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RESEARCH ARTICLE

Effects of host niche and genotype on the diversity and community assembly of the fungal community in peas (*Pisum sativum* L.)

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Highlights

- Niches shaped the pea fungal communities more than genotypes.
- Fungal β-diversity was mainly driven by species replacement.
- Niche- and genotype-based communities presented deterministic and stochastic assembly, respectively.
- The migration rate from exogenous to endogenous niches was low.

Abstract

Fungi play crucial roles in nutrient acquisition, plant growth promotion, and the enhancement of resistance to both abiotic and biotic stresses. However, studies on the fungal communities associated with peas (*Pisum sativum* L.) remain limited. In this study, we systematically investigated the ecological effects of host niches (soil, root, stem, leaf, and pod) and genotypes on the diversity and composition of fungal communities in peas using a multi-level approach that encompassed pattern recognition (β-diversity decomposition), mechanism validation (neutral community model testing), and dynamic tracking methods (migration pathway source-tracking). The results revealed that the dominant fungal phyla across niches and genotypes were Ascomycota, Basidiomycota, and Mortierellomycota, and the community structures of the soil–plant continuum were primarily determined by the pea niches rather than genotypes. β-diversity decomposition was largely attributed to species replacement rather than richness differences, indicating strong niche specificity and microbial replacement across microhabitats. Neutral model analysis revealed that stochastic processes influenced genotype-associated communities, while deterministic processes played a dominant role in niche-based community assembly. Source-tracking analysis identified niche-to-niche fungal migration, with *Erysiphe*, *Fusarium*, *Cephaliophora*, *Ascobolus*, *Alternaria*, and *Aspergillus* as the key genera. Migration rates from exogenous to endogenous niches were low (1.3–61.5%), whereas those within exogenous (64.4–83.7%) or endogenous (73.9–96.4%) compartments were much higher, suggesting that the pea epidermis acts as a selective barrier that filters and enriches microbial communities prior to internal colonization. This study provides comprehensive insights into the mechanisms of host filtering, enrichment and microbial sourcing, which increases our understanding of the assembly rules of the pea-associated fungal microbiome.

Keywords: β-diversity decomposition, fungal community assembly, pea, source-sink relationships, host niche, genotype

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1. Introduction

Fungi play a vital role in soil ecosystems by contributing to nutrient cycling, organic matter decomposition, soil structure preservation, and plant health (Nguyen *et al.* 2016; Guo *et al.* 2024). Their diverse roles ensure crop resilience and enhance soil fertility, thus supporting sustainable agricultural practices (Schmidt *et al.* 2019). The β-diversity of fungal communities across different microhabitats is shaped by a complex interplay of environmental filtering, dispersal limitation, and other ecological mechanisms (Zhao *et al.* 2024). Changes in β-diversity are driven by two processes: 1) species replacement, where one species is substituted by another, leading to shifts in community composition without changes in overall species richness; and 2) richness differences, which involve variations in the presence or absence of species across different sites, resulting in discrepancies in species abundance and diversity (Guo *et al.* 2024). These patterns provide important insights into the underlying community assembly processes. Specifically, community assembly is a key driver of microbial diversity, and it is jointly driven by both deterministic processes (environmental filtering and biotic interactions) and stochastic processes (dispersal limitation and ecological drift) (Wan *et al.* 2023). A previous study demonstrated that deterministic processes play an increasingly dominant role in shaping β-diversity with N addition, leading to distinct changes in the aboveground plant and belowground community compositions (Zhao *et al.* 2024).

Peas (*Pisum sativum* L.) belong to the family Leguminosae and are widely cultivated food legumes valued for their high contents of proteins, starches, vitamins, and minerals; and they serve as an important source of food and animal feed globally (Boakye *et al.* 2022; Calderon and Dangi 2024; Cheng *et al.* 2024). Peas currently rank as the fourth most widely cultivated legume crop globally in terms of harvested area, after soybeans, common beans, and chickpeas (Yang *et al.* 2022). Plant-associated microorganisms contribute significantly to plant health and performance by facilitating water and nutrient uptake, enhancing stress tolerance, suppressing pathogens, and reducing herbivory (Pii *et al.* 2015). While the bacterial community structure was primarily shaped by plant niches (leaves, stems, roots, and soil), fungal communities within leaf niches were greatly impacted by the host genotype. Moreover, bacterial diversity was found to decrease progressively from the soil to internal *Populus* tree compartments, suggesting that environmental filtering imposes strong selective forces on microbial communities (Cregger *et al.* 2018). *Rhizobium*, *Bacteroides*, and *Pseudomonas* are the dominant pea root and nodule endogenous bacteria (Lv *et al.* 2021; Hao *et al.* 2024). However, research on fungal ecological mechanisms is lacking, and the relative contributions of stochastic and deterministic processes and migration patterns in the soil–plant continuum remain unclear. Insights into the structure and assembly processes of pea-associated fungal communities can enhance yield and resource efficiency, so they offer scientific guidance for optimized pea cultivation.

Source tracking analysis is an ecological method used to trace the origin, transmission pathways, and colonization processes of microorganisms in specific niches. Applying this approach to the soil–plant continuum can elucidate the community composition and the source-sink relationships of microbiomes, thereby revealing the ecological processes underlying the assembly of plant microbial communities (Xiong *et al.* 2021; Zhang L *et al.* 2022). To date, our understanding of fungal community diversity, community assembly processes, and migration patterns across different pea genotypes and niches remains limited.

In this study, we aimed to establish an analytical framework that encompasses pattern recognition (β-diversity decomposition), mechanism validation (neutral community process testing), and dynamic tracking (source-tracking of migration pathways) to systematically investigate the interaction mechanisms among genotype, pea niche and fungal community. This study aimed to answer four questions: (1) How does the pea-associated fungal β-diversity change with niches and genotypes? (2) Is pea-associated fungal β-diversity driven by richness differences or species replacement? (3) Is the assembly of pea-associated fungal communities dominated by deterministic or stochastic processes? (4) How do pea-associated fungal communities migrate across different niches? Understanding the interactions between various niches of peas and their associated fungi can strengthen our knowledge of the ecological mechanisms underlying fungal community assembly in peas.

2. Materials and methods

2.1. Site description and sample collection

The field experiment was conducted in Cixi City, Ningbo, Zhejiang Province, China (34°11′N, 121°16′E, 4.5 m above sea level). This region is characterized by saline-alkaline nocturnal tidal soil and a subtropical monsoon climate, with annual mean precipitation of 1,272.8 mm and an average annual temperature of 16.3°C. The experimental site area was 6 m×2.25 m, with three replicates established for each pea genotype. Before sowing, the field was covered with a white plastic film, and each plot was fertilized with 1 kg of compound fertilizer (N-P_2O_5-K_2O, 15:15:15). Seven pea genotypes (Zhongqin1 (ZQ1), Zhongwan9 (ZW9), Rongtao9 (RT9), Lümiao9 (LM9), Gailiangzhongwan4 (GL4), Zhewan2 (ZW2), and Zhongwan6 (ZW6)) were sown on September 12, 2023. Plant height, weight, and yield differed significantly among the seven genotypes (Appendix A).

Samples were collected from eight distinct organ-associated niches and microenvironments, namely bulk soil (BS), rhizosphere soil (RS), rhizosheath soil (R), root endosphere (RE), phylloplane (P), leaf endosphere (LE), stem endosphere (SE), and pod (PO), at the mature stage of the peas (on November 9, 2023). A soil auger was used to collect bulk soil samples at 0–20 cm depth. Full-plant samples were obtained using a spade. Rhizosphere soil samples were collected by vigorously shaking the roots after removing the loosely bound soil. The plant samples were placed in sterile sample bags and transported on ice to the laboratory.

