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Original Article

Bacterial microbiome and their assembly processing in two sympatric desert rodents (*Dipus sagitta* and *Meriones meridianus*) from different geographic sources

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Abstract

The microbiome of mammals has profound effects on host fitness, but the process, which drives the assembly and shift of mammalian microbiome remains poorly understood. To explore the patterns of small mammal microbial communities across host species and geographical sites and measure the relative contributions of different processes in driving assembly patterns, 2 sympatric desert rodent species (*Dipus sagitta* and *Meriones meridianus*) were sampled from 2 geographically distant regions, which differed in the environment, followed by 16S rRNA gene sequencing. The microbiomes differed significantly between *D. sagitta* and *M. meridianus*, and linear mixed modeling (LMM) analysis revealed that microbial diversity was mostly affected by species rather than the environment. For each rodent species, the microbiome diversity and structure differed across geographical regions, with individuals from lower rainfall environments exhibiting greater diversity. The null modeling results suggested dispersal limitation and ecological drift rather than differential selective pressures acting on the microbiome. In addition, each group had a different core genus, suggesting that the taxonomic composition of the microbiome was shaped most strongly by stochastic processes. Our results suggest that variation in the microbiome between hosts, both within and among geographic rodent populations, is driven by bacterial dispersal and ecological drift rather than by differential selective pressures. These results elucidated the diversity patterns and assembly processes of bacterial microbiomes in small desert mammals. Deciphering the processes shaping the assembly of the microbial community is a premise for better understanding how the environment-host-microbe interactions of mammals are established and maintained, particularly in the context of increased environmental disturbances and global changes.

Key words: desert rodents, mammal, microbial ecology, null models, wildlife.

The microbiome of mammals profoundly influences host fitness because they perform many important functions in hosts, including food digestion (Tremaroli and Backhed 2012), immunity regulation (Round and Mazmanian 2009), disease prevention (Tlaskalova-Hogenova et al. 2011) and physical development (Sommer and Backhed 2013). The structure of microbial communities is shaped by a complex set of host attributes (e.g., host genotype, species, ontogeny, and diet) and environmental factors (e.g., habitat, geographic location, and anthropogenic disturbance) (Ley et al. 2008; Spor et al. 2011; Amato et al. 2013), whose relative quantitative importance varies between hosts and geographical locations (Huang et al. 2022). Although the composition, diversity and function of the microbial communities of small mammals have been studied (Kevin et al. 2022; Zahra et al. 2022), the assembly of the microbiome of small mammals has not yet been investigated. Deciphering the processes shaping the assembly of microbial communities is an approach to better understand how the environment-host-microbe interaction of mammals is established and maintained, particularly in the context of increased environmental disturbances and global changes.

Selection, dispersal, and drift have been proposed to be the major processes, which govern the assembly and shift of ecological communities (Vellend 2010; Rosindell et al. 2011; Feng et al. 2018). The niche theory emphasizes the deterministic forces of biotic and abiotic factors in sorting communities (Vellend 2010; Dini-Andreote et al. 2015; Li et al. 2019). Selection may cause communities to converge if they undergo similar environmental conditions (homogeneous selection) or diverge if they undergo distinct environmental conditions (variable selection/Heterogeneous selection). The neutral theory emphasizes stochastic processes such as dispersal and drift (Rosindell et al. 2011; Dini-Andreote et al. 2015). Dispersal influences community assembly by regulating the movement of species across spaces and systems. Dispersal can also cause communities to converge or diverge depending

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on the magnitude of dispersal; that is, high dispersal homogenizes communities through frequent species exchange between communities (homogeneous dispersal), whereas restricted dispersal differentiates communities (dispersal limitation). Ecological drift results in stochastic population fluctuations of species within communities by chance birth and death events and thus generally disperse communities (Ge et al. 2021).

Although knowledge of microbial community assembly in several ecosystems (e.g., gut, soil, and water) has increased in recent years (Zhou et al. 2014; Yan et al. 2016; Dong et al., 2024), there are differences in the relative contributions of assembly processes between free-living and host-associated microbiomes. Generally, deterministic process and stochastic processes work in conjunction in microbiome assembly (Stegen et al. 2012; Dini-Andreote et al. 2015), but the relative contributions of these ecological processes (e.g., diversification, selection, dispersal, and drift) in shaping host microbiomes are likely to vary between ecosystems (Furman et al. 2020; Ge et al.,2021; Xiao et al. 2021). In this study, our main aim was to determine the relative importance of each process (i.e., homogeneous selection, heterogeneous selection, dispersal limitation, homogenizing dispersal, and drift) during the assembly of bacterial microbiomes and to determine how their relative importance varies across biological and geographical scales.

