. All rights reserved. No reuse without permission. | https://doi.org/10.22541/au.169572701.10640197/v1 | This a preprint and has not been peer reviewed. Data may be preliminary.

Here, following this framework, we considered soil-, environment-, and plant community-mediated effects to explore patterns and potential mechanisms of plant pathogens along elevation gradients. However, quantifying plant pathogen communities is complicated by the fact that potential plant pathogens do not always cause disease (i.e. the plant disease triangle; Liu and He, 2019). To overcome this challenge, we measured plant pathogen communities in two ways. We measured foliar fungal disease as a quantitative measurement of the disease severity (i.e. relative abundance, rather than absolute abundance) of pathogens that are currently causing disease on plants. Leaves are readily surveyed for disease, allowing for accurate and reliable surveys of pathogen relative abundance in plant communities. However, the disease observed on a leaf is only one small part of the total pathogen potential of an ecological community. In contrast with leaves, soils can serve as a reservoir of plant pathogens capable of causing disease both above-and below ground, even when those pathogens are not currently causing disease on a plant (Delgado-Baquerizo et al., 2020). Surveying the richness and relative abundance of soil fungal pathogen communities using sequencing-based approaches has become a powerful approach to assess the pathogen potential of a community (van Agtmaal et al., 2017).

We combined these two complementary approaches by measuring foliar fungal disease, soil fungal pathogens, plant community characteristics (richness, evenness, biomass and proneness), environmental conditions, and soil properties along an elevational gradient in an alpine meadow in the northeastern Qinghai-Tibetan Plateau to explore patterns of above- and belowground plant pathogens. We sought to answer the following questions: (*i*) how do abiotic and biotic factors (soil properties, plant community, environment conditions, above- and belowground plant pathogens) change along elevational gradients?; (*ii*) how do soil properties, plant community and environmental conditions affect above- and belowground plant pathogens?; and (*iii*) what is the relative importance of the plant community-, environment- and soil-mediated effects for the correlation between elevation and above- and belowground plant pathogens? We additionally performed a systematic meta-analysis to explore the correlation between elevation and above- and belowground plant pathogens and to assess the generality of our main conclusions.

Materials and methods

Field survey along an elevational gradient

Study site and plot establishment

We conducted our survey along an elevational gradient ranging from 3200 m to 4000 m a.s.l. on the south slope of the Qilian Mountains in Menyuan County, located on the northeastern Qinghai-Tibetan Plateau in China (Fig. S1.1). The region has a continental monsoon climate, with a 6-month growing season from mid-April to mid-October. The study took place in alpine meadow habitat, dominated by some perennial herbaceous genera (e.g. *Gentiana* , *Kobresia* , *Poa* and*Saussurea*), with the species composition shifting with elevation. We established thirty 0.5 × 0.5 m plots (6 replicates for each of five elevations) along the elevational gradient at 3200 m (37°36'39" N, 101°18'16" E), 3400 m (37°39'58" N, 101°20'20" E), 3600 m (37°41'47" N, 101°21'34" E), 3800 m (37°42'13" N, 101°22'14" E) and 4000 m (37°42'29" N, 101°22'27" E). We randomly placed the plots with at least a 10 m buffer zone between two adjacent plots at each elevation.

Plant and soil sampling and climate monitoring

In early August 2020, we harvested all the plant aboveground parts at ground level and sorted them to species at the plot level. We then dried plants at 65 for 48 hours to constant mass and weighed them to 0.01 g; we then summed the dry biomass of each species from each plot to quantify aboveground biomass (hereafter '*AGB* ', see Table 1 for abbreviations). We also collected four soil cores (5 cm in diameter and 10 cm in depth) in the four corners of each plot and pooled them to form one sample per plot. We flushed all fine roots collected from each sample with water and then dried them to measure plant belowground biomass (*BGB*). We measured soil moisture content (W; %) gravimetrically after 5 h of desiccation at 120 and used a pH analyzer and a conductivity analyzer to measure the soil pH (pH) and soil conductivity (C; ms/s), respectively. We extracted 5 g of fresh soil with 50 ml 0.2 M KCl for 1 h at 60 rev s^{-1} using a shaker, then measured nitrate-nitrogen (NO_3^-; mg/kg) and ammonium-nitrogen (NH_4^+; mg/kg) using an auto-analyzer

(AA3, Bran-Luebbe, Germany).

We placed a Temperature-Humidity Recorder Cos-03-0 (Renke Control Technology Co., Ltd., Jinan, Shandong, China) at each elevation, which continuously monitored ambient temperature and humidity every 60 seconds for 72 hours in early August 2020. We then calculated the mean daily temperature (MDT) and mean daily humidity (MDH) for each elevation. Given the relatively short-term temperature and humidity may not represent the environmental conditions that were present during pathogens development, we only test the associations between elevation with MDT andMDH (thereby confirming whether the elevational gradient can serve as a temperature and humidity gradient), but we did not included these measurements in other analyses.

Bioinformatic analysis

We extracted DNA from the soil samples and implemented PCR amplification. We then constructed our library and assessed its quality. We operated the Illumina NovaSeq 6000 platform to quantify and sequence the constructed library for soil fungi. We then used non-clustering direct denoising to generate operational taxonomic units (OTUs) and identified the fungus at the genus level (Detailed supplementary methods in Section S1.1 in Supporting information).

Measurement of above- and belowground plant pathogens

For foliar fungal diseases, we recorded foliar fungal disease severity following the methods provided in Liu et al. (2017). In brief, we visually recorded disease severity (i.e. % leaf area covered by fungal lesion; V_i) from five leaves randomly selected from five individuals for each plant species in each plot. We recorded all available leaves for species with less than 25 leaves. We then calibrated our records by comparing diseased leaves to reference images of known disease severity. We identified the foliar fungal pathogens using an Olympus CX33 light microscope (Shinjuku, Japan) following identification manuals, including the Fungal Identification Manual (Wei, 1979), Plant Disease Diagnosis (Lu, 1997), and also previous studies in this area (Zhang, 2009; Liu et al., 2019; Liu, Lu, et al., 2020). We defined community pathogen load (PL) following Mitchell et al. 2002 as:

$$PL = \frac{\sum_{i=1}^{S} b_i V_i}{\sum_{i=1}^{S} b_i}$$

where S was the total number of plant species in a certain plot, and b_i was the aboveground biomass of plant species i . We then defined a 'disease proneness index' (hereafter 'P_i ') for each species as the average severity index (V_i) across 30 plots of plant species i . We then calculated a 'community proneness index' (hereafter '*Proneness* ') for each plot by calculating a plant aboveground biomass-weighted average of the $P_i$for each plot (Liu et al., 2017):

$$Proneness = \frac{\sum_{i=1}^{S} b_i P_i}{\sum_{i=1}^{S} b_i}$$

where *Proneness* was the expected community pathogen load based on constituent host plant species, which was measured independent of the actual disease in a given plot (Liu et al., 2017). Specifically, each species in each plot was assigned a value of disease proneness based on averaging its disease severity (V_i) in this plot and weighted by its aboveground biomass. Despite a similar mathematical formula for PL and*Proneness* , these values represent two distinct characters of plant community (the actual amount of disease in a community and the amount that would be expected based on the identity of species present and their relative abundances alone). PL and *Proneness* are not always correlated with one another (e.g. Liu et al., 2019), and are often used together in disease ecology studies (e.g. Mitchell et al., 2002; Johnson et al., 2013; Liu et al. 2017; Liu et al., 2019). We log-transformed community pathogen load (PL) and disease proneness index (*Proneness*) to achieve normality of residuals in the following analysis.