2.2. Sample preparation and DNA extraction

Rhizosheath soil and phylloplane samples were collected by placing fresh leaves and roots in sterile sample bags containing phosphate-buffered saline (PBS) solution and ultrasonically shaking and cleaning at 40 kHz for 30 min. Root samples in PBS solution were transferred to a 50 mL centrifuge tube to collect the settled soil by centrifugation, and leaf samples in PBS solution were filtered on a 0.22 μm filter membrane to collect phylloplane fungi. The ultrasonically washed leaves and roots, as well as stems and pods, were immersed in 70% ethanol for 5 min, followed by soaking in 2.5% sodium hypochlorite solution for 5 min and then 70% ethanol for 30 s. Samples were washed five times with sterile water to remove epibiotic microbes (Yang *et al.* 2023). To evaluate the efficacy of surface sterilization, the sterilized plant samples were aseptically sectioned using a sterile scalpel and subsequently inoculated onto Potato Dextrose Agar medium, with blank medium as a negative control. The inoculated plates were incubated at 25°C for 5 d under controlled conditions. The absence of microbial colony formation on all cultured specimens confirmed the effectiveness of the surface sterilization protocol. In total, 168 samples were collected representing seven genotypes (ZQ1, ZW9, RT9, LM9, GL4, ZW2, and ZW6)×eight niches (BS, RS, R, RE, P, LE, SE, and PO)×three repetitions (Fig. 1).

The sterile samples of leaves, roots, stems, and pods, and the 0.22 μm filter membrane containing phylloplane fungi were separately ground into powder with liquid nitrogen, each using a sterilized mortar and pestle before the DNA was extracted (Beckers *et al.* 2017; Yang *et al.* 2023). DNA from the soil and plant tissues was extracted using the FastDNA SPIN Kit (MP Biomedicals, California, USA) (Chen *et al.* 2020). The quality and quantity of the DNA were examined using a NanoDrop spectrophotometer (A260/A280 ratio≥1.8 for purity; concentration≥20 ng μL^{-1}) (NanoDrop Technologies, Wilmington, DE, USA) and agarose gel electrophoresis (intact high-molecular-weight bands without smearing).

2.3. Fungal ITS gene amplification and sequencing

The fungal ITS1 gene region was amplified using barcode tagged-templates and primers ITS5-1737F (5′-GGAAGTAAAAGTCGTAACAA-3′) and ITS2-2043R (5′-GCTGCGTTCTTCATCGATGC-3′) (Zhang *et al.* 2021). PCR amplification was performed using a 50 μL reaction mixture comprising 25 μL of novoprotein (2×*Taq* Master Mix, E005-02, Novoprotein Scientific Inc., Shanghai, China), 18 μL of ddH_2O, 2 μL of each primer, and 3 μL of template DNA. Amplification was conducted with the following program: an initial period at 95°C for 3 min; followed by 34 cycles of 20 s at 95°C, 20 s at 56°C, 30 s at 72°C, and a final extension for 5 min at 72°C (Zhang H *et al.* 2022; Yang *et al.* 2023). The PCR products were recovered using a HiPure Gel Pure Mini Kit (Shanghai Yong Chuan Biological Technology Co., Ltd., China). Libraries were prepared using the ALFA-SEQ DNA Library Prep Kit (Findrop Biosafety Technology (Guangzhou) Co., Ltd., Guangzhou, China). Sequencing was performed using an Illumina Nova 6000 platform and MAGIGENE (Guangdong Magigene Biotechnology Co., Ltd., Guangzhou, China) (Zhu *et al.* 2023).

2.4. Bioinformatics analyses

The raw sequences obtained from the Illumina MiSeq platform were purified and analyzed using QIIME2, and high-quality sequences (q>20) were obtained (Caporaso *et al.* 2010; Cregger *et al.* 2018). The sequences were clustered into 100% amplicon sequence variants (ASVs). The raw sequence data were demultiplexed and quality-filtered using the q2-demux plug-in and denoised using DADA2 (*via* q2-dada2) (Callahan *et al.* 2016). ASVs were aligned using MAFFT (*via* q2-alignment) (Katoh *et al.* 2002), and

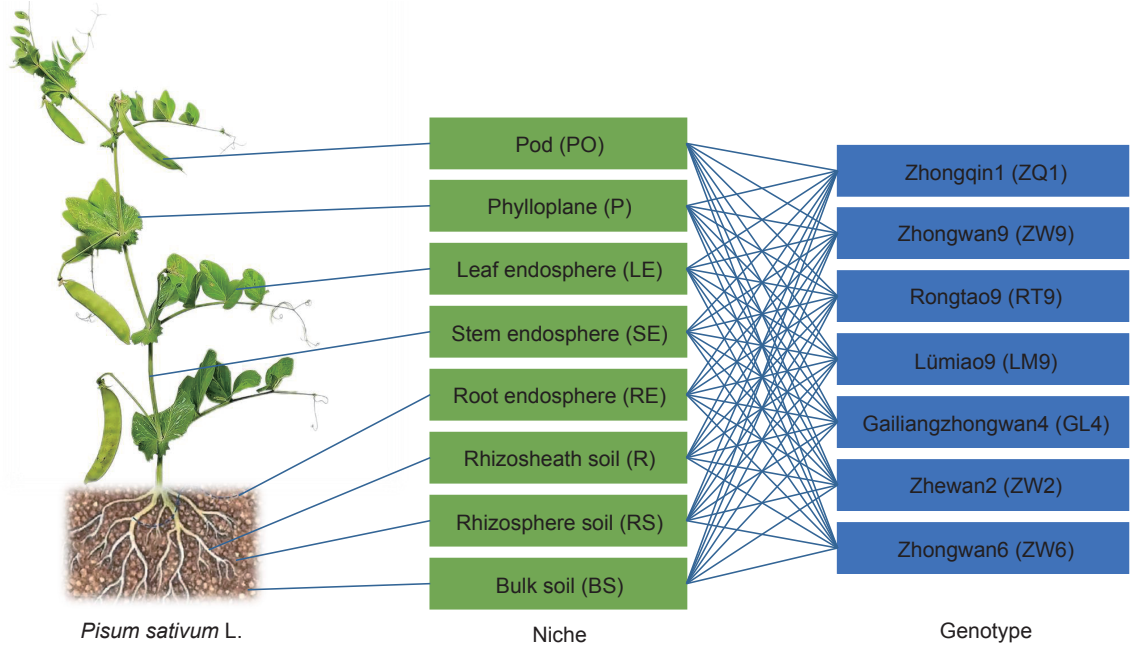


Fig. 1 Experimental design.

phylogenetic trees were constructed using fasttree2 (*via* q2-phylogeny) (Piñeiro *et al.* 2020). ASVs were annotated in QIIME2 using BLAST in the UNITE fungus database (Zhang *et al.* 2021; Yang *et al.* 2023). The obtained nucleotide sequences were submitted to the National Center for Biotechnology Information (NCBI, Bethesda, MD, USA) Sequence Reading Archive (accession number PRJNA1214735). To ensure an even sampling depth, the fungal ASV tables were rarefied to 19,088 sequences prior to microbial diversity analysis (Appendix B). After removing chimeras and low-quality sequences, the ITS sequences of 165 samples were analyzed. Note that three of the original 168 samples were discarded due to insufficient sequencing depth, including two samples of the rhizosheath soil and one sample within the leaf endosphere. The sequences were classified into 4,329 fungal ASVs and clustered using a 100% consistency threshold.

2.5. Statistical analyses

The α-diversity of fungi was evaluated using a one-way analysis of variance (Shannon index and Chao1 index) to reflect the species richness and evenness of microbial fungal communities in different niches and genotypes of peas (Jiya *et al.* 2023; Zhu *et al.* 2023). The differences in fungal community composition between ecotope environments were investigated by calculating the Bray-Curtis dissimilarity matrix for β-diversity and visualized using a non-metric multidimensional scale (Cregger *et al.* 2018). Species replacement and richness differences in the community composition were compared based on the decomposition of β-diversity (Shen *et al.* 2020). Deterministic and stochastic processes in the microbial community assembly were evaluated using neutral community model (NCM) analysis (Sloan *et al.* 2006; Xiong *et al.* 2021). The percentage contributions of the different sources to the overall microbial community composition in each sink were evaluated using the Functional Enrichment Analysis Software Tool (FEAST) (Shenhav *et al.* 2019; Li *et al.* 2023). In FEAST, the Expectation-Maximization algorithm combined with EM_iterations=1,000 was used to ensure model convergence. To address sequencing depth variation, samples were normalized using the rarefy function with COVERAGE set to the minimum sequencing depth, which ensures standardization while minimizing data loss and preserving integrity. In the microbial traceability analysis, “Source” and “Sink” are used to describe the migration and spreading processes of microbial communities. “Source” denotes the potential origins of microbial communities and is used to infer microbial sources in target samples (“Sink”); however, “Sink” represents the target ecosystem or sample whose microbial composition reflects the combined source contributions. Source–sink microbial relationship analysis was used to quantify source contributions, thereby revealing microbial migration patterns and ecological significance. All statistical analyses were conducted in R (http://www.r-project.org) using the adespatial, phyloseq, ggplot2, vegan, and feast R packages.

