



## Urbanization simplifies soil nematode communities and coincides with decreased ecosystem stability

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### ABSTRACT

Urbanization is threatening terrestrial biodiversity and ecosystem functions. Nematodes, as one of the most abundant animals on Earth, play diverse roles within soil food webs and contribute to vital ecosystem services. However, the effects of urbanization on soil nematode functional groups, and the ecological functions and services they provide remain poorly understood. Here, we investigated functional groups of soil nematodes across 12 cities in China and validated the results by using a global nematode dataset. Specifically, we found the proportions of omnivores (by 45–60%) and predators (by 40–50%) as well as temporal ecosystem stability (by 10–20%), as indicated by the normalized difference vegetation index, were reduced in urban ecosystems, while the proportions of fungivores and bacterivores increased (30–60% and 10–20%, respectively). We identified total soil phosphorus and nitrogen as the main drivers influencing soil nematode feeding groups in urban ecosystems. Soil pH, mean annual precipitation, and mean annual temperature were predominant factors in non-urban ecosystems. We further demonstrated that nematode functional guilds contributed to ecosystem stability in urban ecosystems, but this was not true in non-urban ecosystems. Our findings evidence the homogenization effects of urbanization on key functional guilds of soil nematodes and their roles in maintaining ecosystem stability. The fact that ecosystem stability and the abundance of nematode functional guilds are closely related and driven by climatic, edaphic, and anthropogenic factors may help in setting priorities for urban soil management.

### 1. Introduction

Urban soils are crucial components of terrestrial ecosystems by providing essential ecosystem services (Delgado-Baquerizo et al., 2021; Eisenhauer et al., 2017), including green space establishment, water infiltration, and nutrient cycling (Yang et al., 2018). Unfortunately, soils in cities are threatened by construction activities, soil sealing, and

pollution (FAO, 2020; Lorenz and Lal, 2009). Soil research seeks to comprehend the complex interrelations by considering interactions between physical, chemical, and biological factors (Decaëns, 2010), and their feedback on urban soil properties and processes. However, research addressing issues in urban soils is scarce and less attention is paid to urban soil management (McDonald et al., 2020). A multifaceted understanding is vital for developing effective management strategies

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that can maintain or improve urban soil health, functionality, and resilience, in response to environmental pressures and human activities (Wall et al., 2015).

Soil biota forms an essential component for the restoration and conservation of soil health (Sun et al., 2023; Wardle et al., 2004). Including soil biota in soil health assessment allows a better understanding of aboveground-belowground interactions and thereby the development of holistic management regimes (Lehmann et al., 2020). It also contributes to improving predictions of the effects of human-induced environmental changes on biodiversity and ecosystem properties (Fierer et al., 2009), thereby promoting more sustainable approaches to land management in urban ecosystems. As global trends move towards increasing degrees of urban area, it is becoming important to understand the response of biodiversity in urban places (Pavao-Zuckerman and Coleman, 2007). Urbanization may be associated with increased soil compaction and decreased soil nutrient concentrations which are critical factors impacting soil biota (Lepczyk et al., 2017). However, knowledge of changes in the structure and function of soil biota in urban ecosystems is limited (Beninde et al., 2015). Especially soil biota is less studied in urban environments, hampering predictions on the impact of urbanization on the functions and services provided by urban soils.

Nematodes are among the most abundant soil fauna on Earth (van den Hoogen et al., 2019), and changes in their abundance and diversity can greatly influence ecosystem services provided by urban soils (Park et al., 2013). Soil nematodes are key components of the soil food web and are characterized by different feeding groups spanning from primary decomposers to predators (Gebremikael et al., 2016). Bacterivore and fungivore nematodes consume bacteria and fungi, respectively, which may promote soil nutrient cycling. Herbivore nematodes feed on plant root material and function as parasites directly interacting with plants (Topalović and Geisen, 2023). Predatory and omnivorous nematodes rely on a variety of food resources, making them ideal bio-indicators of soil ecosystems (Yeates et al., 1993). Given their diverse roles as bacterivores, fungivores, herbivores, omnivores, and predators in urban soils, the contributions of different nematode feeding groups to ecosystem services in urban soils remain unknown.

Comprising an important part of soil biota, nematodes are tightly interconnected forming 'soil networks' delivering ecosystem services (Morriën, 2016). The complex network of interactions between soil biota plays a key role in regulating ecosystem functions, from nutrient cycling and carbon sequestration to soil fertility and stability (Potapov et al., 2021). Closely connected taxa form modules, which are associated with certain processes and services (Creamer et al., 2016). In soil nematodes, feeding groups are widely characterized as functional guilds. However, taxa from different feeding groups might also interact in providing certain functions, and therefore ecosystem services may benefit from increased diversity within feeding groups (Cesarz et al., 2015).

Here, we applied an annotation-free framework to detect nematode functional guilds (Shan et al., 2023), which is a *de novo* method for defining nematode functional groups. Together, we also annotated the nematodes by classic feeding groups according to the published literature (Yeates et al., 1993). We investigated how nematode functional guilds as well as feeding groups respond to urbanization by comparing paired sites in 12 Chinese cities, including urban and non-urban sites. We selected two typical land-use types for both non-urban and urban environments. In the non-urban environments, four independent sites from natural habitats (Mostly consisting of natural or close to natural forests) and four independent sites from agricultural habitats (Cultivated with maize) were implemented. Further, we extracted data from a global nematode survey to validate our findings on the response of soil nematodes to urbanization (van et al., 2020; Yao et al., 2023). We aimed to decipher the relationship between soil nematodes and ecosystem services and explore how this relationship is modulated by edaphic and climatic factors in urban ecosystems. We hypothesized that (1) the

relative abundance of nematode functional guilds and feeding groups defined as high trophic levels, i.e., omnivores and carnivores that are sensitive to land-use changes, is decreased in urban compared to non-urban sites, and (2) the proportion of nematode functional guilds are correlated with temporal ecosystem stability, with the correlations being determined by edaphic, climatic, and societal factors in both urban and non-urban sites.

## 2. Materials and methods

### 2.1. Study design and soil sampling

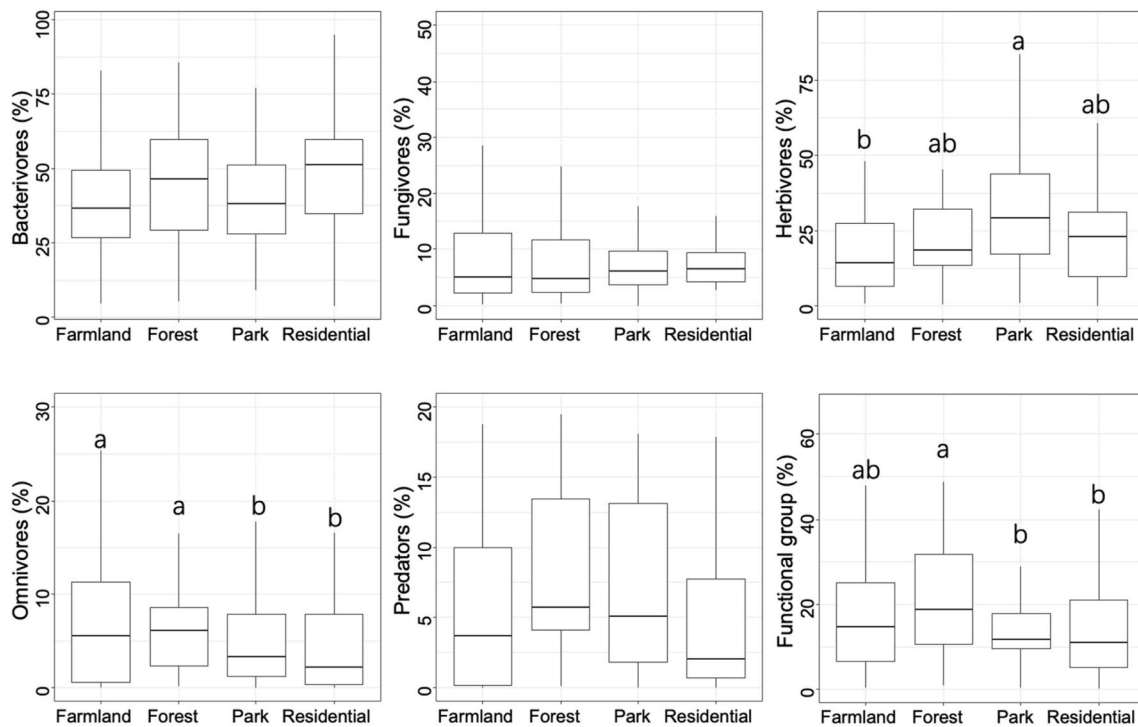
We collected soil samples from 12 cities across China, representing regions with various climates ranging from subtropical to temperate between May and August 2021. To investigate the urbanization effects, we collected samples located outside of urban areas, non-urban environments, and within the urban area, the urban environments. We also selected two land-use types for both non-urban and urban environments. In the urban environments, four independent sites from urban parks and four independent sites from urban residential areas were employed. In each city, the four independent sampling sites of each land-use type were located to each other at a distance of at least 1 km. Thus, we have 192 soil samples (12 cities  $\times$  4 land-use types  $\times$  4 replicates) for the present study (Fig. S1, Table S1). We randomly established a 20 m  $\times$  20 m plot for soil sampling at each sampling site. Within the plot, we collected a total of nine soil cores using a soil corer (5.5 cm in diameter, 0–10 cm in depth) and combined them into a composite sample. The samples were carefully transported under ice to the laboratory, and an aliquot subsequently used for molecular analysis was stored at  $-20^{\circ}\text{C}$ . The remaining soil was air-dried before being subjected to physico-chemical analysis.