Posted on 26 Sep 2023 | The copyright holder is the author/funder. All rights reserved. No reuse without permission. | https://doi.org/10.22541/au.169572701.10640197/v1 | This a preprint and has not been peer reviewed. Data may be preliminary.

Posted on 26 Sep 2023 | The copyright holder is the author/funder. All rights reserved. No reuse without permission. | https://doi.org/10.22541/au.169572701.10640197/v1 | This a preprint and has not been peer reviewed. Data may be preliminary.

For soil fungal pathogens, we defined fungal taxa as putative plant fungal pathogens when they include any pathogenic species which were reported to induce any plant disease symptoms (e.g. canker, rot, leaf spot, blight, rust and mildew) (Liang et al., 2016), as determined by references to published data (Tedersoo et al., 2014), paper in*ISI Web of Science* (if any paper reported their pathogenicity) and the FUNGuild algorithm (Nguyen et al., 2015). Indeed, even if some genera with mixed feeding strategies (e.g. parasitic, mutualistic and saprophytic) were presumed as pathogens based on the above method in this study, they still represent pathogen potential. For instance, genera belong to Dothideomycetes (e.g. *Alternaria* ,*Epicoccum* , *Fusicladium*), Leotiomycetes (e.g. *Coma* ,*Erysiphe* , *Scytalidium*), Sordariomycetes (e.g.*Fusarium* , *Neonectria* , *Valsa*) and other classes were presumed as plant pathogenic. All plant pathogenic genera identified in the field study are listed in Table S2.1. We then calculated the accumulated OTU number of soil fungal pathogens (*sfpOTUs* ; OTU richness of soil fungal pathogens), and also the relative abundance of soil fungal pathogens (*sfpRA* ; copy number of soil fungal pathogens divided by the total number of copy number of soil fungus) for each sample. We log-transformed soil fungal pathogen relative abundance (*sfpRA*) to achieve normality of residuals in the following analysis.

Limitations and caveats in methodology

This study includes three complementary measurements of plant pathogens: the relative abundance of pathogens causing foliar disease, the relative abundance of soil pathogens (*sfpRA*), and the richness of soil pathogens (*sfpOTUs*). We measured damaged on the leaves and fungal pathogen communities in the soil. Although these two approaches do not provide identical assessments of pathogen richness and disease outcomes in foliar and soil- compartments, we believe that this approach is justified, as it reflects the most commonly used approach in these two respective fields of research, and represents comprehensive characteristics of both above- and belowground fungal plant pathogen communities, and thereby provides a more comprehensive understanding of how pathogen communities respond to changing environmental conditions. Furthermore, although the measurements are not identical, they are often positively correlated with one another, and this correlation often transcends study systems (Rottstock et al., 2014; Liu et al., 2016; Halliday et al., 2017; Halliday et al., 2020b). Therefore, we feel confident that the distinct measurements in our study can provide insight into the biogeographic pattern of pathogens across elevation gradients.

In this study, we measured foliar fungal diseases and soil pathogens as representatives of above- and belowground plant pathogens respectively to bring together research from two different fields that tend to study pathogens in different ways, which provides complementary information using complementary measurement approaches. However, we applied these measurements with caveats that visual assessment for foliar fungal diseases does not include all pathogens and can result in an incomplete assessment of pathogen diversity, while sequencing-based assessment for soil fungal pathogens is not directly related to any particular disease outcome. Overall, these measurements still reflect the most commonly used approach in these two respective fields of research, and thereby provide a more comprehensive understanding of how pathogen communities respond to changing environmental conditions.

Root diseases resulted by root-borne pathogens can also affect host mortality, growth and productivity, and further influence ecological succession and biogeochemistry process, thereby regulating ecosystem functioning (e.g. Hansen & Goheen, 2000; Healey et al., 2016). Future studies could combine surveys of foliar and root fungal diseases by both visual measurements and sequencing to more comprehensively explore the responses of above- and belowground plant pathogens to abiotic and biotic factors and their impacts on ecosystem functioning.

The results of the field survey might be sensitive to limitations of the empirical approach. For soil fungal sequencing, fungal ITS1 region may suffer from certain taxonomic biases, like high proportion of mismatches and biased amplification of certain fungal taxa (e.g. basidiomycetes; Tedersoo & Lindahl, 2016). However, fungal ITS1 region indeed possess some advantages which outperformance to other regions, for example, it is easily discriminate fungal taxa from plants and provides wider richness and taxonomic coverage (Mbareche et al., 2020). Future studies could overcome these challenges by incorporating multiple sequencing regions, and using more advanced methods (e.g. exact sequence variants (ESVs), which generate a greater resolution

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

than OTU-based methods; Mbareche et al., 2020). In fact, quantitative PCR is a good choice to calculate the abundance of pathogens (Tellenbach et al., 2010), although the small datasets prevent us from further analyses regarding the absolute abundance of pathogens. In fact, unlike the foliar fungal disease, the relative abundance of soil pathogen profiles cannot indicate the absolute abundance of pathogens. Hence, we can only conclude that elevation had no associated with the relative abundance of soil pathogens. Our empirical results also stem from a single location in a single year, and thus our results might be sensitive to local environmental conditions that are characteristic of the particular year of sampling. Although our empirical results largely agree with the results of the meta-analysis, this does not negate the limitations of the empirical study (i.e., relatively small sample size, single gradient, single year). Future studies conducted over multiple elevational gradients and multiple years remain the gold standard for empirical field surveys along elevational gradients. Large-scale and long-term studies of biotic and abiotic drivers of disease across environmental contexts remain a pressing need if ecologists want identify the underlying effects of temporal dynamics and spatial heterogeneity in the community ecology of infectious disease.

Statistical analysis

(i) Changes of abiotic and biotic factors along elevational gradients

We conducted principal component analysis (PCA) to summarize soil properties (W, pH, C, NO_3^- and NH_4^+) using the "vegan" package (v. 2.5.7; Oksanen et al., 2020) (Fig. S1.2). We then calculated the Spearman rank-order correlation between *Soil PCA1* and each of the soil properties (W, pH, C, NO_3^- and NH_4^+) (Fig. S1.2). We also calculated the plant species richness (*SR*) and Pielou's evenness index (*Evenness*) using the "vegan" package for each plot. To test correlations among various variables, we plotted the correlation matrix for all biotic or abiotic variables (*Elevation* , *SR* ,*Evenness* , *Proneness* , *AGB, BGB, MDT, MDH* and*Soil PCA1*), and we then calculated the Pearson's correlation between these variables and *PL* /various soil pathogen indices (Fig. S1.3a). We also conducted Mantel tests based on "Bray-Curtis" distance between *sfpOTUs* and biotic or abiotic variables using the "ggcor" package (v. 0.9.8.1; Huang et al., 2020) (Fig. S1.3b).

To minimize the influence of the potential spatial autocorrelation on the results, we used the respective coordinates of each sample plot to generate a spatial matrix. Specifically, given the sample plots were distributed along a cambered mountain slope which approximated to the spherical surface, we calculated the spatial matrix based on spherical correlation structure (i.e. "corSpher" class) using the "nlme" package (v. 3.1-152; Pinheiro et al., 2021). We then introduced the spatial matrix into a series of linear mixed-effects models with five elevations as a random effect in following analyses, using the "nlme" package. We set *Elevation* as independent variables in a series of linear mixed-effects models to test its associations with various community-level indices (*SR* , *Evenness* , *Proneness* ,*AGB* and *BGB*) and soil properties (*SoilPCA1*), respectively. At the plant community level, we set *Elevation* as the independent variable and *PL* and soil pathogen indices (*sfpOTUs* and*sfpRA*) as response variables in a series of linear mixed-effects models to test the direct correlations between *Elevation* and above- and belowground plant pathogens.