3. Results

3.1. Host niches and genotypes drive diversity and composition variation in pea

Among the eight niches, fungal α-diversity, as indicated by the Chao1 and Shannon indices, was significantly higher in P and R compared to the other groups, with RE and SE exhibiting the lowest diversity (Fig. 2-A and B). No significant differences were observed in the seven pea genotypes (Fig. 2-A, B, D and E; Appendix C). Clear and independent clustering was observed between the soil samples (BS, RS, and R) and the stems, leaves, and roots (Fig. 2-C), whereas clustering among plant genotypes was less pronounced (Fig. 2-F). The α-diversity analyses (ADONIS analysis) indicated that fungal community variation was mainly explained by pea niches (R^2=72.5%, P<0.001) rather than plant genotypes (R^2=2.3%, P=0.881).

At the phylum level, Ascomycota was the most abundant taxon among the identified fungi in the RE, LE, SE, and PO niches. In the BS, RS, R, and P niches, the relative abundance of Ascomycota was the highest, followed by Mortierellomycota and Basidiomycota (Fig. 2-G). At the genus level, the dominant genera in BS, RS, and R were *Cephaliophora*, *Mortierella*, *Ascobolus*, *Rhizopus*, and *Fusarium*; in RE, the dominant genus was *Rhizopus*; in P, the dominant genera were *Erysiphe*, *Alternaria*, and *Sporobolomyces*; and in LE, the dominant genus was *Erysiphe*. However, many endophytic microorganisms in the plant tissues were unidentified (Fig. 2-H). The dominant fungal phyla within pea genotypes were Ascomycota, Mortierellomycota, and Basidiomycota (Fig. 2-I), and the dominant fungal genera included *Ascobolus*, *Cephaliophora*, *Erysiphe*, and *Fusarium* (Fig. 2-J).

3.2. Dominant factors driving β-diversity partitioning

The differences in fungal β-diversity across the pea niches were primarily driven by species replacement (62.1, 75.2, 72.5, 65.2, 66.8, 73.4, 58.6, and 67.3% of the total dissimilarity in BS, RS, R, P, RE, LE, SE, and PO, respectively), whereas the contributions of richness differences were only 24.7–41.4% (Fig. 3-A; Appendix D). In addition, the fungal β-diversity across pea genotypes was also mainly driven by species replacement (62.2, 62.2, 61.6, 62.8, 55.4, 52.0, and 63.2% of the total dissimilarity in ZQ1, ZW9, RT9, LM9, GL4, ZW2, and ZW6, respectively), with a relatively lower contributions from richness differences (36.8–48.0%) (Fig. 3-B).

3.3. Fungal microbiome community assembly

The neutral community models revealed high explanatory power (64.3–77.0%) for the genotypes within the same niche, and the fungal migration rates between different niches ranged from 0.01006 to 0.1321 (Fig. 4-A). In contrast, the explanatory power for the niches within the same genotype was relatively lower, ranging from –24.0% to –2.1%, and the fungal migration rates between different niches ranged from 0.0009162 to 0.005331 (Fig. 4-B). These results indicated that the variation between different genotypes was driven by stochastic processes, whereas the variation in fungal community structure between different niches was driven by deterministic processes.

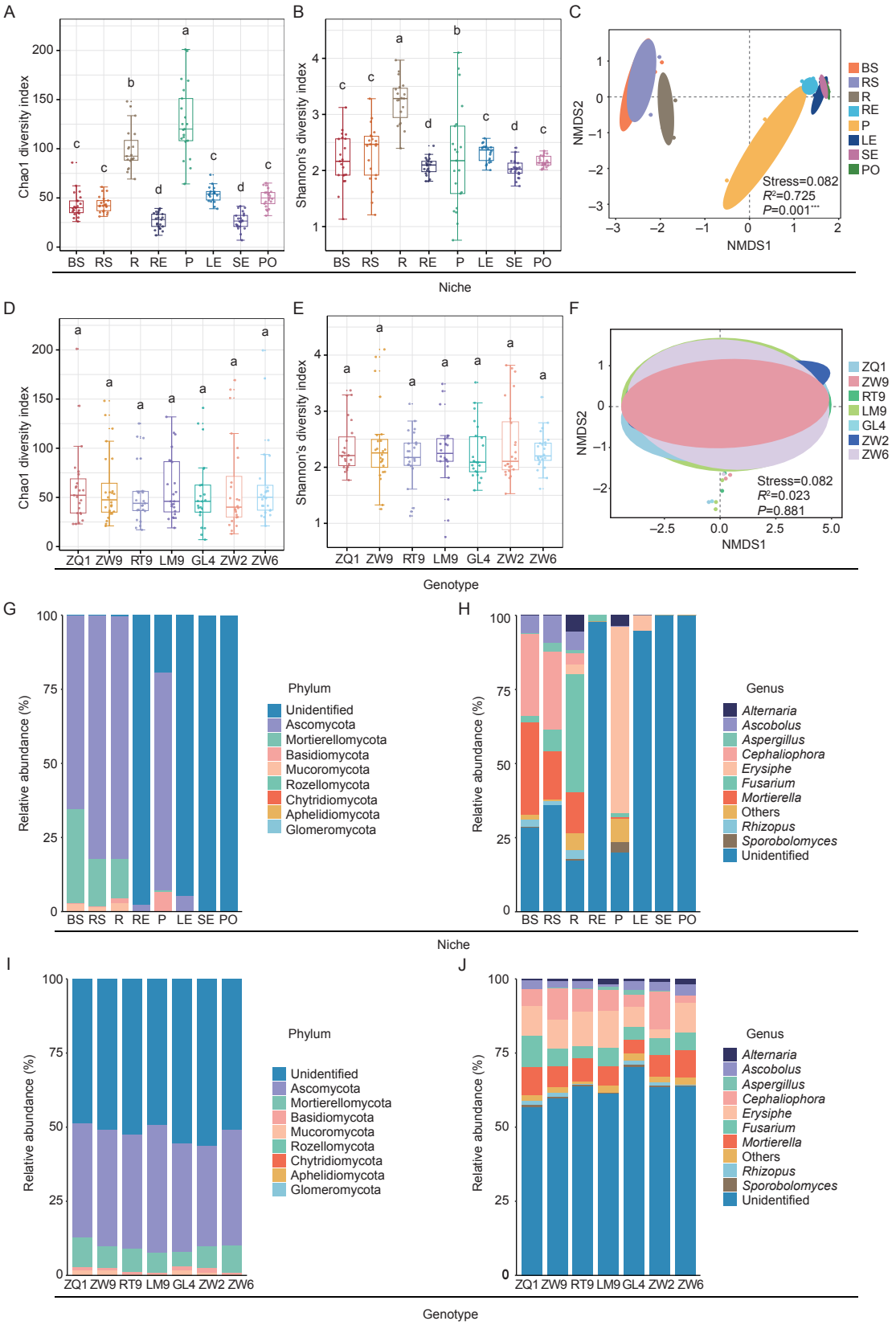


Fig. 2 Diversity and composition of the pea microbiomes across eight niches and seven genotypes. A, B, D and E, comparison of microbial diversity using the Shannon index and Chao1 index (n=21 per niche; n=24 per genotype). C and F, analyses of differences in fungal community composition based on Bray-Curtis and ADONIS analyses. G–J, relative abundances of dominant fungal phyla and genera across different niches and genotypes. BS, bulk soil; RS, rhizosphere soil; R, rhizosheath soil; RE, root endosphere; P, phylloplane; LE, leaf endosphere; SE, stem endosphere; PO, pod; ZQ1, Zhongqin1; ZW9, Zhongwan9; RT9, Rongtao9; LM9, Lumiao9; GL4, Gailiangzhongwan4; ZW2, Zhewan2; ZW6, Zhongwan6. Different lowercase letters indicate significant differences in pea microbiomes across niches and genotypes, as determined by Tukey's test, with error bars representing the standard deviation. ***, P<0.001.