### 2.2. Soil abiotic properties

Soil texture, pH, moisture, total carbon (TC), total nitrogen (TN), and total phosphorus (TP) were measured as soil abiotic variables. A subsample of the soil (10 g) was sieved through a 2.0 mm sieve and fractionated into sand (particle size, 50–2000  $\mu\text{m}$ ), silt (2–50  $\mu\text{m}$ ), and clay (<2  $\mu\text{m}$ ) using the ultrasonic energy method (Roscoe et al., 2000). The results of the particle size analysis were expressed as percentages of the total weight of oven-dried soil. Soil pH was measured using PHS-3C (Shanghai Leici) after shaking in soil-water (1:5 w/v) suspension for 30 min. Soil moisture content was determined using 10 g of fresh soil dried at  $105^{\circ}\text{C}$  for 48 h to constant weight. TC and TN were measured using an elemental analyzer (Elemental Analyzer System Vario Macro Cube, Langensfeld, Germany). TP was measured by Inductively Coupled Plasma-Atomic Emission Spectrometry (ICPS-7500) using the triacid digestion-ICP-AES method (Lu, 1999).

### 2.3. Climate and vegetation properties

For each sampling site, we extracted mean annual precipitation (MAP) and temperature (MAT) from WorldClim version 2.1 (<http://www.worldclim.org/>) at a spatial resolution of 30 s (Fick and Hijmans, 2017). Further, we retrieved the annual maximum value of the Normalized Difference Vegetation Index (NDVI) between 2000 and 2020 for each sampling site with a spatial resolution of 30 m  $\times$  30 m (data was downloaded from Chinese National Ecosystem Science Data Center; <http://www.nesdc.org.cn/sdo/detail?id=60f68d757e28174f0e7d8d49>). The temporal variation of NDVI values was documented to reflect ecosystem stability over a long-term period (Wu et al., 2023). In the present study, we calculated ecosystem temporal stability as the ratio of the annual mean to the standard deviation of the NDVI.



**Fig. 1.** The proportion of nematode feeding guilds and functional groups. Different letters indicate the significant ( $P < 0.05$ ) differences between groups tested by Tukey's HSD.

#### 2.4. Soil nematode community sequencing

Soil samples were thoroughly mixed and 10 g of soil was used for genomic DNA extraction using the MP FastDNA spin kit for soil (MP Biomedicals, Solon, OH, USA) according to the manufacturer's instructions. The quality and quantity of the extracted DNA were certified with 1% agarose gel electrophoresis and Nanodrop-2000 spectrophotometer (Nanodrop Technologies Inc. Wilmington, DE, USA), respectively. The V4 region of the 18S rDNA gene was chosen for amplicon sequencing using primer pair NF1-F/18Sr2b-R (Porazinska et al., 2009). PCR amplification was performed as follows: initial denaturation at 95 °C for 3 min, followed by 30 cycles of denaturing at 95 °C for 30 s, annealing at 55 °C for 30 s, extension at 72 °C for 30 s, and single extension at 72 °C for 10 min, and ended by holding at 4 °C. The PCR mixtures contained 4  $\mu$ L 5  $\times$  TransStart FastPfu buffer, 2  $\mu$ L dNTPs (2.5 mM), 0.8  $\mu$ L forward primer (5  $\mu$ M), 0.8  $\mu$ L reverse primer (5  $\mu$ M), 0.4  $\mu$ L TransStart FastPfu DNA Polymerase, 10 ng template DNA, and 20  $\mu$ L ddH<sub>2</sub>O. The PCR reactions were performed in triplicate and with a negative control replacing template DNA with ddH<sub>2</sub>O. The PCR product was extracted from 2% agarose gel and purified using the AxyPrep DNA Gel Extraction Kit (Axygen Biosciences, Union City, CA, USA) according to the manufacturer's instructions and quantified using Quantus™ Fluorometer (Promega, Madison, WI, USA). Purified amplicons were pooled and sequenced on an Illumina MiSeq PE300 platform (Illumina, San Diego, USA) by Majorbio Bio-Pharm Technology Co. Ltd. (Shanghai, China).

#### 2.5. Sequence data processing

Sequence data were processed using the UPARSE-UNOISE3 pipeline implemented in USEARCH v11, which showed the best balance between resolution and specificity in amplicon data analysis among current analysis pipelines (Himbeek et al., 2024). Paired-end reads were merged using the command `-fastq_mergepairs` with the parameter of 10 maximum in sequences. For quality filtering, we set the parameter "maxee" value to 1.0, which indicated the maximum nucleotide error

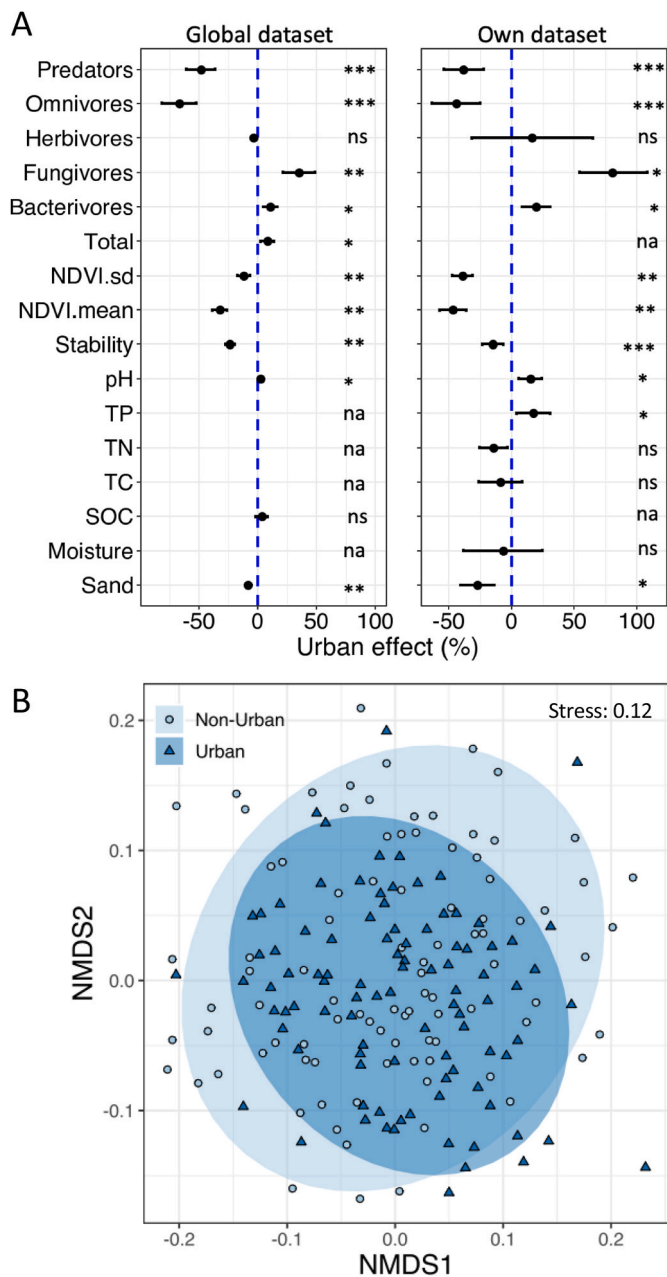
was 1.0 for each sequence. Then, sequences were dereplicated and singleton sequences were removed before phylogroup determinations using the command `-fastx_uniques`. After that, denoised representative sequences were obtained using the command `-unoise3`. The taxonomy of the sequences was assigned using SILVA v138 (Quast et al., 2013). The resulting ASVs were then filtered and those successfully assigned to "Nematode" were retained, which resulted in a rarefied 2300 sequences per sample for downstream analysis.