For the soil fungal pathogen community, we conducted permutational multivariate analysis of variance (PERMANOVA) to test the compositional difference of soil fungal pathogens along the elevational gradient. A significant result of PERMANOVA supports the hypothesis that pathogen communities change along elevational gradients.

(ii) Factors affecting above- and belowground plant pathogens

We set various community-level indices (*SR* , *Evenness* ,*Proneness* , *AGB* and *BGB*), and soil properties (*Soil PCA1*) as independent variables in a series of linear mixed-effects models to test their effects on *PL* , *sfpOTUs*and *sfpRA* , respectively. We using "MuMIn" package (v. 1.47.1; Bartoń, 2022) to conducted full model selections based on a series of linear mixed-effects models for *PL* , *sfpOTUs* and*sfpRA* , respectively. We then extracted the effect sizes with 95% confidence intervals from weighted average standardized coefficients from models with ΔAICc < 4 based on model selections, and compared these models with the null model (i.e. the intercept-only model) based on Akaike's information criterion corrected for small sample sizes

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

(AIC_c) using the "MuMIn" package. We also calculated the log-likelihood (LL) and AIC_c based parameters: change in AIC_c relative to the top-ranked model (ΔAIC_c), AIC_c weight (w AIC_c) and the percent deviance explained (De) (Burnham et al., 2011), to estimate their possibilities of being the best predictor of PL and various soil pathogen indices. When the ratio of$w$ AIC_c of predictor to w AIC_c of null model more than 1.5, it indicates that the corresponding variable is associated withPL , $sfpOTUs$ and $sfpRA$ (Burnham et al., 2011).

(iii) Plant community-, environment- and soil-mediated relationships between elevation and above- and belowground plant pathogens

To explore how soil properties, plant community and environment conditions mediated the relationship between elevation and above- and belowground plant pathogens, we used the "piecewiseSEM" package (v. 2.1.2; Lefcheck, 2016) to build a structural equation model (SEM) based on a series of linear mixed-effects models to test the relationship between elevation and PL and $sfpOTUs$ including paths that were mediated by the plant community and environment (i.e. elevation may first influence plant community characters, soil properties and environmental conditions, which then indirectly change plant fungal pathogen communities) (Halliday et al., 2021; Fig. 2a). We did not include MDT in the SEM, given that there was a strong collinearity between MDT and *elevation* (Pearson' r= 0.985). We calculated the standardized path coefficients (scaled by their mean and standard deviation) and corresponding significance (P values) for each path of the final models. Statistically significant path(s) ($P < 0.1$) highlight potentially important mechanisms generating relationships between elevation and above- and belowground plant pathogens. We used the Fisher's C test to test the goodness-of-fit of the SEM. We then used partial residual plots to interpret the relationships highlighted in the model (Grace, 2006). In brief, we extracted the residuals of each variable in SEM and then set the residuals of plant pathogen related variables (i.e. $PL$and $sfpOTUs$) as functions of residuals of explanatory variable (i.e. *Elevation* , *Soil PCA1* , *Evenness* and*Proneness*) in a series of linear models (Grace, 2006). Significant results of residual analysis correspond to significant paths in the SEM.

Meta-analysis

Data collection

Our empirical approach to study the relationship between elevation and plant pathogens might be sensitive to the relatively small number of replicates across a single environmental gradient in a single year, as trophic interactions can vary across space and time, and are often context dependent (Roslin et al., 2017; Liu, Chen, et al., 2020). Therefore, to test the generality of our main results from the field survey, we conducted a systematic literature search in *ISI Web of Science* and China National Knowledge Infrastructure (www.cnki.net). We searched for research on foliar fungal pathogen [(fungal OTU* OR fungi OTU* OR fung* operational taxonomic unit OR fung* abundance OR fung* richness) AND (elevation* OR altitud*) AND (folia* OR leaf OR leaves)], foliar fungal diseases [(plant disease* OR pathogen* OR infect* OR epidemic*) AND (inciden* OR prevalen* OR load* OR severity OR occur* OR abundance) AND (elevation* OR altitud*) AND (folia* OR leaf OR leaves)] and soil plant pathogens [(fungal OTU* OR fungi OTU* OR fung* operational taxonomic unit OR fung* abundance OR fung* richness) AND (elevation* OR altitud*) AND (soil OR belowground OR underground)]. We finally identified 41 papers (providing a total of 62 effect sizes) that met our criteria (Fig. S1.1; Table S1.1): (i) focused on the relationship between elevation and foliar fungal diseases and foliar/soil plant pathogens in nonagricultural ecosystems; and (ii) reported sample sizes greater than three. Detailed process for literature screening and basic information of 41 papers were provided in Fig. S1.4 and Table S1.1.

We collected the OTU table for studies on foliar and soil plant pathogens, identified the putative plant pathogens according to the aforementioned methods, and calculated $ffpOTUs$ (i.e. foliar fungal pathogen OTU richness), $sfpOTUs$ and $sfpRA$ for each study. All plant pathogenic genera identified in the meta-analysis are listed in the Table S2.1. We extracted the sample sizes and Pearson's correlation coefficients (r) from the main text, tables, figures (using WebPlotDigitizer v. 4.4; Rohatgi, 2020), or raw data. We also recorded background information on location and the elevation range of sampling (as highest sampling

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

elevation minus lowest sampling elevation) from original papers, then we extracted the mean annual temperature and mean annual precipitation of the lowest elevation location for each study based on the WorldClim database (Fick and Hijmans, 2017).

Effect sizes

We calculated effect sizes as the Fisher's z -transformation of Pearson's correlation coefficients (r) (Rosenberg et al., 2013):

$$Z = \left(\frac{1+r}{1-r}\right)$$

The corresponding variance for each Z was estimated as:

$$Var = \frac{1}{n-3}$$

where n is the sample size. Positive values of Z indicate that increasing elevation is associated with increases in foliar fungal disease or foliar/soil pathogens, while negative values indicate decreases.

Statistical analysis

We used the *metafor* package (v. 3.0.2; Viechtbauer, 2010) to calculate the mean effect size of elevation on *ffpOTUs* , foliar fungal diseases, *sfpOTUs,* and *sfpRA* , with 'study' nested in 'paper' as random effects (Nakagawa et al., 2017). The effect size (Z) was considered to be significant when the 95% confidence interval of the mean did not include zero (Lajeunesse, 2013). We tested the overall effect of elevation on *ffpOTUs* , foliar fungal diseases, *sfpOTUs* , and *sfpRA* , and respective effect in forest and grassland ecosystem for foliar fungal disease (due to insufficient study in grassland ecosystems for other response variables). We then introduced mean annual temperature, mean annual precipitation, latitude and elevation to test the context dependence of effect size (Z). The amount of heterogeneity explained by each variable was estimated by the Q_{m} statistic and its corresponding P value (Viechtbauer, 2010). For assessing the potential publication bias, we conducted Kendall's rank test for funnel plot asymmetry (Borenstein et al., 2009), and also ran a meta-regression between effect size (Z) and studies' publication years/journal impact factors. All statistical analyses were conducted using R v. 4.1.1 (R Development Core Team, 2021).

Results

Field survey along an elevational gradient

(i) Changes of abiotic and biotic factors along elevational gradients

For soil properties, the first principal component (*Soil PCA1*) explained 41.63% of the total variance and *Soil PCA1* was positively correlated with pH, C, and NO_3^- and negatively correlated with W and NH_4^+ (Fig. S1.2).

We found 73 plant species in total along the elevational gradient in the alpine meadow. *Elevation* had a strong correlation with *MDT*rather than *MDH* , showed a highly collinearity between them (Fig. S1.3). Due to this collinearity, we feel confident interpreting elevation as a proxy for temperature. Besides, increasing*Elevation* was associated with a significant decrease in $AGB (P = 0.006$, Marginal$R^2 = 0.733)$, SR ($P = 0.028$, Marginal $R^2 = 0.482$) and *Soil PCA1*($P = 0.038$, Marginal $R^2 = 0.610$) (Fig. S1.5).