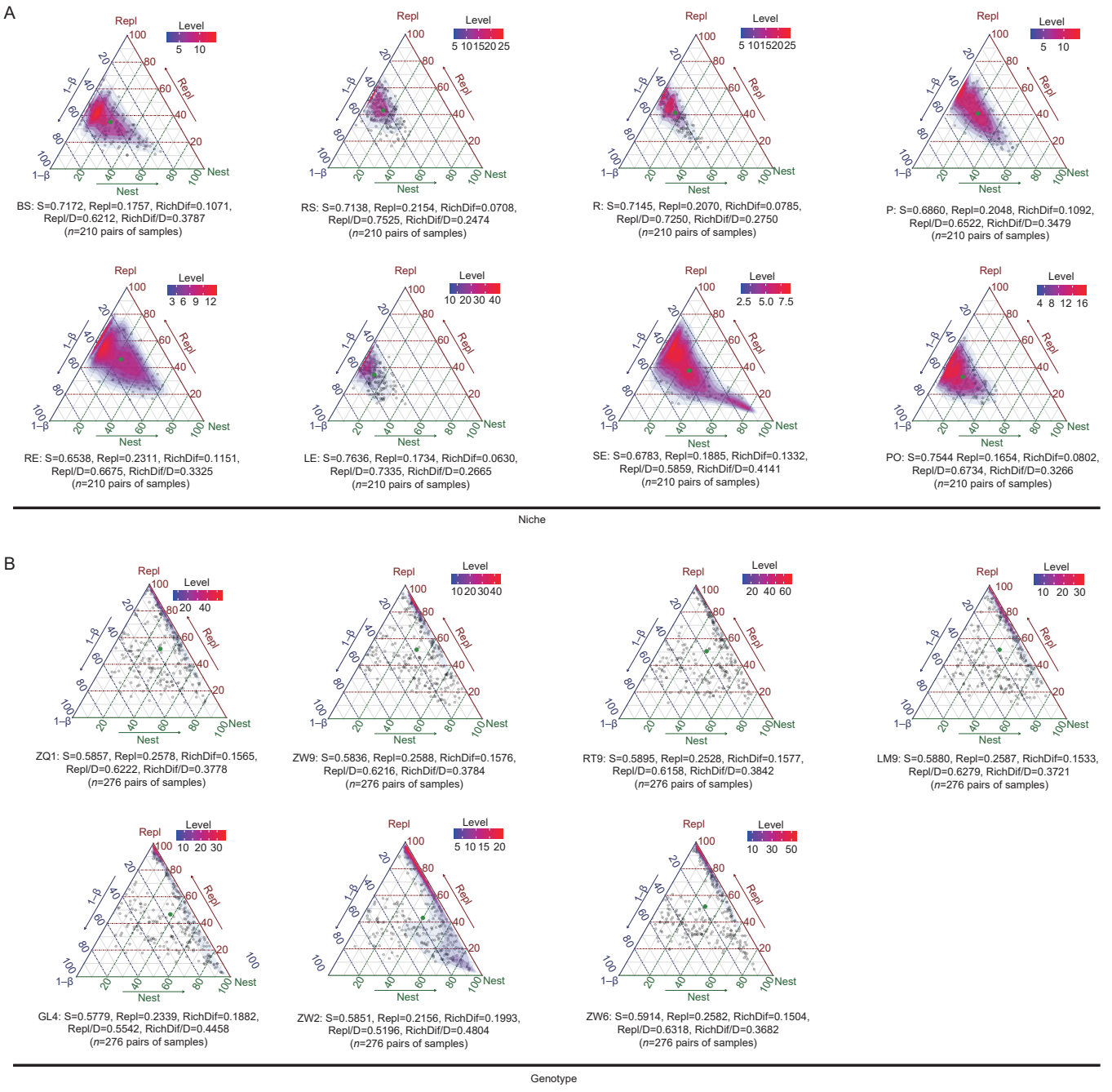


Fig. 3 β-Diversity decomposition of the impacts of species replacement and richness differences on the fungal microbiome composition in peas. A and B, ternary plots illustrating the effects of species replacement and richness differences on microbial β-diversity in pea niches (A) and pea genotypes (B). Each point represents a pair of samples, and its position is determined by the values of the species replacement and richness difference matrices. S, similarity; D, dissimilarity; Repl, species replacement; RichDiff, richness difference; BS, bulk soil; RS, rhizosphere soil; R, rhizosheath soil; RE, root endosphere; P, phylloplane; LE, leaf endosphere; SE, stem endosphere; PO, pod; ZQ1, Zhongqin1; ZW9, Zhongwan9; RT9, Rongtao9; LM9, Lumiao9; GL4, Gailiangzhongwan4; ZW2, Zhewan2; ZW6, Zhongwan6.

3.4. Potential sources of the pea plant niche fungal microbiome

The fungal communities associated with peas primarily originated from the soil, and they were gradually accumulated and filtered through different niches (Fig. 5). Overall, 83.7 and 3.0% of the microorganisms in RS and P niches originated from BS, respectively, whereas 64.4% of those in the R niche originated from the RS. In addition, 1.26% of the microorganisms in the RE niche originated from the R, while 96.4 and 92.8% of microorganisms in the SE and PO niches, respectively, originated from the RE. Moreover, 89.8 and 93.2% of the microorganisms in the LE and PO niches, respectively, originated from the SE, while 61.5 and 94.3% of the P and PO niches, respectively, originated from the LE. These results reflected an exogenous-to-endogenous migration process for the soil–pea plant continuum.