#### 2.6. Determination of feeding groups and functional guilds of nematodes

The feeding group for each nematode ASV was allocated according to the record collection from NEMAPLEX (<http://nemaplex.ucdavis.edu/>) with the taxonomy information collected from amplicon sequencing. Subsequently, the most relevant functional guilds of nematodes to ecosystem stability were determined using the Ensemble Quotient Optimization method, which was designed to find out an assemblage of species (nematode ASVs in the present study) that as a whole is most strongly indicative of a functional readout (Shan et al., 2023). The results were further subjected to cross-validation implemented in the R package mEQO and visualized in aggregation networks using the R package visNetwork (Almende et al., 2022; Shan et al., 2023).

#### 2.7. Statistical analyses

All the analyses and figures were generated using R 4.1.2 (R Core Team, 2022). In the analyses, we considered four categories of variables, i.e., space, climate, urban, and edaphic factors, to explore variations in nematode functional guilds in both urban and non-urban soils. Space was included as the latitude and longitude aiming to account for any potential influence of spatial autocorrelation. Climate variables were the MAT (mean annual temperature) and MAP (mean annual precipitation). To quantify the impact of urbanization, the population density for each city was retrieved from the World Bank (<https://data.worldbank.org/>). The edaphic variables were soil texture, pH, and soil nutrients. To quantify the contribution of each explanatory variable to variations in



**Fig. 2.** Effects of urbanization on nematode communities. Panel A, changes of nematode feeding groups, soil properties, and ecosystem stability in urban and non-urban soils. Points represent mean values of changes in response ratios calculated by the differences between urban and non-urban sites divided by the values in non-urban sites. Bars are 95% confidence intervals. Points to the right of the dashed line indicate that the properties are higher in urban sites compared to the corresponding non-urban areas. \*, \*\*, \*\*\* represent significant differences from zero,  $p < 0.05$ ,  $0.01$ , and  $0.001$ , resulting from students t-tests. ns, not significant ( $p > 0.05$ ); na, data not available. SOC, soil organic carbon; TC, soil total carbon; TN, soil total nitrogen; TP, soil total phosphorus. Panel B, nematode community composition as indicated by non-metric multidimensional scaling (NMDS). Circles represent 95% confidence intervals.

nematode richness, represented by ASV numbers in the present study, a random forest (RF) analysis with 5000 permutations was performed to evaluate the most important factors using the R package rfPermute (Archer, 2020). To analyze variations in nematode community composition in non-urban and urban environments, nonmetric multidimensional scaling (NMDS) calculated with Jaccard distance was performed using metaMDS in the R package vegan (Oksanen et al., 2020). To

determine the direct and indirect effects of environmental variations on soil nematode feeding groups, a partial least squares path model (PLS-PM) was constructed using the R package plsmpm (Sanchez et al., 2015). To build the model, four categories of variables and the land-use type (0 and 1 for non-urban and urban, respectively) were defined as latent variables. To examine the effects of environmental variables on the relationships between the proportion of functional guilds and ecosystem stability, a multiple regression tree analysis was performed and visualized using the R package ggparty (Borkovec and Madin, 2019). The differences in the proportions of nematode feeding guilds and functional groups between land-use types were compared using Tukey's HSD test.

Further, to validate the findings in our study, we also retrieved data, including the abundance of nematode feeding groups, climate, soil, and NDVI values, from the published global dataset (van den Hoogen et al., 2019; 2019). In the global dataset, the spatial resolutions for nematodes and environmental variables were matched (1 km). We extracted the sites located in urban areas within the global nematode dataset, which finally resulted in 22 cities outside of China. By co-extracting comparable non-urban sites in the vicinity of those cities, we calculated the urban effects on soil and nematode feeding groups.

### 3. Results

#### 3.1. Urban effects on soil properties, nematodes, and ecosystem stability

For the proportion of feeding guilds and functional groups, the differences between the two habitats within non-urban sites, i.e., farmlands and forests, were not significant (Fig. 1). This was the same case for the two habitats within urban sites, i.e., parks and residential areas (Fig. 1). Parks were greater in the proportion of herbivores than farmlands, omnivores were higher in farmlands and forests than in parks and residential areas (Fig. 1). Forests have the highest proportion of functional groups that were most related to ecosystem stability, followed by farmlands, parks and residential areas (Fig. 1). In both the global and our nematode dataset, the proportion of predatory (by 40~50%) and omnivore nematodes (by 45~60%), ecosystem stability (by 10~20%), and sand content (by 7~25%) was significantly lower in urban than non-urban soils (Fig. 2A). By contrast, both the proportion of bacterivore and fungivore nematodes were higher in urban than in non-urban soils, with the difference being greater for fungivores than for bacterivores (30~60% and 10~20%, respectively; Fig. 2A). In the national dataset, we also found the content of total phosphorus to be increased in urban compared to non-urban soils (by ~15%; Fig. 2A). NDVI was decreased in urban than non-urban soils by 38% and 49% for the global and our nematode dataset, respectively (Fig. 2A). Soil pH was increased in urban than non-urban soils by 8% and 10% for the global and our nematode dataset, respectively (Fig. 2A). Further, nematode community composition was homogenized, with a narrower confidence interval in urban compared to the non-urban soils (Fig. 2B).

#### 3.2. Environmental drivers of the relative abundance of nematode feeding groups

Soil chemical factors including TP and TN were identified as the main drivers of the relative abundance of nematode feeding groups, while in non-urban soils, pH, MAT, MAP, and longitude were more important (Fig. 3). Further, climatic factors and soil properties were significantly negatively associated with the relative abundance of nematode feeding groups in the pathway analysis (Fig. 4). Land-use change from non-urban to urban increased the proportion of bacterivores but decreased that of predators. Space, i.e. distance between sites, showed significant positive effects on the proportion of fungivores.

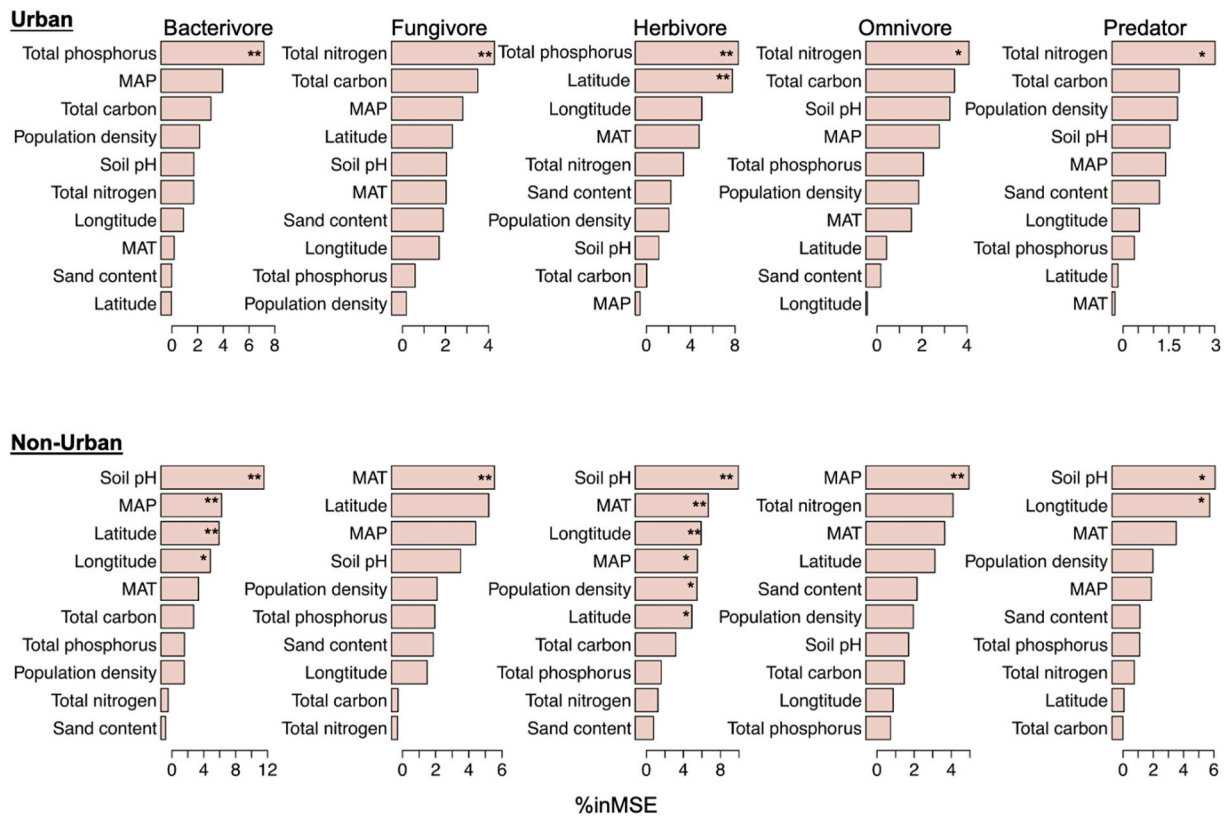


Fig. 3. Environmental factors associated with variations in the relative abundance of nematode feeding guilds in urban (upper panel) and non-urban soils (lower panel). Bars show the importance of the random forest predictor, with \* and \*\* representing significant differences from zero at  $p < 0.05$  and  $0.01$ , respectively.