At the plant population-level, fungal leaf spot was the most commonly identified symptom, compared with others (e.g. rusts, blight, smuts and downy mildew; Table S1.3). Multiple species belonging to several genera (i.e. *Alternaria* , *Ascochyta* , *Peronospora* ,*Puccinia* , *Trichometasphaeria* , *Urosystis* and*Ustiligo*) were identified as pathogens associated with the observed foliar fungal diseases (Table S1.2). Host species varied inV_i from 0 to 27.78 (*Deschampsia caespitosa*) (Table S1.2). With respect to soil pathogens, we identified

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

putative plant fungal pathogens belonging to 106 genera. Soil plant fungal pathogens occupied an average of about 7.72% relative abundance, compared to all soil fungi across all plots.

For foliar fungal diseases and soil pathogens, increasing *Elevation* was associated with a significant reduction in *sfpOTUs* ($P = 0.020$, Marginal $R^2 = 0.433$), but not *PL* ($P = 0.403$, Marginal $R^2 = 0.034$) or *sfpRA* ($P = 0.973$, Marginal $R^2 < 0.001$) (Fig. S1.6). PERMANOVA results indicated that the composition of soil fungal pathogens varied with elevation ($F_{4,25} = 2.744$, $P < 0.001$) (Table S1.3).

(ii) Factors affecting above- and belowground plant pathogens

Among different plant community-level indices, environmental factors and soil properties, linear mixed-effects models result showed that only *Proneness* ($P < 0.001$, Marginal $R^2 = 0.348$) had a significant positive effect on *PL* , while no variable had a significant effect on *sfpOTUs* and *sfpRA* (Fig. S1.7, S1.8, S1.9). Among various univariable and multivariable linear mixed-effects models, only univariable of *Pronenss* ($P < 0.001$) and *Eveness* ($P = 0.043$) had significant effects on *PL* , and the combination of these factors (*Proneness* + *Eveness*) provided the best fit ($AIC_c = 82.510$, $w\ AIC_c = 0.269$, $R^2 = 0.438$) compared to the null model ($AIC_c = 93.719$, $w\ AIC_c = 0.001$), accounting for *c.* 44% of the deviance explained in *PL* (Fig. 1a; Table S1.4). *sfpOTUs* were directly associated with *Elevation* ($P = 0.005$), and strongly influenced by *Soil PCA1* ($P = 0.021$) (Fig. 1b). The combination of *Elevation* and *Soil PCA1* provided the best fit ($AIC_c = 76.842$, $w\ AIC_c = 0.294$, $R^2 = 0.534$) for *sfpOTUs* among various multivariable models (Table S1.5). *sfpRA* had no statistically significant association with any ecological variable tested, and was therefore not included in the SEM (Table S1.6)

(iii) Plant community-, environment- and soil-mediated relationships between elevation and above- and belowground plant pathogens

The final SEM (Fisher's C = 11.855, $P = 0.158$, *d.f.* = 8) revealed that increasing *Elevation* was associated with a significant decrease in *Soil PCA1* (standardized path coefficient β = -0.768, $P = 0.038$) (Fig. 2b; Table S1.7). While *Elevation* showed strong negative direct correlation with *sfpOTUs* (β = -1.109, $P = 0.015$) but not *PL* (β = -0.159, $P = 0.563$). Furthermore, *Evenness* (β = -0.296, $P = 0.082$) and *Proneness* (β = 0.665, $P < 0.001$), rather than *Soil PCA1* (β = -0.180, $P = 0.482$), were associated with variation in *PL* . In contrast, significant reduction in *sfpOTUs* was associated with increasing *Soil PCA1* (β = -0.540, $P = 0.026$), but not *Evenness* ($\beta = 0.063$, $P = 0.672$) or *Proneness* ($\beta = 0.097$, $P = 0.529$) (Fig. 2b; Table S1.7). The final SEM accounted for 45.772% of variation in *PL* and 55.855% of variation in *sfpOTUs* . In addition, the residual plots also showed that changes in both *Proneness* and *Evenness* were associated with variation in *PL* , while increasing *Soil PCA1* was associated with a significant decrease in *sfpOTUs* (Fig. S1.10). This indicates the important role of plant community in explaining variation in *PL* , and highlights the indirect connection between elevation and belowground plant pathogens via abiotic factors.

Meta-analysis

Kendall's rank test for funnel plot asymmetry indicated that no publication bias existed for all the tests ($P > 0.05$) (Table S1.8; Fig. S1.11), and there was no significant correlation between effect size (Z) and studies' publication years/journal impact factors ($P > 0.05$) (Fig. S1.12).

Overall, elevation was not associated with foliar fungal pathogen OTU richness ($Z \pm 95\%$ CI = -0.157 $\pm$ 0.263, $P = 0.242$), foliar fungal diseases ($Z \pm 95\%$ CI = -0.047 $\pm$ 0.242, $P = 0.703$) or *sfpRA* ($Z \pm 95\%$ CI =-0.101 $\pm$ 0.183, $P = 0.281$), whereas increasing elevation was significantly associated with *sfpOTUs* ($Z \pm 95\%$ CI = -0.257 $\pm$ 0.172, $P = 0.003$) (Fig. 3; Fig. S1.13; Table S1.9). Elevation was not significantly associated with foliar fungal disease in forest ($Z \pm 95\%$ CI = -0.127 $\pm$ 0.440, $P = 0.571$) or grassland ($Z \pm 95\%$ CI = 0.023 $\pm$ 0.153, $P = 0.772$) ecosystems (Fig. 3; Fig. S1.13; Table S1.9). There was no significant correlation between the climatic variables (mean annual temperature, mean annual precipitation) and the effect size (Z) on *ffpOTUs* , foliar fungal diseases, *sfpOTUs* or *sfpRA* (Fig. S1.14; Table S1.10). Nevertheless, increasing absolute latitude was associated with decreasing effect size (Z) on *ffpOTUs* ($Q_m = 4.231$, $P = 0.040$) but not foliar fungal disease ($Q_m = 0.364$, $P = 0.547$), *sfpOTUs* ($Q_m = 1.264$, $P = 0.261$) or *sfpRA* (Q

$_{m}$ = 1.647, P = 0.199) (Fig. S1.14; Table S1.10). Furthermore, studies conducted over larger elevational ranges tended to observe stronger negative relationships between elevation and *sfpOTUs*(Q_{m} = 4.572, P = 0.033) and *sfpRA*(Q_{m} = 4.469, P = 0.033), though the elevational range of sampling did not moderate this relationship for*ffpOTUs* (Q_{m} = 0.150, P = 0.698) or foliar fungal disease (Q_{m} = 0.415, P = 0.520) (Fig. S1.14; Table S1.10).

Discussion

Changes of abiotic and biotic factors along elevational gradients

In our field survey, abiotic and biotic factors (soil properties, plant community characters and environment conditions) greatly varied along elevational gradients, consistent with past studies (e.g. Lomolino, 2001), and those abiotic and biotic factors, in turn, explained relationships between elevation and above- belowground plant pathogens. By integrating this field survey with a systematic meta-analysis, we showed a general, negative association between elevation and soil pathogen richness, but did not find support for a general association with foliar fungal pathogen richness, foliar fungal diseases or soil pathogen relative abundance. Our meta-analysis further suggests that the elevation range of sampling may potentially shape which relationships are observed between elevation and soil fungal pathogens. Larger elevation gradients mean that studies are comparing among different habitat types with different sets of species, whereas smaller elevation gradients are usually making comparisons across environmental conditions within a single habitat type. These results provided important evidence that elevational patterns of plant pathogens are consistent despite great variation in plant and pathogen species pools.