Hosts, which occupy a broad range of environments may exhibit greater spatiotemporal variation in their microbiome than those constrained as specialists to narrower subsets of resources or habitats. To explore the patterns of small mammal bacterial microbiomes across host species and geographical sites and measure the relative contributions of different processes (i.e., homogeneous selection, heterogeneous selection, dispersal limitation, homogenizing dispersal, and ecology drift) in driving assembly patterns, we surveyed the bacterial microbiomes of sympatric and allopatric populations of 2 rodent species residing in the wild throughout western China: *Dipus sagitta* and *Meriones meridianus*. These host species are ideal for comparison because they are relatively abundant and widespread in the inland deserts of East Asia (IUCN 2016), and their distribution range has been significantly influenced by climate change (Bu et al. 2022; Wang et al. 2022). They are both social animals, which inhabit similar environments and mostly survive in high-altitude deserts and semideserts. The diet of *D. sagitta* largely consists of nuts, seeds, and insects, whereas that of *M. meridianus* is dominated by stems and a leaf of herbaceous plants (Wilson et al. 2017).

We tested 3 hypotheses: first, the gut microbiota of sympatric species differs in the compositional and predicted functional characteristics of their gut microbiomes, reflecting that host phylogeny plays a crucial role in shaping the gut microbiota; second, host species exhibit different degrees of spatiotemporal turnover in the composition and diversity of their gut microbiomes. Regions with greater rainfall show greater host microbial diversity, reflecting ecological differences in how hosts respond to rainfall-driven environmental changes; third, deterministic processes drive the process of community assemblage of the gut microbiota in each group because selection is the result of biotic and abiotic pressures.

Materials and Methods

Sampling

To explore the patterns of small mammals microbial communities across host species and geographical sites and measure the relative contributions of different processes in driving assembly patterns, 4 groups (LD, LM, HD, and HM) of 2 species (D: *D.sagitta* and M: *M.meridianus*) within 2 types of environments on the basis of mean annual precipitation (MAP) (L: MAP < 50 mm, H: MAP > 100 mm) were considered here, with 6 and 5 geographical sites selected from each region (separated by distances of up to approximately 2,500 km). We collected a total of 39 fecal samples in the summer of 2021–2022 (Figure 1). The information for each sample is provided in Table S1. We used the same methods of sample collection and preservation at all of the sampling sites. Briefly, on capture, the animals were euthanized via cervical dislocation to minimize animal suffering in the field, and the entire gut was dissected from each animal and preserved separately in sterile tubes with ethanol. Finally, after all of the samples were transferred to the laboratory, we washed the entire gut 3× with sterile water and expelled the fresh feces from it into new sterile tubes under sterile conditions via ethanol-sterilized forceps. Before DNA extraction, the fecal samples were stored separately in sterile tubes at –20°C.

All animal works were conducted in accordance with Animal Research Protocol IOZ-2006 approved by the Animal Care Committee of the Institute of Zoology, Chinese Academy of Sciences (IOZCAS), China.

DNA extraction and sequencing

The total DNA of feces was extracted using a TGuide 96 Soi genomic DNA extraction kit (TIANGEN Biotech, China) following the manufacturer's protocol. The extracted DNA was diluted to 10 ng/μL for PCR amplification. We used the universal primers 515F (5′-ACTCCTACGGGAGGCAGCA-3′)/806R(5′-GGACTACHVGGGTWTCTAAT-3′) to amplify the V3-V4 region of the microbial 16S rRNA gene (Tamaki et al. 2011). The 515F primer with a 12bp barcode was inserted to differentiate each sample during sequencing analysis. PCR amplifications were performed in duplicate with 10 μL reaction mixture containing 5 μL of PCR buffer, 0.2 μL of each deoxynucleoside triphosphate (dNTP) at 2 mM, 0.3 μM of each primer, 0.2 U of KOD FX Neo (TOYOBO, Beijing) and 10 ng genomic DNA. The thermal cycling procedure consisted of an initial denaturation step at 95°C for 5 min, followed by 25 cycles of 95°C for 30 s, 50°C for 30 s, and 72°C for 40 s, and a final extension at 72°C for 7 min. Ultimately, we used an Illumina Novaseq6000 (Illumina, USA) platform to sequence the PCR amplicons after acquiring 2 × 250 bp paired-end sequences. The above services were provided by Berry Genomics (Beijing, China).