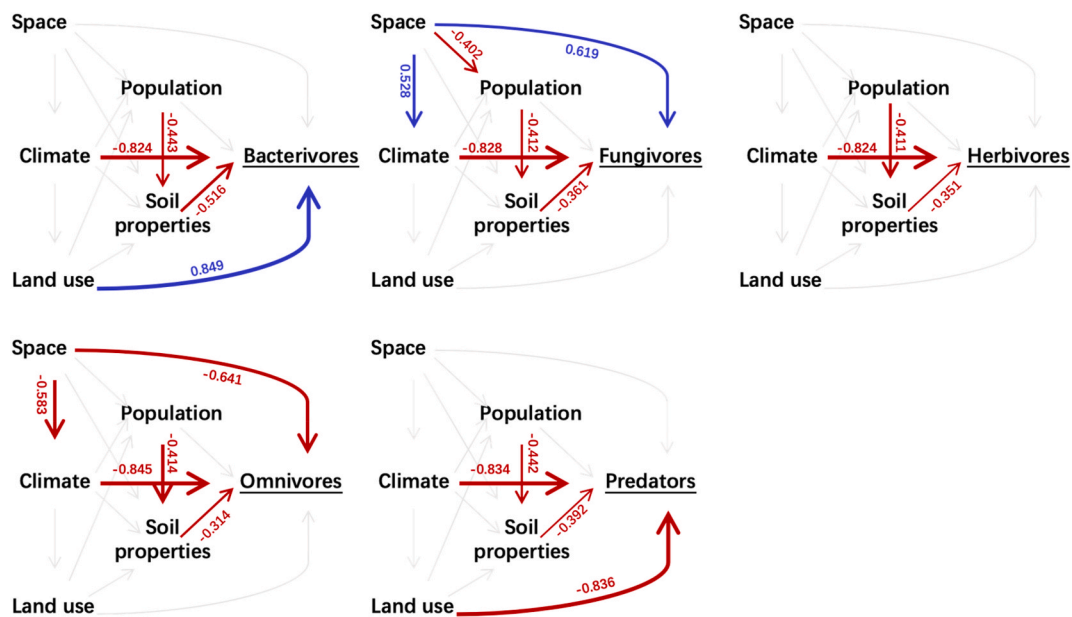
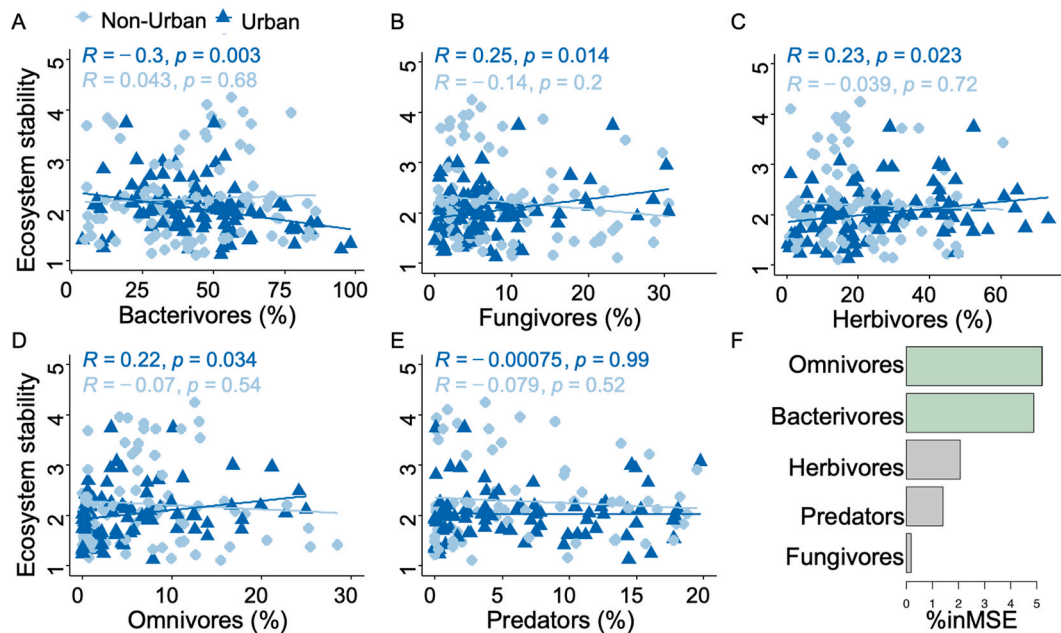


Fig. 4. Pathways associated with variations in nematode feeding guilds. Partial Least Squares Path Modeling (PLS-PM) of the direct and indirect associations among space (average distance among sites to control for spatial autocorrelation), climate, urban population, land use (urban compared with non-urban), and soil properties in driving the proportion of nematode functional guilds. Different categories of predictors (climate, soil, population, land use, and spatial influence) are grouped in the model for graphical simplicity. Numbers adjacent to arrows are the coefficients of the relationship, only significant relationships are included; red and blue represent significant negative and positive relationships, respectively. In the analyses, we considered four categories of variables, i.e., space, climate, urban, and edaphic factors, to explore variations in nematode functional guilds in both urban and non-urban soils. Space was included as the latitude and longitude aiming to account for any potential influence of spatial autocorrelation. Climate variables were the MAT (mean annual temperature) and MAP (mean annual precipitation). To quantify the impact of urbanization, the population density for each city was retrieved from the World Bank (<https://data.worldbank.org/>). The edaphic variables were soil texture, pH, and soil nutrients. Goodness-of-Fit of the models for bacterivores, fungivores, herbivores, omnivores, and predators were 0.81, 0.83, 0.79, 0.80, and 0.78.



**Fig. 5.** Relationship between the proportion of nematode feeding guilds and ecosystem stability. A-E, The linear relationships between the proportion of nematode feeding guilds and ecosystem stability. Statistics ( $r$ - and  $p$ -values) for the correlation analysis are indicated. Lines indicate significant relationships between the proportion of nematode feeding guilds and ecosystem stability. Solid and dashed lines represent significant ( $p < 0.05$ ) and non-significant ( $p > 0.05$ ) relationships, respectively. F, the contribution of the proportion of nematode feeding guilds to ecosystem stability tested by random forest analysis. The relative importance of each driver (%incMSE) was shown in the x-axis and the color bars denote the corresponding variables were significant ( $P < 0.05$ ) and the grey bars were not significant ( $P > 0.05$ ).

### 3.3. Associations between nematode functional guilds and ecosystem stability

While the proportions of bacterivores, fungivores, herbivores, and omnivores were significantly ( $P < 0.05$ ) related to temporal ecosystem stability in urban sites, this was not the case in non-urban sites ( $P > 0.05$ ; Fig. 5). All five nematode feeding groups interactively contributed to ecosystem stability (Fig. 6A). The proportion of functional guilds was significantly lower in urban than non-urban soils (Fig. 6B and C). MAP, latitude, and population density contributed to the classification of the relationships between nematode functional guilds and stability in a regression tree (Fig. 6D). The separation nodes were 1266 mm, 37.9 °C, and 1575 people per km<sup>2</sup> for MAP, latitude, and population density, respectively.