In our field survey, we did not find any evidence supporting a direct association between elevation and foliar fungal diseases at either the host population or community levels. These results are inconsistent with a previous study in the Swiss Alps along a 1101 m elevational gradient, which found that elevation not only associated with disease directly, but also indirectly regulated plant diseases by shifting the relationship between host plant composition and disease (Halliday et al., 2021). The differences between these two studies might be due to variation in climate (higher temperature and precipitation in the Swiss Alps), vegetation type (our site was a typical alpine meadow, while the Swiss Alps had more montane habitat), and/or elevation range of sampling (1101 m for Swiss Alps *vs.* 800 m here). Elevation is also associated with a series of changes in plant communities, soil properties and other environmental factors (Lomolino, 2001). These factors may have both positive (e.g. warming; Siebold and Tiedemann, 2013; Liu et al., 2019) and negative (e.g., dilution effect; Mitchell et al., 2002; Rottstock et al., 2014; Halliday et al., 2020) effects on foliar fungal diseases. These effects could also offset each other, and environmental gradients can further modify how host community structure affects disease (Halliday et al., 2021), resulting in imperceptible associations between elevation with foliar fungal diseases. Hence, the relationship between elevation and foliar fungal diseases was difficult to explain using temperature, precipitation, latitude and elevation range of sampling in our meta-analysis.

Factors affecting above- and belowground plant pathogens

Our results provide empirical support that both *Evenness* and*Proneness* can shape community pathogen loads, highlighting the importance of including host community indices into disease ecology models (Liu et al., 2017; Halliday et al., 2019). *Proneness*increased with elevation, indicating that plant communities at higher elevations contained greater proportions of competent species (i.e. species with good growth but weak defensive abilities, thus harboring more pathogens; Liu et al., 2017), though this point was not fully supported by the study in the Swiss Alps (Halliday et al., 2021). We attribute the variation in the plant species level proneness index partly to the growth-defense trade-off, which suggests that the enhancement of defense systems comes at the cost of reduced growth, based on the assumption that plants share limited resources (Coley et al., 1985; Cappelli et al., 2020). The community proneness index was the best predictor of pathogen loads along the elevational gradient in the alpine meadow (explaining ˜ 34.0% of variation), suggesting that plant communities dominated by host species with higher proneness (i.e. weak defense systems) might experience more serious disease. Thus, our study calls for the inclusion of host community composition into models that predict infectious disease (Halliday, Rohr, et al., 2020).

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Our study provided empirical evidence for a negative biodiversity-disease relationship, which ecologists have long debated (Rottstock et al., 2014; Halliday et al., 2019; Halliday and Rohr, 2019; Liu, Chen, et al., 2020). Pielou's evenness index was an effective predictor of pathogen load, even along such a sharp elevation gradient, consistent with the theoretical prediction that host community evenness should be a better predictor of disease than species richness (Chen and Zhou, 2015). The evenness index incorporates the distribution of relative abundances among host species, which can better describe the mechanism of encounter reduction between host biodiversity and plant diseases (Mitchell et al., 2002; Liu et al., 2016). Therefore, a higher community evenness index translates into a lower probability of focal individuals being infected through encounter reduction (Keesing et al., 2006).

Plant community-, environment- and soil-mediated relationships between elevation and above- and belowground plant pathogens

Together, our results provide broad evidence that above- and belowground pathogens had distinct geographical vertical pattern which were shaped by different mechanisms. For foliar fungal pathogens, we found empirical evidence that plant community characteristics (i.e. community proneness index and evenness), rather than soil properties, were the main drivers of community pathogen load in our field survey. However, results did not support any direct or indirect relationship between elevation and foliar fungal disease.

These results highlight the importance of host identity to determine community-level diseases. In the field survey, shifts in plant community proneness to diseases ultimately led to difference in community pathogen load, a finding that is consistent with other studies from both plant diseases (Mitchell et al., 2002; Liu et al., 2017) and also*Ribeiroia ondatrae* caused amphibian diseases (Johnson et al., 2013). However, although only a small part of the 73 plant species' distributions overlaps with each other and generates considerable variance in the composition of host communities for pathogens along the elevation gradient, the degree of communities prone to disease did not synchronously change. Therefore, plant community mediated effect was not found in our study.

In general, our results indicated that abiotic factors mediated the association between elevation and soil fungal pathogen richness, while elevation showed no significant direct or indirect association with foliar fungal diseases and soil fungal pathogen relative abundance. Indeed, different measurements for pathogens (i.e. OTU richness versus relative abundance) may explain the inconsistent responses of pathogens to abiotic and biotic factors. For instance, positive plant richness- pathogen diversity relationship and negative diversity-disease relationship can be observed in natural plant communities, given that increasing plant species diversity provides more diverse hosts while simultaneously inducing dilution effects (Rottstock et al., 2014). However, there is insufficient evidence to determine the relative strength of which pathogen richness responds to the abiotic and biotic environment. In addition, the different patterns of above- and belowground plant pathogens along elevational gradients can be partly explained by their differences in life history characteristics. Rusts (e.g. *Phragmidium* , *Puccinia* and *Uromyces*) are the dominant foliar fungal pathogens in our study site (Liu et al., 2019); these fungi belong to a group of obligate biotrophic pathogens that can only extract nutrients from living plant cells (Duplessis et al., 2021) and have relatively narrow host ranges (one or just a few phylogenetically close plant species; Zhang, 2009). In contrast, soil plant pathogens are commonly necrotrophic with a relatively broad host range (Delgado-Baquerizo et al., 2020). Therefore, we expect that negative biodiversity-disease relationships might occur more commonly for foliar pathogens; foliar diseases are strongly dependent on their hosts, so they can be easily captured by host composition rather than environmental factors (e.g. temperature). For soil biota, we found empirical evidence that increasing elevation was associated with reductions in soil fungal pathogen richness via changes in soil properties (*Soil PCA1*). Our meta-analysis providing further evidence of a general negative association between soil fungal pathogen richness and elevation among studies. However, elevation had no significant association with soil fungal pathogen relative abundance. Unlike the severity of foliar fungal disease or fungal pathogen relative abundance, soil fungal pathogen richness depends on the relative rate of fungal colonization and extinction of fungal taxa. On the one hand, temperature may promote soil fungal pathogen richness through the following three mechanisms: First, temperature is a key limiting factor for plant biomass and richness (Chu et al.,

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

2019). Broad evidence from plants suggests that the species richness of lower trophic levels can determine the diversity of higher trophic levels, including fungal pathogens (Kamiya et al., 2014; Rottstock et al., 2014; Liu et al., 2016). Moreover, increased plant biomass provides greater host availability and more diverse habitats for pathogens. These plant-mediated paths can shape a potential negative relationship between elevation and soil fungal pathogen richness. Second, temperature can promote coevolution between hosts and pathogens, given the stronger inter-trophic interactions in warm areas (Roslin et al., 2017; Liu, Chen, et al., 2020); this is thought to be a main factor shaping pathogen richness. Third, temperature can determine the distribution of soil pathogens via environment filtering (Tedersoo et al., 2014); the fitness of pathogens is largely affected by temperature and humidity through impacts on survival, growth, dispersal and reproduction, both at local and global scales (Tedersoo et al., 2014; Liu et al., 2019; Delgado-Baquerizo et al., 2020).