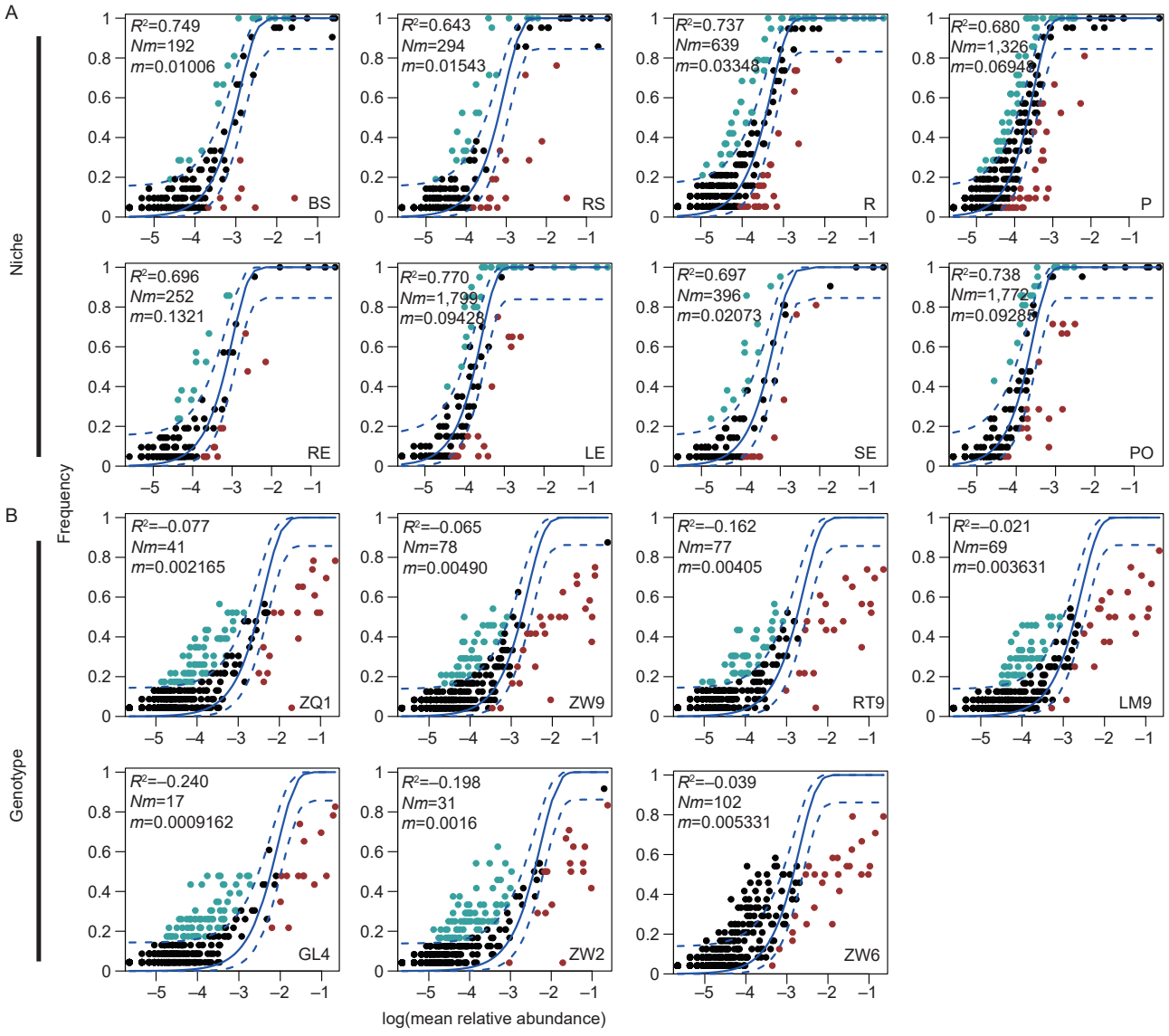


Fig. 4 Stochastic and non-stochastic (deterministic) processes of fungal microbiome community assembly in the pea plant niche and genotype using neutral community models (NCM). R^2 represents the overall goodness of fit of the NCM, *Nm* denotes the meta-community size, and *m* quantifies the migration rate at the community level. In each graph, the solid blue line indicates the best fit of the NCM, while the dashed blue line represents the 95% confidence interval of the model. Amplicon sequence variants with frequencies higher or lower than those predicted by the NCM are shown in different colors. BS, bulk soil; RS, rhizosphere soil; R, rhizosheath soil; RE, root endosphere; P, phylloplane; LE, leaf endosphere; SE, stem endosphere; PO, pod; ZQ1, Zhongqin1; ZW9, Zhongwan9; RT9, Rongtao9; LM9, Lumiao9; GL4, Gailiangzhongwan4; ZW2, Zhewan2; ZW6, Zhongwan6.

3.5. Migration of potential fungal genera from exogenous-to-endogenous pea niches

We observed 152 unique ASVs in endogenous niches (RE, SE, LE, PO), 473 unique ASVs in exogenous niches (BS, RS, R, P), and 95 ASVs shared between endogenous and exogenous niches (Fig. 6-A). Among them, *Chrysosporium* and *Humicola* were specific to endogenous niches; *Rhizopus*, *Mortierella*, *Sporobolomyces*, and *Papiliotrema* were specific to exogenous niches; and *Erysiphe*, *Fusarium*, *Cephaliophora*, *Ascobolus*, and *Alternaria* were shared in both endogenous and exogenous niches (Fig. 6-B). In the endogenous niches, 47 (19.0%) ASVs overlapped in RE, SE, LE, and PO (Fig. 6-C); however, all shared ASVs were unidentified (Fig. 6-D). In exogenous niches, 55 (9.7%) ASVs overlapped in BS, RS, R, and P (Fig. 6-E), and *Erysiphe*, *Mortierella*, *Cephaliophora*, and *Fusarium* were common in the BS, RS, R, and P habitats (Fig. 6-F).

4. Discussion

4.1. Effects of pea niches and genotypes on fungal community structure

Significant differences in pea-associated fungal communities were observed across niche compartments, whereas the differences among genotypes were less pronounced. This result aligned with previous studies on *Populus* (Cregger *et al.* 2018) and *Capsicum annuum* L. (Gao *et al.* 2021), confirming that niche differentiation, rather than pea genotype, was the primary diver of microbial community assembly (Fig. 7). This

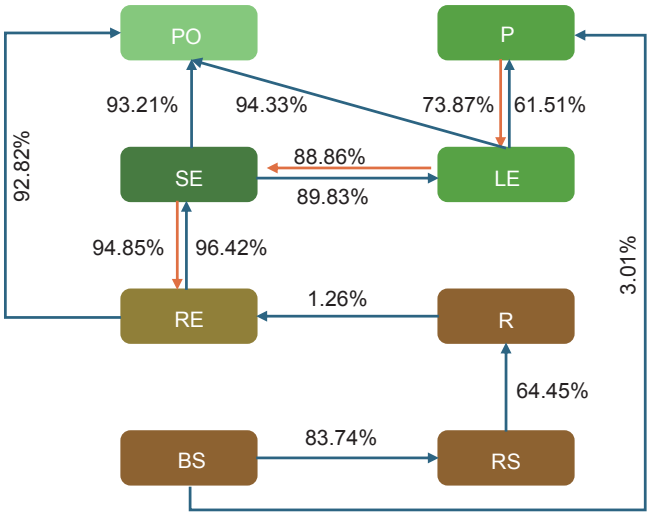


Fig. 5 Source tracing analysis model of the niche fungal microbiome of the pea plant. BS, bulk soil; RS, rhizosphere soil; R, rhizosheath soil; RE, root endosphere; P, phylloplane; LE, leaf endosphere; SE, stem endosphere; PO, pod. The direction of each arrow indicates the source-to-sink relationships. The blue arrows represent source-to-sink transfer from the soil to plant systems, and the orange arrows represent the reverse transfer from the plant system. The percentages represent the contribution of each source.

may be because the surface environments of pea plants in the BS, RS, R, and P niches provided richer resources and more favorable niches, whereas endogenous organ-associated microenvironments, such as RE, SE, and LE, are relatively enclosed and face more biological defense mechanisms, which could limit the survival and reproduction of microorganisms (Remus-Emsermann and Schlechter 2018; Qian *et al.* 2019).

We further explored the compositions of fungal communities and found that the fungal microbiomes of the pea niches and genotypes were primarily composed of Ascomycota, Mortierellomycota, and Basidiomycota. Ascomycota encompass plant symbiotic fungi that can promote growth *via* hormone production or enhanced nutrient uptake, with some species exhibiting antagonism through antibiotics or resource competition (Silva *et al.* 2019; Manici *et al.* 2024). Mortierellomycota is prevalent in soils and drives organic matter decomposition, nutrient cycling, and plant growth through lipid metabolites (Ozimek and Hanaka 2021). Basidiomycota form mycorrhizal symbioses, aiding water and nutrient uptake, while some species can suppress pathogens *via* organic matter decomposition or antimicrobial production (Manici *et al.* 2024). These results confirmed the critical roles of the relevant fungi in plant growth, pathogenicity, and antagonism, which profoundly influence the functionality and stability of ecosystems.

4.2. Fungal β-diversity decomposition and the community assembly processes across pea niches and genotypes

The impact of species replacement on fungal microbial composition was significantly higher than that of richness differences, and species replacement served as the primary cause of the differences in fungal communities across pea niches and genotypes. Species replacement depends on the dispersal potential of microbial communities and external environmental disturbances, with the microbial dispersal potential influenced by individual dispersal ability and environmental filtering (Guo *et al.* 2023; Wan *et al.* 2023). To further evaluate the relative contributions of deterministic and stochastic processes on species replacement, we used a neutral model to analyze the community assembly mechanisms in pea niches and genotypes. The results showed that the NCM fitting and *Nm* values for fungal communities were higher in pea genotypes than in niches, indicating a greater role of stochastic processes in genotype community assembly, while deterministic processes dominated in niches. These results suggested that species replacement dominated the differences in fungal communities among pea genotypes while neutral tests supported stochastic processes, which was consistent with the findings of previous studies on microbes in soil and sediments along the Yangtze River (Wan *et al.* 2023). This may be related to the degree of nutrient availability and cell size in the environment (Chase 2010; Goldford *et al.* 2018). Moreover, fungal cells are less easily dispersed in a nutrient-rich state (Zinger *et al.* 2019).