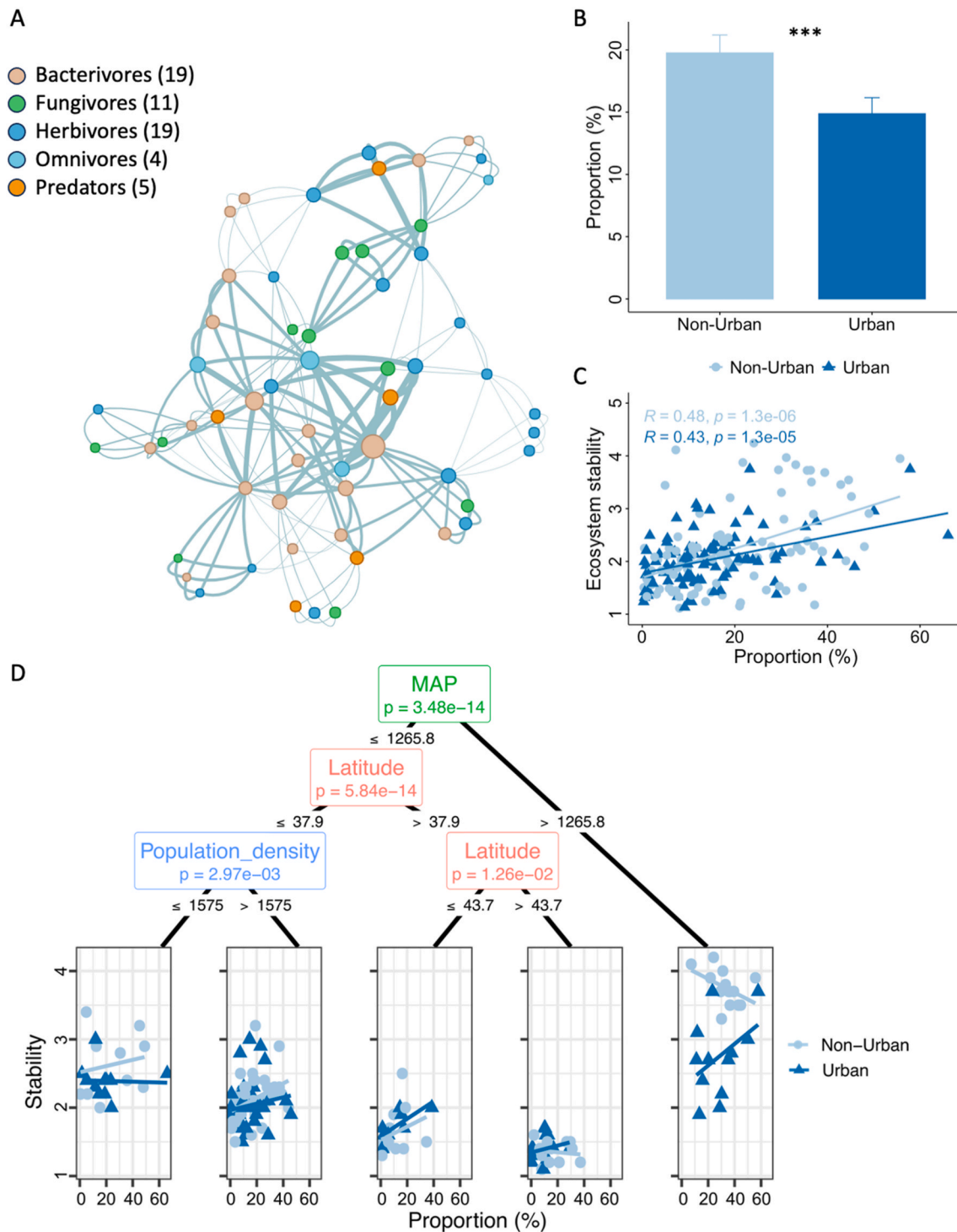
## 4. Discussion

### 4.1. Urban effects on soil nematode functional groups

In both the global dataset and the cities across China, we found that a lower proportion of nematode predators and omnivores were present in urban than non-urban soils, which supports our first hypothesis. Overall, habitat loss and fragmentation have been identified to be responsible for the reduced abundance and diversity of animals and plants in cities (Beninde et al., 2015; Fahrig, 2003). We found that urban areas were also characterized by lower sand content in the present study. The mechanisms responsible for the fact, however, remain unclear. Three indirect reasons might be related to this result. First, new residential sites were documented to have significantly lower sand contents compared to old residential sites (Scharenbroch et al., 2005), which might be applied to the sites in the present study. Secondly, the initial processes of urbanization significantly altered soil conditions in a variety of ways, within which removal and replacement of topsoil was the most common (Marcotullio et al., 2008). Given the fact that the contents of sand were lower in the remaining subsoil compared with the removed

or reformed surface soils, the altered soil might be shown with characteristics of lower sand content. Further, soil sealing due to urban infrastructure constructions, e.g., architecture in residential areas and pave roads in urban parks, causes the urban soil surface to be impervious (Pickett and Cadenasso, 2009). Thus, infiltration is limited for most urban soils, resulting in a relatively different hydrological condition of urban soils, compared to the soils in the open field including forests and farmlands. Lower sand content may have contributed to the lower relative abundance of nematode predators and omnivores, as the ideal pore size for nematodes is  $> 30 \mu\text{m}$  (Erktan et al., 2020). Smaller pore sizes associated with lower sand content in urban soils likely cooccur with lower soil organic carbon (Kong et al., 2009), which might detrimentally affect nematodes, especially large body-sized predators and omnivores. However, these effects have to be evaluated in future studies. Further, other factors associated with urbanization, such as pollution, were shown to reduce the diversity of native plants, and more adverse climatic conditions in urban compared to non-urban ecosystems likely contributed to the reduction in nematode predators and omnivores known to be more sensitive to disturbances than bacterial and fungal feeding nematodes (Zhao and Neher, 2013).

Increased relative abundance of bacterial and fungal-feeding nematodes in urban ecosystems supports our first hypothesis. Bacterial and fungal-feeding nematodes grow fast and quickly recover from population declines (Bongers, 1990). Thus, microbivore nematodes likely serve as important drivers of nutrient cycling in both urban and non-urban ecosystems, and this is supported by generally high densities of nematodes in urban ecosystems (Li et al., 2022). Notably, the increase in fungivore nematodes in urban ecosystems exceeded that of bacterivores and the content of total nitrogen was the most important driver for fungivores in urban sites. Therefore, the proportion of fungivores is supposed to be driven by mineralized fertilization (Ferris et al., 2004). Compared to other nematode trophic groups, fungivore nematodes were positively related to the spatial distance of the study sites in our path analysis. This suggests that fungivore nematodes may be less sensitive to habitat fragmentation than other nematode feeding groups, potentially



**Fig. 6.** Functional guilds of nematode ASVs associated with ecosystem stability. (A) aggregation network of the functionally cohesive group of nematodes that are most associated with ecosystem stability. The thickness of links represents the cohesion of two nodes. The nodes are nematode ASVs colored by feeding guilds. The size of each node represents the degree of each node. (B) Proportion of the detected functional guild in urban and non-urban ecosystems. (C) Relationship between the proportion of nematode functional guild and ecosystem stability. (D) Regression tree showing the effects of environmental factors on the relationships between ecosystem stability and the proportion of detected functional guild. \*\*\*,  $p < 0.001$  as indicated by Wilcoxon test. Lines indicate significant relationships between the proportion of nematode feeding guilds and ecosystem stability.

due to the abundant fungal food sources (Anthony et al., 2023; Tedersoo et al., 2014). It is worth noting that our findings were consistent across global and continental scales and valid for the traditional and molecular methods. Given the complementary effects of the two techniques, we provide the possibilities of concurrently exploiting both methods that are used for soil nematode community analysis (Donhauser et al., 2023).

#### 4.2. Urban effects on the association between nematodes and ecosystem stability

Nematode functional guilds most strongly associated with temporal ecosystem stability comprised bacterivores, fungivores, herbivores, and omnivores in urban sites partly supporting our second hypothesis. This is

consistent with earlier findings that niche complementarity among soil animals enhances ecosystem functions (Hättenschwiler et al., 2005; Heemsbergen Hal, 2004), suggesting that nematode functional groups complement each other in supporting ecosystem functions and long-term ecosystem stability. Further, nematodes of the same feeding group may detrimentally affect each other due to competition and this may contribute to reduced ecosystem stability (Zheng et al., 2022). The competitive interactions were documented to be an important process in shaping nematode species coexistence (Ettema, 1998). Moreover, the mechanisms were also evidenced by relevant experiments that found the substances controlling interspecific competition in nematodes (Bose et al., 2014).

The fact that the relative abundance of nematode feeding groups, except for predators, correlated with temporal ecosystem stability in urban but not in non-urban ecosystems may have been due to the fact that non-urban ecosystems are generally less variable in time than urban ecosystems (Price et al., 2019). In contrast to bacterivore nematodes, fungivores, herbivores, and omnivores were negatively correlated with temporal ecosystem stability. Among nematode feeding groups, bacterivores are most opportunistic and quickly respond to external inputs (Bongers and Bongers, 1998), and therefore may not reflect long-term changes in plant community composition (Lorenz and Lal, 2009). Their ability to quickly respond to changes in resource availability and environmental changes likely explains their higher proportion in urban than non-urban soils and thereby their association with lower temporal ecosystem stability.