On the other hand, soil properties may be a regulator of plant pathogens. Firstly, soil nutrients may benefit pathogens in both agro- and natural ecosystems by increasing tissue nitrogen concentration (Huber and Watson, 1974; Veresoglou et al., 2013; Liu et al., 2017). Secondly, soil properties can alter the competition dynamics and shape plant community composition (e.g. inducing the light asymmetry under nitrogen addition; Xiao et al., 2021), which possibly indirectly influence soil plant pathogens by preferring species with better growth ability (Cappelli et al., 2020). The soil fungal pathogen relative abundance showed no significant variation along the elevation gradient, despite considerable changes in temperature and soil properties. We attribute this to the small proportion of soil fungal pathogens relative to total soil fungi (i.e. percentage of soil fungal pathogens copy number; on average of 7.72% in our study), which may potentially limit the ability of soil pathogens to respond to biotic and abiotic variables.

Conclusion

Our multi-method approach provides evidence that community pathogen load is more commonly associated with host community composition (evenness and proneness) than with linear changes along the elevational gradient. In contrast, we did find that increasing elevation was associated with soil fungal pathogen richness (but not relative abundance), via the effects of temperature and soil properties. These results highlight elevational patterns of above- and belowground plant pathogens may be regulated by distinct mechanisms, and are consistent with a growing body of studies of foliar diseases (Mitchell et al., 2002; Rottstock et al., 2014; Cappelli et al., 2020; Halliday et al., 2021) and soil pathogens (Tedersoo et al., 2014; Delgado-Baquerizo et al., 2020). More importantly, there is increasing evidence that environmental gradients can modify how host community structure affects disease (Halliday, Heckman, et al., 2020; Halliday et al., 2021). These interaction effects may help explain the variation among different studies, highlighting that only looking at environmental factors or community structure would miss key information relevant to diseases. Nevertheless, for the first time, to the best of our knowledge, we distinguish the different mechanisms driving different groups of plant pathogens in an alpine meadow. Because of rapid changes in both plant communities and climate change in the Anthropocene, our study implies that incorporating different information to build models predicting above- and belowground plant pathogens is of great importance for understanding ecosystem health.

References

1. Allan, E., van Ruijven, J., and Crawley, M. J. 2010. Foliar fungal pathogens and grassland biodiversity. – Ecology 91: 2572–2582.
2. Allen, W. J., DeVries, A. E., Bologn, N. J., Bickford, W. A., Kowalski, K. P., Meyerson, L. A., and Cronin, J. T. 2020. Intraspecific and biogeographical variation in foliar fungal communities and pathogen damage of native and invasive*Phragmites australis* . – Glob. Ecol. Biogeogr. 29: 1199–1211.
3. Bartoń, K. 2022. MuMIn: Multi-Model Inference. Retrieved from https://CRAN.R-project.org/package=MuMIn
4. Bebber, D. P. 2015. Range-expanding pests and pathogens in a warming world. – Annu. Rev. Phytopathol. 53: 335–356.
5. Borenstein, M., Hedges, L., Higgins, J., and Rothstein, H. 2009.*Introduction to meta-analysis* . New York, NY, USA: John Wiley & Sons.
6. Burdon, J. J., and Chilvers, G. A. 1982. Host density as a factor in plant-disease ecology. – Annu.

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Rev. Phytopathol. 20: 143–166.
7. Burnham, K. P., Anderson, D. R., and Huyvaert, K. P. 2011. AIC model selection and multimodel inference in behavioral ecology: some background, observations, and comparisons. – Behav. Ecol. Sociobiol. 65: 23–35.
8. Cappelli, S. L., Pichon, N. A., Kempel, A., and Allan, E. 2020. Sick plants in grassland communities: a growth-defense trade-off is the main driver of fungal pathogen abundance. – Ecol. Lett. 23: 1349–1359.
9. Chen, L. F., and Zhou, S. R. 2015. A combination of species evenness and functional diversity is the best predictor of disease risk in multihost communities. – Am. Nat. 186: 755–765.
10. Chu, C. J. *et al* . 2019. Direct and indirect effects of climate on richness drive the latitudinal diversity gradient in forest trees. – Ecol. Lett. 22: 245–255.
11. Coley, P. D., Bryant, J. P., and Chapin, F. S. 1985. Resource availability and plant antiherbivore defense. – Science 230: 895–899.
12. Delgado-Baquerizo, M., Guerra, C. A., Cano-Diaz, C., Egidi, E., Wang, J. T., Eisenhauer, N., Singh, B. K., and Maestre, F. T. 2020. The proportion of soil-borne pathogens increases with warming at the global scale. – Nat. Clim. Chang. 10: 550–554.
13. Duplessis, S., Lorrain, C., Peter, B., Figueroa, M., Dodds, P. N., and Aime, M. C. 2021. Host adaptation and virulence in heteroecious rust fungi. – Annu. Rev. Phytopathol. 59: 403–422.
14. Fick, S. E., and Hijmans, R. J. 2017. WorldClim 2: new 1km spatial resolution climate surfaces for global land areas. – Int. J. Climatol. 37: 4302–4315.
15. Grace, J. B. 2006. *Structural equation modeling and natural systems* . Cambridge University Press, Cambridge, UK.
16. Halbritter, A. H., Fior, S., Keller, I., Billeter, R., Edwards, P. J., Holderegger, R., Karrenberg, S., Pluess, A. R., Widmer, A., and Alexander, J. M. 2018. Trait differentiation and adaptation of plants along elevation gradients. –*J. Evol. Biol.* 31: 784–800.
17. Halliday, F. W., Heckman, R. W., Wilfahrt, P. A., and Mitchell, C. E. 2017. A multivariate test of disease risk reveals conditions leading to disease amplification. – Proc. R. Soc. B-Biol. Sci. 284: 20171340.
18. Halliday, F. W., Heckman, R. W., Wilfahrt, P. A., and Mitchell, C. E. 2019. Past is prologue: host community assembly and the risk of infectious disease over time. – Ecol. Lett. 22: 138–148.
19. Halliday, F. W., and Rohr, J. R. 2019. Measuring the shape of the biodiversity-disease relationship across systems reveals new findings and key gaps. – Nat. Commun. 10: 5032.
20. Halliday, F. W., Rohr, J. R., and Laine, A. L. 2020. Biodiversity loss underlies the dilution effect of biodiversity. – Ecol. Lett. 23: 1611–1622.
21. Halliday, F. W., Jalo, M., and Laine, A. L. 2021. The effect of host community functional traits on plant disease risk varies along an elevational gradient. – eLife 10: e67340.
22. Hansen, E. M., and Goheen, E. M. 2000 *Phellinus weirii* and other native root pathogens as determinants of forest structure and process in western North America. – Annu. Rev. Phytopathol. 38: 515–539.
23. Healey, S. P., Raymond, C. L., Lockman, I. B., Hernandez, A. J., Garrard, C., and Huang, C. 2016. Root disease can rival fire and harvest in reducing forest carbon storage. – Ecosphere 7: e01569.
24. Huang, H. Y., Zhou, L., Chen, J., and Wei, T. Y. 2020. ggcor: Extended tools for correlation analysis and visualization. R package version 0.9.8.1. Retrieved from https://github.com/houyunhuang/ggcor
25. Huber, D. M., and Watson, R. D. 1974. Nitrogen form and plant disease. – Annu. Rev. Phytopathol. 12: 139–165.
26. Johnson, P. T., Preston, D. L., Hoverman, J. T., and Richgels, K. L. 2013. Biodiversity decreases disease through predictable changes in host community competence. – Nature 494: 230–233.
27. Kamiya, T., O'Dwyer, K., Nakagawa, S., and Poulin, R. 2014. Host diversity drives parasite diversity: meta-analytical insights into patterns and causal mechanisms. – Ecography 37: 689–697.
28. Keesing, F., Holt, R. D., and Ostfeld, R. S. 2006. Effects of species diversity on disease risk. – Ecol. Lett. 9: 485–498.
29. Keesing, F. *et al* . 2010. Impacts of biodiversity on the emergence and transmission of infectious