Preprocessing and quality control

We processed the raw sequencing data with Amplicon Analysis Pipeline-Version 1.14 (Liu et al. 2021). First, the paired reads were merged to obtain amplicon sequences, and the primers were removed. Then, low-quality amplicon sequences (those with quality scores less than 20, lengths less than 350 bp, or ambiguous bases) were removed. All these procedures were performed with USEARCH (Edgar 2010). All sequences were denoised to amplicon sequence variants (ASVs) using unoise3 in USEARCH (Edgar and Flyvbjerg 2015). The aligned 16S rRNA gene sequences were input into the UCHIME algorithm for a chimera check (Edgar et al. 2011). Additionally, we filled out a feature table (amplicon sequence variant ASV table) by quantifying the frequency of the feature sequences in each sample. Owing to possible contamination of the

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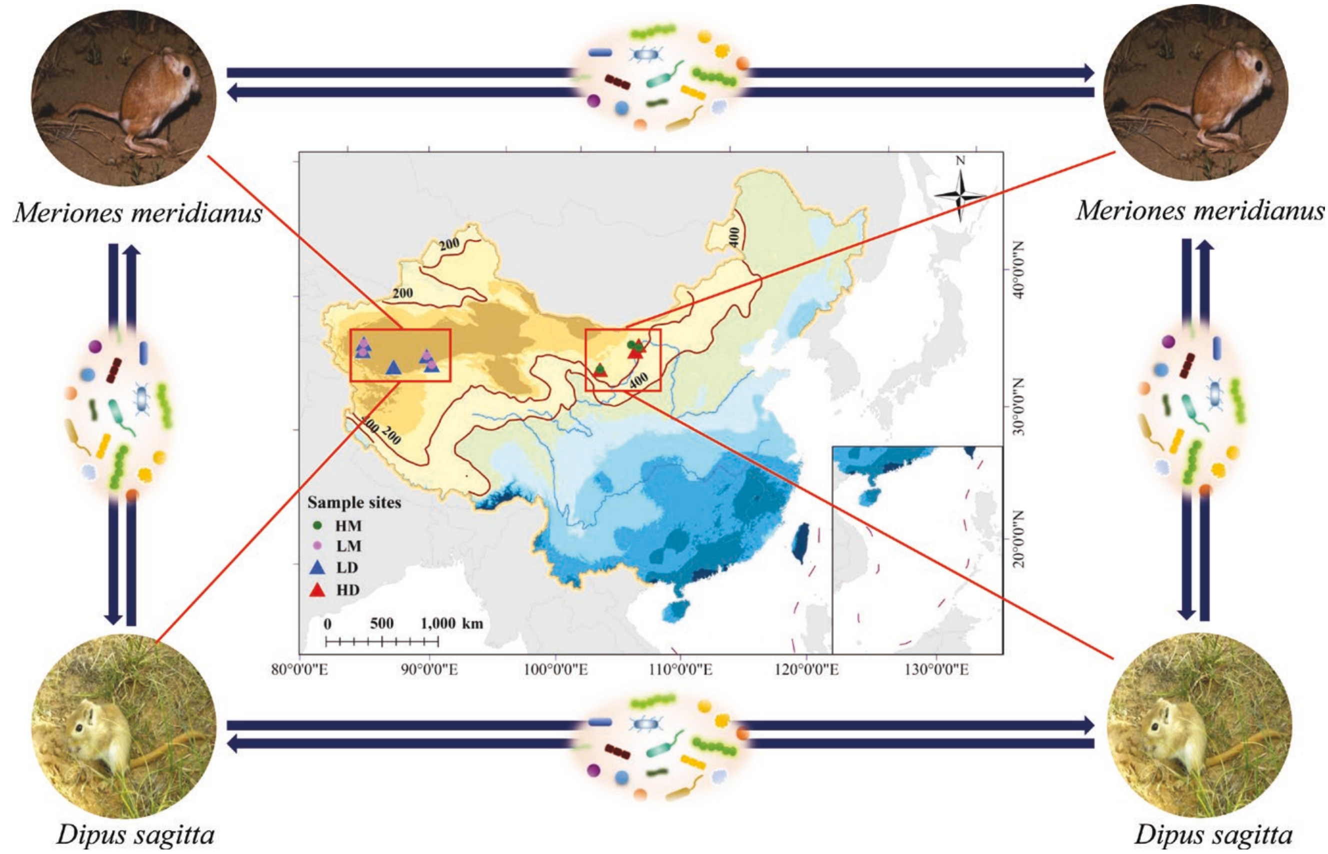