Our results that the relative abundance of nematode functional guilds, a sub-community detected to collectively contribute to ecosystem stability by the *de novo* method, was significantly correlated with temporal ecosystem stability in both non-urban and urban ecosystems also support our second hypothesis. Further, it confirms the usefulness of functional guilds in reflecting ecosystem properties. As discussed above, nematodes of the same feeding group may hamper each other due to occupying similar niches (Vestergård et al., 2019), and this may compromise their contributions to ecosystem processes and in the long term to ecosystem stability. Further, nematodes can occupy different niches, and contribute to greater nutrient use efficiency. Resource depletion may result in greater diversity as nematodes are forced to seek additional food sources (Neher, 2010). Climate variables, i.e., MAP, as well as latitude, longitude, and population density, were the main classifiers of the associations between temporal ecosystem stability and the relative abundance of nematode functional guilds. This is consistent with the idea that ecosystem stability emerges from the functional organization of food webs (de Castro et al., 2021). Moreover, less precipitation (<1266 mm) and low latitudes (<38°) modified the association between temporal ecosystem stability and nematode functional guilds. It is documented that conclusions about the effects of urbanization on nematodes were not elucidated (Szabó et al., 2023). As increased population density is associated with an increased demand for resources, it may potentially result in over-exploitation of resources in soil (Smith et al., 2016), thus impacting the soil nematodes. This reinforces the importance of sustainable management of urban soils, which only is possible by improved knowledge of the response of soil biota, such as nematodes, contributing to the main functions of soils.

## 5. Conclusion

Combining global data and data from cities across China, the results of our study suggest that urbanization is associated with a reduced abundance of key functional guilds of soil biota, resulting in simplified nematode communities and reduced temporal ecosystem stability. These results were due to the detrimental effects of urbanization on edaphic factors, such as the soil structure and nutrients. The findings call for actions to pay more attention to soil biodiversity in cities, especially for key functional guilds. For example, by improving the habitat integrity and resource availability of those taxa. Our assessment of soil

properties and nematode communities as soil biodiversity indicators provides novel insights into the drivers and regulating screws for management strategies towards more healthy soils in cities.

## CRedit authorship contribution statement

**Xin Gong:** Writing – review & editing, Writing – original draft, Visualization, Data curation. **Zhihong Qiao:** Methodology, Investigation. **Haifeng Yao:** Methodology, Investigation. **Dan Zhao:** Writing – review & editing, Software, Data curation. **Nico Eisenhauer:** Writing – review & editing, Supervision. **Stefan Scheu:** Writing – review & editing, Supervision. **Chao Liang:** Writing – review & editing. **Manqiang Liu:** Writing – review & editing. **Yong-Guan Zhu:** Writing – review & editing, Supervision, Conceptualization. **Xin Sun:** Writing – review & editing, Supervision, Funding acquisition, Conceptualization.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

Data will be made available on request.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.soilbio.2023.109297>.

## References

- Almende, B., Thieurmel, B., Robert, T., 2022. visNetwork: Network Visualization using “vis.js” Library. R package version 2.1.2. <https://CRAN.R-project.org/package=visNetwork>.
- Anthony, M.A., Bender, S.F., van der Heijden, M.G.A., 2023. Enumerating soil biodiversity. Proceedings of the National Academy of Sciences 120. <https://doi.org/10.1073/pnas.2304663120>.
- Archer, E., 2020. rfPermute: estimate permutation p-values for random forest importance metrics. In: R Package Version 2.1.81.
- Beninde, J., Veith, M., Hochkirch, A., 2015. Biodiversity in cities needs space: a meta-analysis of factors determining intra-urban biodiversity variation. Ecology Letters 18, 581–592. <https://doi.org/10.1111/ele.12427>.
- Bongers, T., 1990. The maturity index: an ecological measure of environmental disturbance based on nematode species composition. Oecologia 83, 14–19. <https://doi.org/10.1007/BF00324627>.
- Bongers, T., Bongers, M., 1998. Functional diversity of nematodes. Applied Soil Ecology 10, 239–251.
- Borkovec, M., Madin, N., 2019. Ggparty: “Ggplot” Visualizations for the “Partykit” Package. R Package Version 1.0.0. <https://CRAN.R-project.org/package=ggparty>.
- Bose, N., Meyer, J.M., Yim, J.J., Mayer, M.G., Markov, G.V., Ogawa, A., Schroeder, F.C., Sommer, R.J., 2014. Natural variation in dauer pheromone production and sensing supports intraspecific competition in nematodes. Current Biology 24, 1536–1541. <https://doi.org/10.1016/j.cub.2014.05.045>.