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

diseases. – Nature 468: 647–652.
30. Lajeunesse, M. J. 2013. *Recovering missing or partial data from studies: a survey of conversions and imputations for meta-analysis* . In: Koricheva J, Gurevitch J, Mengersen K. eds. *Handbook of meta-analysis in ecology and evolution* . Princeton, NJ, USA: Princeton University Press, 195–206.
31. Lefcheck, J. S. 2016. piecewiseSEM: Piecewise structural equation modelling in r for ecology, evolution, and systematics. – Methods Ecol. Evol. 7: 573–579.
32. Liang, M. X., Liu,. X. B., Gilbert, G. S., Zheng, Y., Luo, S., Huang, F. M., and Yu, S X. 2016. Adult trees cause density-dependent mortality in conspecific seedlings by regulating the frequency of pathogenic soil fungi. – Ecol. Lett. 19: 1448–1456.
33. Liu, X., Lyu, S. M., Zhou, S. R., and Bradshaw, C. J. A. 2016. Warming and fertilization alter the dilution effect of host diversity on disease severity. – Ecology 97: 1680–1689.
34. Liu, X., Lyu, S. M., Sun, D. X., Bradshaw, C. J. A, and Zhou, S. R. 2017. Species decline under nitrogen fertilization increases community-level competence of fungal diseases. – Proc. R. Soc. B-Biol. Sci. 284: e20162621.
35. Liu, X., Ma, Z. Y., Cadotte, M. W., Chen, F., He, J. S., and Zhou, S. R. 2019. Warming affects foliar fungal diseases more than precipitation in a Tibetan alpine meadow. – New Phytol. 221: 1574–1584.
36. Liu, X., Chen, L. F., Liu, M., Garcia-Guzman, G., Gilbert, G. S., and Zhou, S. R. 2020. Dilution effect of plant diversity on infectious diseases: latitudinal trend and biological context dependence. – Oikos 129: 457–465.
37. Liu, X., Lu, Y. W., Zhang, Z. H., and Zhou, S. R. 2020. Foliar fungal diseases respond differently to nitrogen and phosphorus additions in Tibetan alpine meadows. – Ecol. Res. 35: 162–169.
38. Liu, X., Zhang, L., Huang, M. J., and Zhou, S. R. 2021. Plant diversity promotes soil fungal pathogen richness under fertilization in an alpine meadow. – J. Plant Ecol. 14: 323–336.
39. Liu, Y., and He, F. L. 2019. Incorporating the disease triangle framework for testing the effect of soil-borne pathogens on tree species diversity. – Funct. Ecol. 33: 1211–1222.
40. Lomolino, M. V. 2001. Elevation gradients of species-density: historical and prospective views. – Glob. Ecol. Biogeogr. 10: 3-13.
41. Lu, J. Y. 1997. *Plant disease diagnosis* . Beijing, China: China Agricultural Press.
42. Mbareche, H., Veillette, M., Bilodeau, G., and Duchaine, C. 2020. Comparison of the performance of ITS1 and ITS2 as barcodes in amplicon-based sequencing of bioaerosols. – PeerJ 8: e8523.
43. Mitchell, C. E., Tilman, D., and Groth, J. V. 2002. Effects of grassland plant species diversity, abundance, and composition on foliar fungal disease. – Ecology 83: 1713–1726.
44. Nakagawa, S., Noble, D. W. A., Senio, A. M., and Lagisz, M. 2017. Meta-evaluation of meta-analysis: ten appraisal questions for biologists. – BMC Biol. 15: 18.
45. Nguyen, N. H., Song, Z. W., Bates, S. T., Branco, S., Tedersoo, L., Menke, J., Schilling, J. S., and Kennedy, P. G. 2015. FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. – Fungal Ecol. 20: 241-248.
46. Oksanen, J. *et al* . 2020. vegan: Community Ecology Package. R package version 2.5.7. Retrieved from https://CRAN.R-project.org/package=vegan
47. Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., and R Core Team. 2021. nlme: Linear and Nonlinear Mixed Effects Models. Retrieved from https://CRAN.R-project.org/package=nlme
48. R Development Core Team. 2021. R: A language and environment for statistical computing. Version 4.1.1. Vienna, Austria: R Foundation for Statistical Computing. Retrieved from http://www.R-project.org
49. Rohatgi, A. 2020. WebPlotDigitizer (Version 4.4). Retrieved from https://automeris.io/WebPlotDigitizer
50. Romero, F., Cazzato, S., Walder, F., Vogelgsang, S., Bender, S. F., and van der Heijden, M. G. A. 2021. Humidity and high temperature are important for predicting fungal disease outbreaks worldwide. – New Phytol. 234, 1553–1556.
51. Rosenberg, M. S., Rothstein HR, and Gurevitch J. 2013. *Effect sizes: conventional choices and calculations* . In: Koricheva J, Gurevitch J, Mengersen K. eds. *Handbook of meta-analysis in ecology and*

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

evolution . Princeton, NJ, USA: Princeton University Press, 61–71.
52. Roslin, T. *et al* . 2017. Higher predation risk for insect prey at low latitudes and elevations. – Science, 356: 742–744.
53. Rottstock, T., Joshi, J., Kummer, V., and Fischer, M. 2014. Higher plant diversity promotes higher diversity of fungal pathogens, while it decreases pathogen infection per plant. – Ecology 95: 1907–1917.
54. Rowe, R. J. 2009. Environmental and geometric drivers of small mammal diversity along elevational gradients in Utah. – Ecography 32: 411–422.
55. Roy, B. A., Gusewell, S., and Harte, J. 2004. Response of plant pathogens and herbivores to a warming experiment. – Ecology 85: 2570–2581.
56. Siebold, M., and Tiedemann, A. 2013. Effects of experimental warming on fungal disease progress in oilseed rape. – Glob. Change Biol. 19: 1736–1747.
57. Stukenbrock, E. H., McDonald, B. A. 2008. The origins of plant pathogens in agro-ecosystems. – Annu. Rev. Phytopathol. 46: 75–100.
58. Tedersoo, L. *et al* . 2014. Global diversity and geography of soil fungi. – Science 346: 1078–1078.
59. Tedersoo, L., and Lindahl, B. 2016. Fungal identification biases in microbiome projects. – Env. Microbiol. Rep. 8: 774–779.
60. Tellenbach, C., Grunig, C. R., and Sieber, T. N. 2010. Suitability of quantitative real-time PCR to estimate the biomass of fungal root endophytes. – Appl. Environ. Microb. 76: 5764–5772.
61. Wei, J. C. 1979. *Fungal Identification Manual* . Shanghai Science and Technology Press, Shanghai, China.
62. van Agtmaal, M., Straathof, A., Termorshuizen, A., Teurlincx, S., Hundscheid, M., Ruyters, S., Busschaert, P., Lievens, B., and de Boer, W. 2017. Exploring the reservoir of potential fungal plant pathogens in agricultural soil. – Appl. Soil Ecol. 121: 152–160.
63. Veresoglou, S. D., Barto, E. K., Menexes, G., and Rillig, M. C. 2013. Fertilization affects severity of disease caused by fungal plant pathogens. – Plant Pathol. 62: 961–969.
64. Viechtbauer, W. 2010. Conducting meta-analyses in R with the metafor package. – J. Stat. Softw. 36: 1–48.
65. Xiao, Y., Liu, X., Zhang, L., Song, Z. P., and Zhou, S. R. 2021. The allometry of plant height explains species loss under nitrogen addition. – Ecol. Lett. 24: 553–562.
66. Zhang, R. 2009. Survey and identification of the alpine grassland's major fungal diseases in Gannan region of Gansu province. Master thesis, Gansu Agricultural University, Lanzhou, Gansu, China.
67. Zhu, J. T., Zhang, Y. J., Yang, X., Chen, N., Li, S. P., Wang, P. D., Jiang, L. 2020. Warming alters plant phylogenetic and functional community structure. – J. Ecol. 108: 2406–2415.