4.3. Migration pathways of fungal communities across pea niches

The belowground exogenous niches (BS, RS, and R) contributed significantly to fungal communities in the aboveground niches (RE, SE, LE, and PO). In addition, aboveground niches such as RE, SE, LE and P exhibited a certain degree of fungal biodiversity exchange. For example, the SE niche acted as a sink for RE, while serving as a source for LE and PO. This indicated that each niche was a potentially significant source of fungi, thereby maintaining biodiversity. Previous studies suggested that when environmental conditions fluctuate, sinks can become sources for sustaining the diversity of fungal communities (Zhang L *et al.* 2022; Li *et al.* 2023). These results confirmed the importance of maintaining the diversity of fungal communities in both soil and plant niches. Previous studies found that specific groups from one habitat can migrate to adjacent niches (Xiong *et al.* 2020; Zwirzitz *et al.* 2020). Consistent with these findings, this study also revealed similar patterns in which the main fungal genera migrating across pea plant niches included *Erysiphe*, *Fusarium*, *Cephaliophora*, *Ascobolus*, *Alternaria*, and *Aspergillus*. *Fusarium* and *Erysiphe* are plant pathogenic fungi that are widely distributed in soil and plants, common in agricultural production systems, and easily isolated and identified. They can induce plant disease outbreaks under suitable environmental conditions (Lahlali *et al.* 2024). However, some soil-borne fungal pathogen species enhance seedling drought resistance (Pappas *et al.* 2018; Zhou *et al.* 2021). These results suggested that endogenous fungi may exert either pathogenic or beneficial effects on the host plant, although further investigation is required to confirm these effects. Moreover, given the significant role of ecological conditions at the site, preventive measures should be implemented in follow-up studies to mitigate the risk of plant diseases.

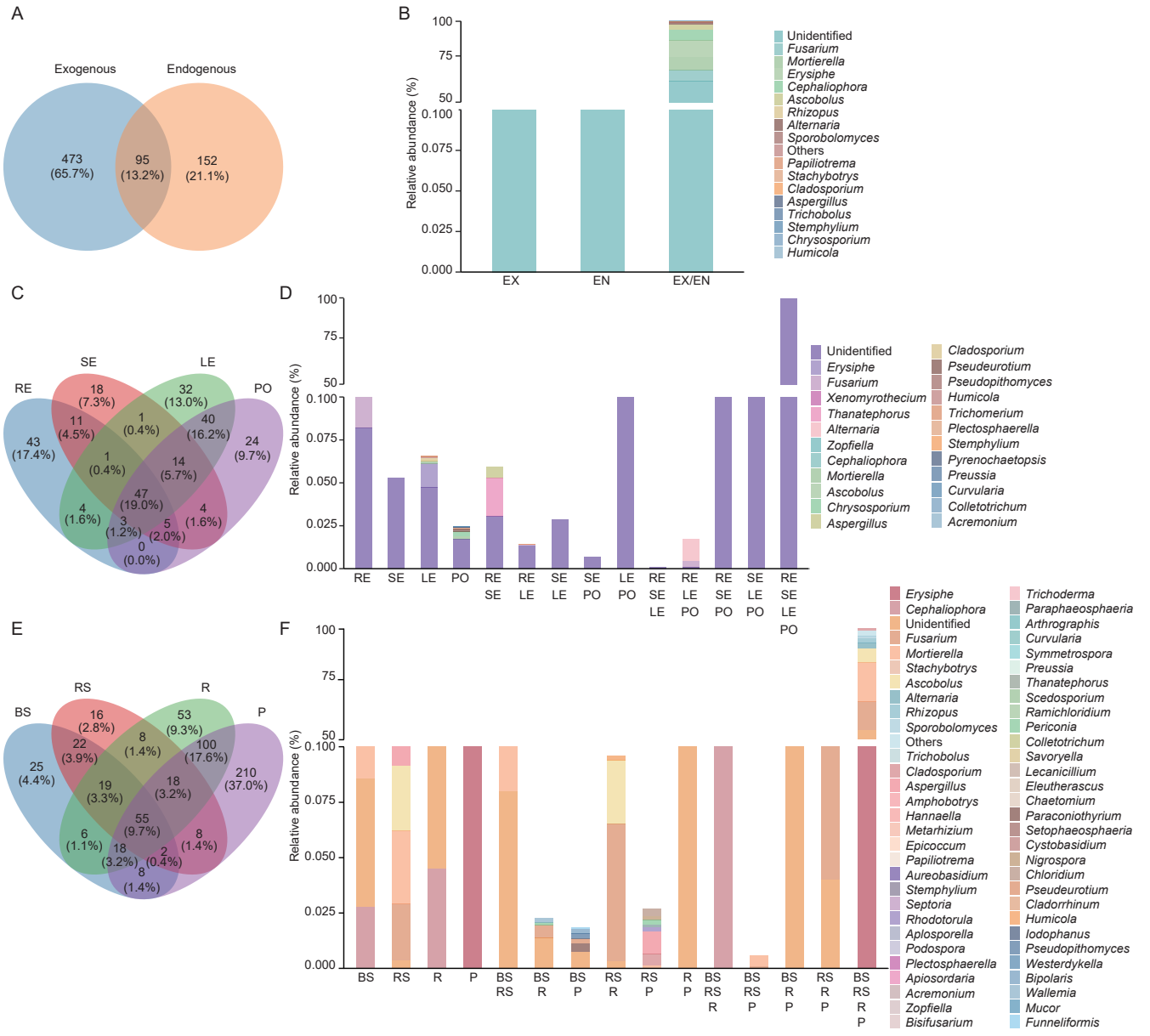


Fig. 6 Shared and unique fungal amplicon sequence variants (ASVs) and genera taxonomy in the endogenous and exogenous niches of pea plants at the genus level. A–C, Venn diagrams illustrating the numbers of shared and unique ASVs in the endogenous (RE, SE, LE, and PO) and exogenous (BS, RS, R, and P) habitats. D–F, stacked bar charts of the shared and unique genera in endogenous and exogenous habitats. EX, exogenous; EN, endogenous; BS, bulk soil; RS, rhizosphere soil; R, rhizosheath soil; RE, root endosphere; P, phylloplane; LE, leaf endosphere; SE, stem endosphere; PO, pod.

5. Conclusion

This study systematically investigated the ecological assembly mechanisms of fungal communities across different pea niches and genotypes. The key findings were as follows: (1) Fungal community composition was primarily shaped by plant-associated niches rather than host genotypes. Significant niche differentiation was observed, with clear separation between the belowground (soil and root) and aboveground (leaf, stem, and pod) communities. (2) Community assembly was more significantly affected by species replacement than by richness differences in both pea niches and genotypes. (3) Niche-based fungal assembly was predominantly governed by deterministic processes, including environmental filtering and niche partitioning, whereas genotype-driven assembly was more influenced by stochastic processes. This implies that optimizing soil conditions (e.g., nutrient availability, moisture, and pH) can selectively enrich beneficial fungi, thereby enhancing plant growth and nutrient acquisition. (4) The potential sources and cross-habitat migration patterns of the pea fungal community were identified through source traceability analysis. The results revealed that each niche could act as a source of fungal taxa. Notably, the main fungal genera migrating across pea plant niches included *Erysiphe*, *Fusarium*, *Cephaliophora*, *Ascobolus*, *Alternaria*, and *Aspergillus*. These results increase our understanding of fungal microbiome diversity and species composition and community assembly mechanisms in the soil–pea continuum. Moreover, the results