- Cesarz, S., Reich, P.B., Scheu, S., Ruess, L., Schaefer, M., Eisenhauer, N., 2015. Nematode functional guilds, not trophic groups, reflect shifts in soil food webs and processes in response to interacting global change factors. *Pedobiologia* 58, 23–32. <https://doi.org/10.1016/j.pedobi.2015.01.001>.
- Creamer, R.E., Hannula, S.E., Leeuwen, J.P.V., Stone, D., Rutgers, M., Schmelz, R.M., Ruiter, P.C.d., Hendriksen, N.B., Bolger, T., Bouffaud, M.L., Buee, M., Carvalho, F., Costa, D., Dirilgen, T., Francisco, R., Griffiths, B.S., Griffiths, R., Martin, F., Silva, P. M. da, Mendes, S., Morais, P.V., Pereira, C., Philippot, L., Plassart, P., Redecker, D., Römcke, J., Sousa, J.P., Wouterse, M., Lemanceau, P., 2016. Ecological network analysis reveals the inter-connection between soil biodiversity and ecosystem function as affected by land use across Europe. *Applied Soil Ecology* 97, 112–124. <https://doi.org/10.1016/j.apsoil.2015.08.006>.
- de Castro, F., Adl, S.M., Allesina, S., Bardgett, R.D., Bolger, T., Dalzell, J.J., Emmerson, M., Fleming, T., Garlaschelli, D., Grilli, J., Hannula, S.E., de Vries, F., Lindo, Z., Maule, A.G., Öpik, M., Rillig, M.C., Veresoglou, S.D., Wall, D.H., Caruso, T., 2021. Local stability properties of complex, species-rich soil food webs with functional block structure. *Ecology and Evolution* 11, 16070–16081. <https://doi.org/10.1002/eec3.8278>.
- Decaens, T., 2010. Macroecological patterns in soil communities. *Global Ecology and Biogeography* 19, 287–302. <https://doi.org/10.1111/j.1466-8238.2009.00517.x>.
- Delgado-Baquerizo, M., Eldridge, D.J., Liu, Y.R., Sokoya, B., Wang, J.T., Hu, H.W., He, J. Z., Bastida, F., Moreno, J.L., Bamigboye, A.R., Blanco-Pastor, J.L., Cano-Díaz, C., Illán, J.G., Makhalyane, T.P., Siebe, C., Trivedi, P., Zaady, E., Verma, J.P., Wang, L., Wang, J., Grebenc, T., Peñaloza-Bojacá, G.F., Nahberger, T.U., Teixido, A. L., Zhou, X.Q., Berdugo, M., Duran, J., Rodríguez, A., Zhou, X., Alfaro, F., Abades, S., Plaza, C., Rey, A., Singh, B.K., Tedersoo, L., Fierer, N., 2021. Global homogenization of the structure and function in the soil microbiome of urban greenspaces. *Science Advances* 7, eabg5809. <https://doi.org/10.1126/sciadv.abg5809>.
- Donhauser, J., Briones, M.J.I., Mikola, J., Jones, D.L., Eder, R., Filser, J., Frossard, A., Krogh, P.H., Sousa, J.P., Cortet, J., Desie, E., Domene, X., Djuric, S., Hackenberger, D., Jimenez, J.J., Iamandi, M., Rissmann, C., Schmidt, O., Shanskiy, M., Silfver, T., Vancampenhout, K., Vasutova, M., Velizarova, E., Frey, B., 2023. Extracting DNA from soil or directly from isolated nematodes indicate dissimilar community structure for Europe-wide forest soils. *Soil Biology and Biochemistry* 185, 109154. <https://doi.org/10.1016/j.soilbio.2023.109154>.
- Eisenhauer, N., Antunes, P.M., Bennett, A.E., Birkhofer, K., Bissett, A., Bowker, M.A., Caruso, T., Chen, B., Coleman, D.C., Boer, W., de Ruiter P de, DeLuca, T.H., Frati, F., Griffiths, B.S., Hart, M.M., Hättenschwiler, S., Haimi, J., Heethoff, M., Kaneko, N., Kelly, L.C., Leinaas, H.P., Lindo, Z., Macdonald, C., Rillig, M.C., Ruess, L., Scheu, S., Schmidt, O., Seastedt, T.R., Straalen, N.M. van, Tiuonov, A.V., Zimmer, M., Powell, J. R., 2017. Priorities for research in soil ecology. *Pedobiologia* 63, 1–7. <https://doi.org/10.1016/j.PEDOBIO.2017.05.003>.
- Erktan, A., Or, D., Scheu, S., 2020. The physical structure of soil: determinant and consequence of trophic interactions. *Soil Biology and Biochemistry* 148, 107876. <https://doi.org/10.1016/j.soilbio.2020.107876>.
- Ettema, C.H., 1998. Soil nematode diversity: species coexistence and ecosystem function. *Journal of Nematology* 30, 159–169.
- Fahrig, L., 2003. Effects of habitat fragmentation on biodiversity. *Annual Review of Ecology Evolution and Systematics* 34, 487–515. <https://doi.org/10.1146/annurev.ecolsys.34.011802.132419>.
- FAO, 2020. State of Knowledge of Soil Biodiversity - Status, Challenges and Potentialities. ITPS, GSBI, SCBD, EC. FAO, Rome. <https://doi.org/10.4060/cb1928en>. Report 2020.
- Ferris, H., Venette, R.C., Scow, K.M., 2004. Soil management to enhance bacterivore and fungivore nematode populations and their nitrogen mineralisation function. *Applied Soil Ecology* 25, 19–35. <https://doi.org/10.1016/j.apsoil.2003.07.001>.
- Fick, S.E., Hijmans, R.J., 2017. WorldClim 2: new 1-km spatial resolution climate surfaces for global land areas. *International Journal of Climatology* 37, 4302–4315. <https://doi.org/10.1002/joc.5086>.
- Fierer, N., Strickland, M.S., Liptzin, D., Bradford, M.A., Cleveland, C.C., 2009. Global patterns in belowground communities. *Ecology Letters* 12, 1238–1249. <https://doi.org/10.1111/j.1461-0248.2009.01360.x>.
- Gebremikael, M.T., Steel, H., Buchan, D., Bert, W., De Neve, S., 2016. Nematodes enhance plant growth and nutrient uptake under C and N-rich conditions. *Scientific Reports* 6, 1–10. <https://doi.org/10.1038/srep32862>.
- Hättenschwiler, S., Tiuonov, A.V., Scheu, S., 2005. Biodiversity and litter decomposition in terrestrial ecosystems. *Annual Review of Ecology Evolution and Systematics* 36, 191–218. <https://doi.org/10.1146/annurev.ecolsys.36.112904.151932>.
- Heemsbergen Hal, V., 2004. Biodiversity effects on soil processes explained by interspecific functional dissimilarity. *Science* 306, 8–10. <https://doi.org/10.1126/science.1101865>.
- Himbeek, R. Van, Giullia, S., Elsen, S. Van Den, Oude, J., Karst, S., Visser, J., Helder, J., 2024. A full-length SSU rRNA-based workflow for high-resolution monitoring of nematode communities reveals direct and indirect responses to plant-based manipulations. *Soil Biology and Biochemistry* 189, 109263. doi:10.1016/j.soilbio.2023.109263.
- Kong, X., Dao, T.H., Qin, J., Qin, H., Li, C., Zhang, F., 2009. Effects of soil texture and land use interactions on organic carbon in soils in North China cities' urban fringe. *Geoderma* 154, 86–92. <https://doi.org/10.1016/j.geoderma.2009.09.016>.
- Lehmann, J., Bossio, D.A., Kögel-Knabner, I., Rillig, M.C., 2020. The concept and future prospects of soil health. *Nature Reviews Earth & Environment* 1, 544–553. <https://doi.org/10.1038/s43017-020-0080-8>.
- Lepczyk, C.A., Aronson, M.F.J., Evans, K.L., Goddard, M.A., Lerman, S.B., Macivor, J.S., 2017. Biodiversity in the city: fundamental questions for understanding the ecology of urban green spaces for biodiversity conservation. *BioScience* 67, 799–807. <https://doi.org/10.1093/biosci/bix079>.
- Li, X., Liu, T., Li, H., Geisen, S., Hu, F., Liu, M., 2022. Management effects on soil nematode abundance differ among functional groups and land-use types at a global scale. *Journal of Animal Ecology* 91, 1770–1780. <https://doi.org/10.1111/1365-2656.13744>.
- Lorenz, K., Lal, R., 2009. Biogeochemical C and N cycles in urban soils. *Environment International* 35, 1–8. <https://doi.org/10.1016/j.envint.2008.05.006>.
- Lu, R., 1999. *Analysis Methods of Soil Agricultural Chemistry*. China Agricultural Science and Technology Press, Beijing.
- Marcotullio, P.J., Braimoh, A.K., Onishi, T., 2008. The impact of urbanization on soils. In: Braimoh, A.K., Vlek, P.L.G. (Eds.), *Land Use and Soil Resources*. Springer Netherlands, Dordrecht, pp. 201–250. [https://doi.org/10.1007/978-1-4020-6778-5\\_10](https://doi.org/10.1007/978-1-4020-6778-5_10).
- McDonald, R.I., Mansur, A.V., Ascensão, F., Colbert, M., Crossman, K., Elmqvist, T., Gonzalez, A., Güneralp, B., Haase, D., Hamann, M., Hillel, O., Huang, K., Kahnt, B., Maddox, D., Pacheco, A., Pereira, H.M., Seto, K.C., Simkin, R., Walsh, B., Werner, A. S., Ziter, C., 2020. Research gaps in knowledge of the impact of urban growth on biodiversity. *Nature Sustainability* 3, 16–24. <https://doi.org/10.1038/s41893-019-0436-6>.
- Morriën, E., 2016. Understanding soil food web dynamics, how close do we get? *Soil Biology and Biochemistry* 102, 10–13. <https://doi.org/10.1016/j.soilbio.2016.06.022>.
- Neher, D.A., 2010. Ecology of plant and free-living nematodes in natural and agricultural soil. *Annual Review of Phytopathology* 48, 371–394. <https://doi.org/10.1146/annurev-phyto-073009-114439>.
- Oksanen, J., Blanchet, F.G., Friendly, M., Kindt, R., Legendre, P., McGlenn, D., Minchin, P.R., O'Hara, R.B., Simpson, A.V., Solymos, P., Stevens, M.H.H., Szoecs, E., Wagner, H., 2020. *Vegan: community ecology package*. R Package Version 2, pp. 5–7. <https://doi.org/10.4135/9781412971874.n145>.
- Park, S.J., Taylor, R.A.J., Grewal, P.S., 2013. Spatial organization of soil nematode communities in urban landscapes: Taylor's Power Law reveals life strategy characteristics. *Applied Soil Ecology* 64, 214–222. <https://doi.org/10.1016/j.apsoil.2012.11.001>.
- Pavao-Zucker, M.A., Coleman, D.C., 2007. Urbanization alters the functional composition, but not taxonomic diversity, of the soil nematode community. *Applied Soil Ecology* 35, 329–339. <https://doi.org/10.1016/j.apsoil.2006.07.008>.
- Pickett, S.T.A., Cadenasso, M.L., 2009. Altered resources, disturbance, and heterogeneity: a framework for comparing urban and non-urban soils. *Urban Ecosystems* 12, 23–44. <https://doi.org/10.1007/s11252-008-0047-x>.
- Porazinska, D.L., Giblin-Davis, R.M., Faller, L., Farmerie, W., Kankani, N., Morris, K., Powers, T.O., Tucker, A.E., Sung, W., Thomas, W.K., 2009. Evaluating high-throughput sequencing as a method for metagenomic analysis of nematode diversity. *Molecular Ecology Resources* 9, 1439–1450. <https://doi.org/10.1111/j.1755-0998.2009.02611.x>.
- Potapov, A.M., Rozanova, O.L., Semenina, E.E., Leonov, V.D., Belyakova, O.I., Bogatyeva, V.Y., Degtyarev, M.I., Esaulov, A.S., Korotkevich, A.Y., Kudrin, A.A., Malysheva, E.A., Mazei, Y.A., Tsurikov, S.M., Zuev, A.G., Tiuonov, A.V., 2021. Size compartmentalization of energy channeling in terrestrial belowground food webs. *Ecology* 102, 1–14. <https://doi.org/10.1002/ecy.3421>.
- Price, E.L., Sertić Perić, M., Romero, G.Q., Kratina, P., 2019. Land use alters trophic redundancy and resource flow through stream food webs. *Journal of Animal Ecology* 88, 677–689. <https://doi.org/10.1111/1365-2656.12955>.
- Quast, C., Pruesse, E., Yilmaz, P., Gerken, J., Schweer, T., Yarza, P., Peplies, J., Glöckner, F.O., 2013. The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. *Nucleic Acids Research* 41, 590–596. <https://doi.org/10.1093/nar/gks1219>.
- R Core Team, 2022. R: a Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. URL: <http://www.R-project.org/>.
- Roscoe, R., Buurman, P., Velthorst, E.J., 2000. Disruption of soil aggregates by varied amounts of ultrasonic energy in fractionation of organic matter of a clay Latosol: carbon, nitrogen and  $\delta^{13}C$  distribution in particle-size fractions. *European Journal of Soil Science* 51, 445–454. <https://doi.org/10.1046/j.1365-2389.2000.00321.x>.
- Sanchez, G., Trinchera, L., Russolillo, G., 2015. *plsmp: tools for partial least squares path modeling (PLS-PM)*. In: R Package Version 0.4.9.
- Scharenbroch, B.C., Lloyd, J.E., Johnson-Maynard, J.L., 2005. Distinguishing urban soils with physical, chemical, and biological properties. *Pedobiologia* 49, 283–296. <https://doi.org/10.1016/j.pedobi.2004.12.002>.
- Shan, X., Goyal, A., Gregor, R., Cordero, O.X., 2023. Annotation-free discovery of functional groups in microbial communities. *Nature Ecology & Evolution* 7, 716–724. <https://doi.org/10.1038/s41559-023-02021-z>.
- Smith, P., House, J.I., Bustamante, M., Sobotková, J., Harper, R., Pan, G., West, P.C., Clark, J.M., Adhya, T., Rumpel, C., Paustian, K., Kuikman, P., Cotrufo, M.F., Elliott, J.A., McDowell, R., Griffiths, R.I., Asakawa, S., Bondeau, A., Jain, A.K., Meersmans, J., Pugh, T.A.M., 2016. Global change pressures on soils from land use and management. *Global Change Biology* 22, 1008–1028. <https://doi.org/10.1111/gcb.13068>.
- Sun, X., Liddicoat, C., Tiuonov, A., Wang, B., Zhang, Y., Lu, C., Li, Z., Scheu, S., Breed, M. F., Geisen, S., Zhu, Y.-G., 2023. Harnessing soil biodiversity to promote human health in cities. *Npj Urban Sustainability* 3, 5. <https://doi.org/10.1038/s42949-023-00086-0>.
- Szabó, B., Korányi, D., Gallé, R., Lóvei, G.L., Bakonyi, G., Batáry, P., 2023. Urbanization decreases species richness, and increases abundance in dry climates whereas decreases in wet climates: a global meta-analysis. *The Science of the Total Environment* 859, 160145. <https://doi.org/10.1016/j.scitotenv.2022.160145>.
- Tedersoo, L., Tedersoo, L., Bahram, M., Pölme, S., Kõljalg, U., Yorou, N.S., Wijesundera, R., Ruiz, L.V., Vasco-palacios, A.M., Thu, P.Q., Suija, A., Smith, M.E.,