Table 1. All abbreviations and their corresponding annotations in the main text.

Category	**Abbreviation**	**Annotation**
Soil properties	*Soil PCA1*	The first principal component of soil properties PCA
Plant community characters	*AGB*	Plant aboveground biomass
	BGB	Plant belowground biomass
	SR	Plant community species richness
	Evenness	Plant community Pielou's evenness index
	Proneness	Plant community proneness index
Environmental conditions	*MDT*	Mean daily temperature
	MDH	Mean daily humidity
Plant pathogen indices	*PL*	Plant community pathogen load
	sfpOTUs	Soil fungal pathogen OTU richness
	sfpRA	Soil fungal pathogen relative abundance

Figure legends

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Figure 1. Effects of *Elevation* , soil properties (*Soil PCA1*), aboveground biomass (*AGB*), Belowground biomass (*BGB*), Pielou's evenness index (*Evenness*), community proneness index (*Proneness*) and species richness (*SR*) on (a) community pathogen load (*PL*), (b) soil fungal pathogen OTU richness (*sfpOTUs*) and (c) soil fungal pathogen relative abundance (*sfpRA*). Effect sizes with 95% confidence intervals are weighted average standardized coefficients from models with ΔAICc < 4 based on model selections (Table S4, S5, S6). Blue lines indicate significant negative effects, grey lines indicate non-significant effects, and red lines indicate significant positive effects.

Figure 2. The piecewise structural equation model. (a) The hypothesized piecewise structural equation model. Hypothesized plant community-mediated and soil-mediated association between elevation and community pathogen load and soil fungal pathogen OTU richness. Specifically, paths a-c represent the association between elevation and host plant community and soil properties, paths d, e represent the direct association between elevation and community pathogen load and soil fungal pathogen OTU richness, while paths f-h and paths i-k represent the community-mediated and soil-mediated effects on community pathogen load and soil fungal pathogen OTU richness, respectively. (b) The final piecewise structural equation model. Numbers on arrows are standardized path coefficients (scaled by their mean and standard deviation), and asterisks indicate statistical significance (***$P < 0.001$; * $0.01 < P < 0.05$; + $0.05 <$ P < 0.1). Red arrows represent significant positive relationships while blue arrows represent significant negative relationships at the $P <$ 0.1 level; gray arrows with dashed lines represent insufficient statistical evidence for path coefficients ($P >$ 0.1). Arrow thickness is proportional to the strength of the path. *De*indicates the deviance explained by predictive variables.

Figure 3. Overall result of the meta-analysis testing the associations between elevation and foliar fungal pathogen OTU richness, foliar fungal diseases, soil fungal pathogen OTU richness and soil fungal pathogen relative abundance. Shown are the mean values of the effect sizes (Z) and the corresponding 95% confidence intervals (95% CI).

Figure 1.

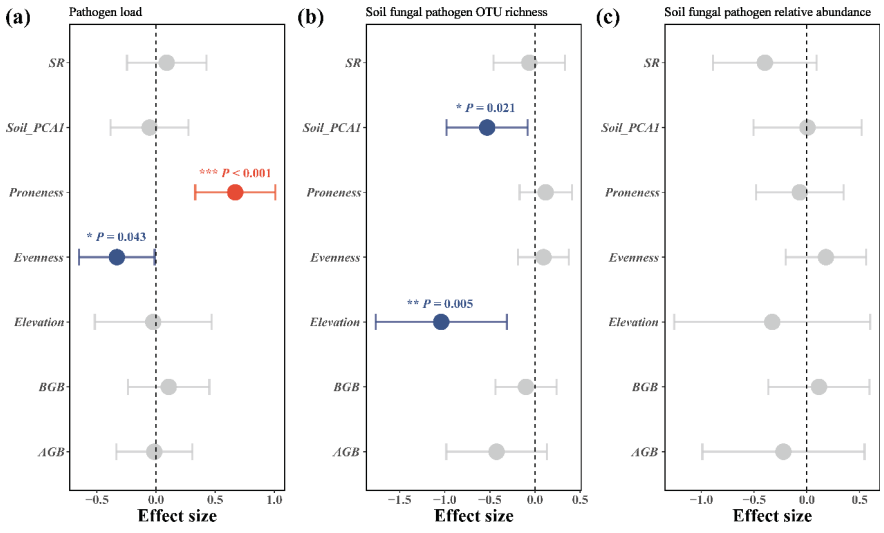

Figure 2.

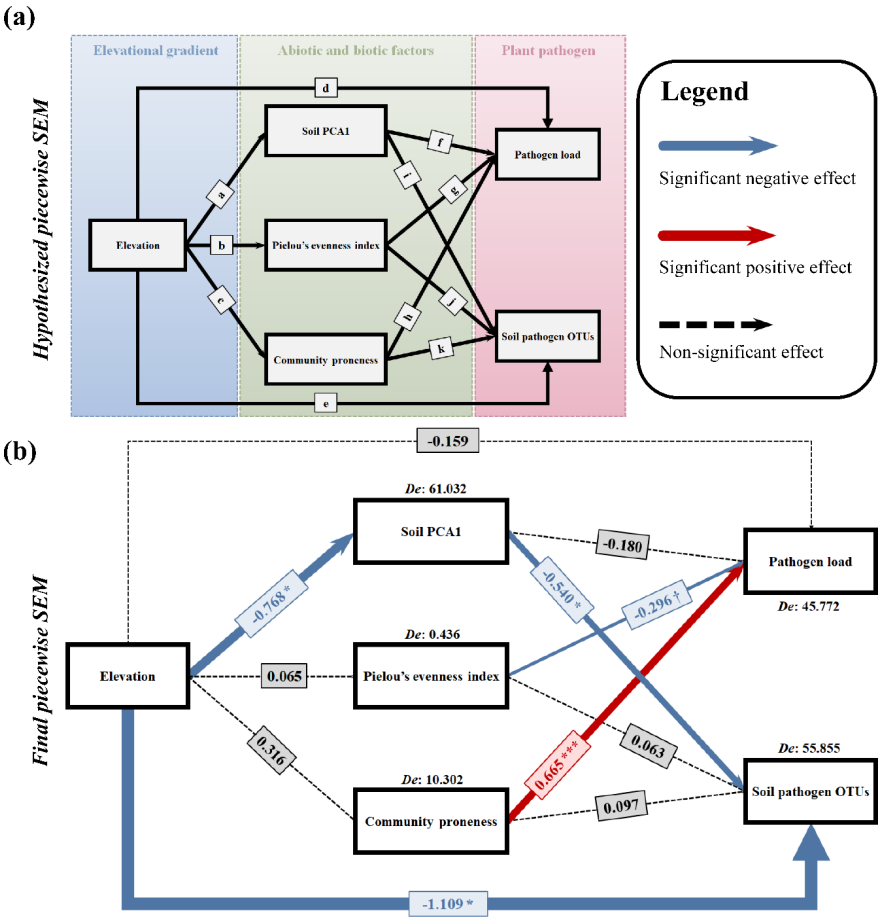

Figure 3.

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.

Posted on 26 Sep 2023 — The copyright holder is the author/funder. All rights reserved. No reuse without permission. — https://doi.org/10.22541/au.169572791.10640197/v1 — This a preprint and has not been peer reviewed. Data may be preliminary.