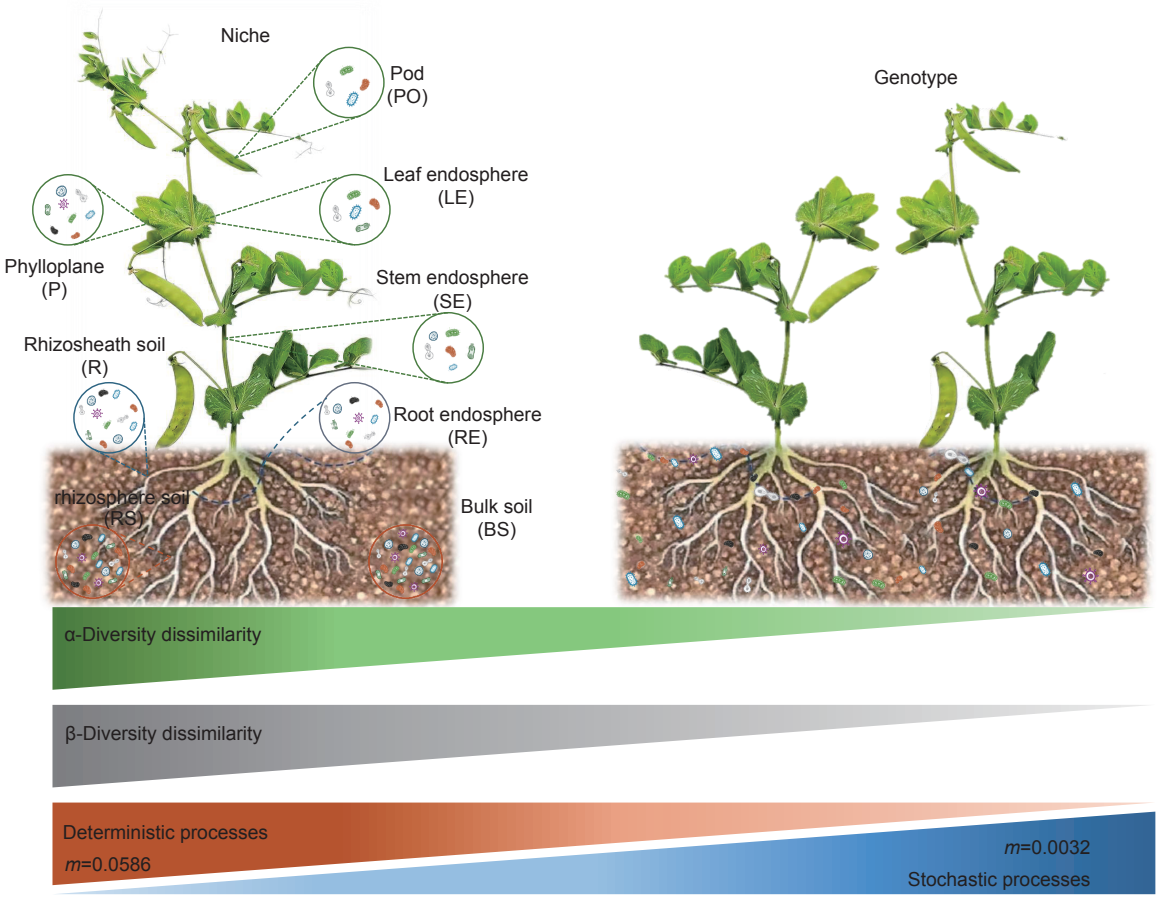


Fig. 7 Conceptual diagram of how the fungal community assembly mechanisms are influenced by host niche and genotype. *m* quantifies the migration rate at the community level.

provide a new theoretical basis for the sustainable production of peas, and guiding significance for the sustainable development of global agriculture.

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Declaration of competing interest

The authors declare that they have no conflict of interest.

Declaration of generative AI and AI-assisted technologies in the writing process

The authors declare that they did not use AI in the preparation and writing of this manuscript.

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References

Beckers B, Beeck M O D, Weyens N, Boerjan W, Vangronsveld J. 2017. Structural variability and niche differentiation in the rhizosphere and endosphere bacterial microbiome of field-grown poplar trees. *Microbiome*, **5**, 25.

Boakye T A, Li H, Osei R, Boamah S, Min Z, Ni C, Wu J, Shi M, Qiao W. 2022. Antagonistic effect of *Trichoderma longibrachiatum* (TL6 and TL13) on *Fusarium solani* and *Fusarium avenaceum* causing root rot on snow pea plants. *Journal of Fungi*, **8**, 1148.

Calderon R B, Dangi S R. 2024. Arbuscular mycorrhizal fungi and rhizobium improve nutrient uptake and microbial diversity relative to dryland site-specific soil conditions. *Microorganisms*, **12**, 667.

Callahan B J, McMurdie P J, Rosen M J, Han A W, Johnson A J A, Holmes S P. 2016. DADA2: High-resolution sample inference from Illumina amplicon data. *Nature Methods*, **13**, 581–583.

Caporaso J G, Kuczynski J, Stombaugh J, Bittinger K, Bushman F D, Costello E K, Fierer N, Peña A G, Goodrich J K, Gordon J I, Huttley G A, Kelley S T, Knights D K, Koenig J E, Ley R E, Lozupone C A, Mcdonald D, Muegge B D, Pirrung M, Reeder J, *et al.* 2010. QIIME allows analysis of high-throughput community sequencing data. *Nature Methods*, **7**, 335–336.

Chase J M. 2010. Stochastic community assembly causes higher biodiversity in more productive environments. *Science*, **328**, 1388–1391.

Chen X, Krug L, Yang H, Li H, Yang M, Berg G, Cernava T. 2020. Nicotiana tabacum seed endophytic communities share a common core structure and genotype-specific signatures in diverging cultivars. *Computational and Structural Biotechnology Journal*, **18**, 287–295.

Cheng S, Feng C, Chen B, Hofer J, Shi Y, Jiang M, Song B, Cheng H, Lu L, Wang L, Howard A, Bendahmane A, Fouchal A, Moreau C, Sawada C, Lesignor C, Barclay E, Vikeli E, Tsanakas G, Zhao H, *et al.* 2024. Genomic and genetic insights into Mendel's pea genes. *bioRxiv*, **5**, 596837.