- Sharp, C., Saluveer, E., Saitta, A., Rosas, M., Riit, T., Ratkowsky, D., Pritsch, K., Põldmaa, K., Otsing, E., Nohra, E., Njouonkou, A.L., Nilsson, R.H., Morgado, L.N., Mayor, J., May, T.W., Majuakim, L., Lodge, D.J., Lee, S.S., Larsson, K., Kohout, P., Hosaka, K., Hiiesalu, I., Henkel, T.W., Harend, H., Guo, L., Greslebin, A., Grelet, G., Geml, J., Gates, G., Dunstan, W., Dunk, C., Drenkhan, R., Dearnaley, J., Kesel, A. De, Dang, T., Chen, X., Buegger, F., Brearley, F.Q., 2014. Global diversity and geography of soil fungi. *Science* 346, 1078. <https://doi.org/10.1126/science.1256688>.
- Topalović, O., Geisen, S., 2023. Nematodes as suppressors and facilitators of plant performance. *New Phytologist* 2305–2312. <https://doi.org/10.1111/nph.18925>.
- van den Hoogen, J., Geisen, S., Routh, D., Ferris, H., Traunspurger, W., Wardle, D.A., de Goede, R.G.M., Adams, B.J., Ahmad, W., Andriuzzi, W.S., Bardgett, R.D., Bonkowski, M., Campos-Herrera, R., Cares, J.E., Caruso, T., de Brito Caixeta, L., Chen, X., Costa, S.R., Creamer, R., Mauro da Cunha Castro, J., Dam, M., Djigal, D., Escuer, M., Griffiths, B.S., Gutiérrez, C., Hohberg, K., Kalinkina, D., Kardol, P., Kergunteuil, A., Korthals, G., Krashevskaya, V., Kudrin, A.A., Li, Q., Liang, W., Magilton, M., Marais, M., Martín, J.A.R., Matveeva, E., Mayad, E.H., Mulder, C., Mullin, P., Neilson, R., Nguyen, T.A.D., Nielsen, U.N., Okada, H., Rius, J.E.P., Pan, K., Peneva, V., Pellissier, L., Carlos Pereira da Silva, J., Pitteloud, C., Powers, T. O., Powers, K., Quist, C.W., Rasmann, S., Moreno, S.S., Scheu, S., Setälä, H., Sushchuk, A., Tiunov, A.V., Trap, J., van der Putten, W., Vestergård, M., Villenave, C., Waeyenberge, L., Wall, D.H., Wilschut, R., Wright, D.G., Yang, J., Crowther, T.W., 2019. Soil nematode abundance and functional group composition at a global scale. *Nature* 572, 194–198. <https://doi.org/10.1038/s41586-019-1418-6>.
- van den Hoogen, J., Geisen, S., Wall, D.H., Wardle, D.A., Traunspurger, W., de Goede, R.G.M., Adams, B.J., Ahmad, W., Ferris, H., Bardgett, R.D., Bonkowski, M., Campos-Herrera, R., Cares, J.E., Caruso, T., de Brito Caixeta, L., Chen, X., Costa, S.R., Creamer, R., da Cunha e Castro, J.M., Dam, M., Djigal, D., Escuer, M., Griffiths, B.S., Gutiérrez, C., Hohberg, K., Kalinkina, D., Kardol, P., Kergunteuil, A., Korthals, G., Krashevskaya, V., Kudrin, A.A., Li, Q., Liang, W., Magilton, M., Marais, M., Martín, J.A.R., Matveeva, E., Mayad, E.H., Mzough, E., Mulder, C., Mullin, P., Neilson, R., Nguyen, T.A.D., Nielsen, U.N., Okada, H., Rius, J.E.P., Pan, K., Peneva, V., Pellissier, L., da Silva, J.C.P., Pitteloud, C., Powers, T.O., Powers, K., Quist, C.W., Rasmann, S., Moreno, S.S., Scheu, S., Setälä, H., Sushchuk, A., Tiunov, A.V., Trap, J., van der Putten, W., Vestergård, M., Villenave, C., Waeyenberge, L., Wall, D.H., Wilschut, R., Wright, D.G., Yang, J., Routh, D., Crowther, T.W., 2020. A global database of soil nematode abundance and functional group composition. *Scientific Data* 7, 103. <https://doi.org/10.1038/s41597-020-0437-3>.
- Vestergård, M., Dam, M., Mortensen, L.H., Dyckmans, J., Christensen, B.T., 2019. Natural <sup>13</sup>C abundance reveals age of dietary carbon sources in nematode trophic groups. *Soil Biology and Biochemistry* 130, 1–7. <https://doi.org/10.1016/j.soilbio.2018.11.024>.
- Wall, D.H., Nielsen, U.N., Six, J., 2015. Soil biodiversity and human health. *Nature* 528, 69–76. <https://doi.org/10.1038/nature15744>.
- Wardle, D.A., Bardgett, R.D., Klironomos, J.N., Setälä, H., van der Putten, W.H., Wall, D. H., 2004. Ecological linkages between aboveground and belowground biota. *Science* 304, 1629–1633. <https://doi.org/10.1126/science.1094875>.
- Wu, L., Chen, H., Chen, D., Wang, S., Wu, Y., Wang, B., Liu, S., Yue, L., Yu, J., Bai, Y., 2023. Soil biota diversity and plant diversity both contributed to ecosystem stability in grasslands. *Ecology Letters* 1–11. <https://doi.org/10.1111/ele.14202>.
- Yang, G., Wagg, C., Veresoglou, S.D., Hempel, S., Rillig, M.C., 2018. How soil biota drive ecosystem stability. *Trends in Plant Science* 23, 1057–1067. <https://doi.org/10.1016/j.tplants.2018.09.007>.
- Yao, H., Li, Z., Geisen, S., Qiao, Z., Breed, M.F., Sun, X., 2023. Degree of urbanization and vegetation type shape soil biodiversity in city parks. *The Science of the Total Environment* 899, 166437. <https://doi.org/10.1016/j.scitotenv.2023.166437>.
- Yeates, G.W., Bongers, T., De Goede, R.G., Freckman, D.W., Georgieva, S.S., 1993. Feeding habits in soil nematode families and genera—an outline for soil ecologists. *Journal of Nematology* 25, 315–331.
- Zhao, J., Neher, D.A., 2013. Soil nematode genera that predict specific types of disturbance. *Applied Soil Ecology* 64, 135–141. <https://doi.org/10.1016/j.apsoil.2012.11.008>.
- Zheng, J., Dini-Andreote, F., Luan, L., Geisen, S., Xue, J., Li, H., Sun, B., Jiang, Y., 2022. Nematode predation and competitive interactions affect microbe-mediated phosphorus dynamics. *mBio* 13, 1–16. <https://doi.org/10.1128/mbio.03293-21>.