Cregger M A, Veach A M, Yang Z K, Crouch M J, Vilgalys R, Tuskan G A, Schadt C W. 2018. The *Populus* holobiont: Dissecting the effects of plant niches and genotype on the microbiome. *Microbiome*, **6**, 31.
Gao M, Xiong C, Gao C, Tsui C K M, Wang M, Zhou X, Zhang A M, Cai L. 2021. Disease-induced changes in plant microbiome assembly and functional adaptation. *Microbiome*, **9**, 187.
Goldford J E, Lu N, Bajić D, Estrela S, Tikhonov M, Sanchez-Gorostiaga A S, Segrè D, Mehta P, Sanchez A. 2018. Emergent simplicity in microbial community assembly. *Science*, **361**, 469–474.
Guo B, Zhang H, Liu Y, Chen J, Li J. 2024. Assembly of cereal crop fungal communities under water stress determined by host niche. *Environmental and Experimental Botany*, **219**, 105650.
Guo Q, Wen Z, Ghanizadeh H, Fan Y, Zheng C, Yang X, Yan X, Li W. 2023. Stochastic processes dominate assembly of soil fungal community in grazing excluded grasslands in northwestern China. *Journal of Soils and Sediments*, **23**, 156–171.
Hao J, Liu Q, Song F, Cui X, Liu L, Fu L, Zhang S, Wu X, Zhang X. 2024. Community diversity of endophytic bacteria in the leaves and roots of pea seedlings. *Agronomy*, **14**, 2030.
Jiya N, Shede P, Sharma A. 2023. Diversity and composition of fungal communities across diverse environmental niches in Antarctica. *Polar Science*, **38**, 100973.
Katoh K, Misawa K, Kuma K, Miyata T. 2002. MAFFT: A novel method for rapid multiple sequence alignment based on fast Fourier transform. *Nucleic Acids Research*, **30**, 3059–3066.
Lahlali R, Taoussi M, Laasli S E, Gachara G, Ezzouggari R, Belabess Z, Aberkani K, Assouguem A, Meddich A, Jarroudi M E, Barka E A. 2024. Effects of climate change on plant pathogens and host-pathogen interactions. *Crop and Environment*, **3**, 159–170.
Li S, Ren K, Yan X, Tsyganov A N, Mazei Y, Smirnov A, Mazei N, Zhang Y, Rensing C, Yang J. 2023. Linking biodiversity and ecological function through extensive microeukaryotic movement across different habitats in six urban parks. *iMeta*, **2**, e103.
Lv X, Wang Q, Zhang X, Hao J, Li L, Chen W, Li H, Wang Y, Ma C, Wang J, Liu Q. 2021. Community structure and associated networks of endophytic bacteria in pea roots throughout plant life cycle. *Plant and Soil*, **468**, 225–238.
Manici L M, Caputo F, De Sabata D D, Fornasier F. 2024. The enzyme patterns of ascomycota and basidiomycota fungi reveal their different functions in soil. *Applied Soil Ecology*, **196**, 105323.
Nguyen N H, Song Z, Bates S T, Branco S, Tedersoo L, Menke J, Schilling J S, Kennedy P G. 2016. FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecology*, **20**, 241–248.
Ozimek E, Hanaka A. 2021. *Mortierella* species as the plant growth-promoting fungi present in the agricultural soils. *Agriculture*, **11**, 7.
Pappas M L, Liapoura M, Papantoniou D, Avramidou M, Kavroulakis N, Weinhold A, Broufas G D, Papadopoulou K K, 2018. The beneficial endophytic fungus *Fusarium solani* strain K alters tomato responses against spider mites to the benefit of the plant. *Frontiers in Plant Science*, **9**, 1603.
Pii Y, Mimmo T, Tomasi N, Terzano R, Cesco S, Crecchio C. 2015. Microbial interactions in the rhizosphere: Beneficial influences of plant growth-promoting rhizobacteria on nutrient acquisition process. A review. *Biology and Fertility of Soils*, **51**, 403–415.
Piñeiro C, Abuín J M, Pichel J C. 2020. Very Fast Tree: Speeding up the estimation of phylogenies for large alignments through parallelization and vectorization strategies. *Bioinformatics*, **36**, 4658–4659.
Qian X, Li H, Wang Y, Wu B, Wu M, Chen L, Li X, Zhang Y, Wang X, Shi M, Zheng Y, Guo L, Zhang D. 2019. Leaf and root endospheres harbor lower fungal diversity and less complex fungal co-occurrence patterns than rhizosphere. *Frontiers in Microbiology*, **10**, 1015.
Remus-Emsermann M N P, Schlechter R O. 2018. Phyllosphere microbiology: At the interface between microbial individuals and the plant host. *New Phytologist*, **218**, 1327–1333.
Schmidt R, Mitchell J, Scow K. 2019. Cover cropping and no-till increase diversity and symbiotroph: Saprotroph ratios of soil fungal communities. *Soil Biology and Biochemistry*, **129**, 99–109.
Shen C, Gunina A, Luo Y, Wang J, He J, Kuzyakov Y, Hemp A, Classen A T, Ge Y. 2020. Contrasting patterns and drivers of soil bacterial and fungal diversity across a mountain gradient. *Environmental Microbiology*, **22**, 3287–3301.
Shenhav L, Thompson M, Joseph T A, Briscoe L, Furman O, David B, Mizrahi I, Pe'er I, Halperin E. 2019. FEAST: Fast expectation-maximization for microbial source tracking. *Nature Methods*, **16**, 627–632.
Silva R N, Monteiro V N, Steindorff A S, Gomes E V, Noronha E F, Ulhoa C J. 2019. Trichoderma/pathogen/plant interaction in pre-harvest food security. *Fungal Biology*, **123**, 565–583.
Sloan W T, Lunn M, Woodcock S, Head I M, Nee S, Curtis T P. 2006. Quantifying the roles of immigration and chance in shaping prokaryote community structure. *Environmental Microbiology*, **8**, 732–740.
Wan W, Gadd G M, Gu J, Liu W, Chen P, Zhang Q, Yang Y. 2023. Beyond biogeographic patterns: Processes shaping the microbial landscape in soils and sediments along the Yangtze River. *mLife*, **2**, 89–100.
Xiong C, He J, Singh B K, Zhu Y, Wang J, Li P, Zhang Q, Han L, Shen J, Ge A, Wu C, Zhang L. 2020. Rare taxa maintain the stability of crop mycobiomes and ecosystem functions. *Environmental Microbiology*, **23**, 1907–1924.
Xiong C, Zhu Y, Wang J, Singh B, Han L, Shen J, Li P, Wang G, Wu C, Ge A, Zhang L, He J. 2021. Host selection shapes crop microbiome assembly and network complexity. *New Phytologist*, **229**, 1091–1104.
Yang H, Ye W, Yu Z, Shen W, Li S, Wang X, Chen J, Wang Y, Zheng X. 2023. Host niche, genotype, and field location shape the diversity and composition of the soybean microbiome. *Journal of Integrative Agriculture*, **22**, 2412–2425.
Yang T, Liu R, Luo Y, Hu S, Wang D, Wang C, Pandey M K, Ge S, Xu Q, Li N, Li G, Huang Y, Saxena R K, Ji Y, Li M, Yan X, He Y, Liu Y, Wang X, Xiong C, *et al.* 2022. Improved pea reference genome and pan-genome highlight genomic features and evolutionary characteristics. *Nature Genetics*, **54**, 1553–1563.
Zhang H, Wu C, Wang F, Wang H, Chen G, Cheng Y, Chen J, Yang J, Ge T. 2022. Wheat yellow mosaic enhances bacterial deterministic processes in a plant–soil system. *Science of the Total Environment*, **812**, 151430.
Zhang L, Zhang M, Huang S, Huang S, Li L, Gao Q, Wang Y, Zhang S, Huang S, Yuan L, Wen Y, Liu K, Yu X, Li D, Zhang L, Xu X, Wei H, He P, Zhou W, Philippot L, Ai C. 2022. A highly conserved core bacterial microbiota with nitrogen-fixation capacity inhabits the xylem sap in maize plants. *Nature Communications*, **13**, 3361.
Zhang Z, Zhang J, Jiao S. 2021. Fungi show broader environmental thresholds in wet than dry agricultural soils with distinct biogeographic patterns. *Science of the Total Environment*, **750**, 141761.
Zhao M, Loreau M, Ochoa-Hueso R, Zhang H, Yang J, Zhang Y, Liu H, Jiang Y, Han X. 2024. Decoupled responses of above- and below-ground beta-diversity to nitrogen enrichment in a typical steppe. *Ecology Letters*, **27**, e14339.
Zhou X R, Dai L, Xu G F, Wang H S. 2021. A strain of *Phoma* species improves drought tolerance of *Pinus tabulaeformis*. *Scientific Reports*, **11**, 7637.
Zinger L, Taberlet P, Schimann H, Bonin A, Boyer F, Barba M D, Gaucher P, Gielly L, Giguet-Covex C, Iribar A, Réjou-Méchain M, Rayé G, Rioux D, Schilling V, Tymen B, Viers J, Zouiten C, Thuiller W, Coissac E, Chave J. 2019. Body size determines soil community assembly in a tropical forest. *Molecular Ecology*, **28**, 528–543.
Zhu T, Zhang L, Yan Z, Liu B, Li Y, You X, Chen Mo, Liu T, Xu Y, Zhang J. 2023. Niche-dependent microbial assembly in salt-tolerant tall fescue and its contribution to plant biomass. *Industrial Crops and Products*, **206**, 117736.
Zwirzitz B, Wetzels S U, Dixon E D, Stessl B, Zaiser A, Rabanser I, Thalguter S, Pinior B, Roch F, Strachan C, Zanghellini J, Dzieciol M, Wagner M, Selberherr E. 2020. The sources and transmission routes of microbial populations throughout a meat processing facility. *NPJ Biofilms and Microbiomes*, **6**, 26.

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