Figure 1 A map showing the locations of sampling sites marked with different symbols across the research area. Symbols include green circles for group HM: *M. meridianus* from high rainfall regions, purple circles for group LM: *M. meridianus* from low rainfall regions, blue triangles for group LD: *D. sagitta* from low rainfall regions, and red triangles for group HD: *D. sagitta* from high rainfall regions. Boundaries of countries and rivers were acquired from https://malagis.com/category/gis-resource/

chloroplast sequences during PCR amplification, we then removed nonbacterial ASVs, chloroplasts and singletons from our ASV list (https://www.festinalente.me/bioinf/). The ASVs were assigned to the taxonomy group against the Silva 128 database (http://www.arb-silva.de). A total of 119,719,48 sequences were obtained from 39 samples, and a total of 575,640 high-quality sequences were obtained after splicing and quality control of the data (Table S2, Supporting Information). In total, we recovered 2,928 bacterial ASVs from all samples. Because uneven sequencing depth across samples may confuse the results of community comparisons, we rarefied each sample to the same number of reads (14,000 sequences) with the "vegan" package (Oksanen et al., 2018) in R software.

Diversity and composition measures

The "vegan" package was used to calculate 4 indices of alpha diversity: the Chao1, ACE, Shanon and Simpson diversity indices. Analysis of Variance (ANOVA) was then performed to determine whether the alpha diversity differed between the groups. Pairwise comparisons were performed using Tukey's Honest Significant Difference (HSD) test. We used Principal coordinate analysis (PCoA) with the Bray–Curtis distance to evaluate the dissimilarities in community composition between samples and assess their beta diversity. To determine differences in the community composition of microbiomes among samples, a permutational multivariate analysis of variance (PERMANOVA) was carried out with 9,999 permutations using the adonis function (Anderson 2014). For the analysis of the core microbiota, we defined ASVs, which were present in more than 50% of the samples and had a relative abundance of more than 0.5% as core ASVs. Similarly, the genera associated with these ASVs were categorized as core genera. Linear discriminant analysis (LDA) effect size (LEfSe) (Segata et al. 2011) was performed to identify the specialists for the various bacterial communities among groups at the ASV level using the criteria of an LDA score ≥4.0 and a Kruskal–Wallis test significance level ≤0.05. Furthermore, we obtained species-specific ASVs with the stricter criteria of an LDA score ≥4.0 and a significance level ≤0.05 to facilitate visualization as a phylogenetic tree. The phylogenetic tree was annotated and visualized online (https://www.bic.ac.cn/ImageGP/).

We performed LMM with microbiome richness as the response variable and model predictors that included species, environment, and the species × environment interaction, with the collection site representing a random intercept to account for repeated sampling of populations, in the package "lme4" in R (Bates et al. 2015).

Functional prediction of the microbiome

Phylogenetic Investigation of Communities by Reconstruction of Unobserved States (PICRUSt) v1.1.0 (Langille et al. 2013) was used to predict the metagenomic functional composition (Zheng et al. 2019) of Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways (Kanehisa and Goto 2000).

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Community assembly process

The ecological processes were quantified using a novel method named infer community assembly mechanisms by the phylogenetic-bin-based null model (iCAMP), which was developed from the method of Stegen et al (Stegen et al. 2013; Ning et al. 2020). First, iCAMP assigned the observed taxa into phylogenetic-closed groups (bins). We subsequently calculated the within-bin beta Net Relatedness Index (βNRI) and the modified Raup–Crick metric (RC) to estimate the relative contributions of homogeneous selection (HoS; $\beta NRI < -1.96$), heterogeneous selection (HeS; $\beta NRI > 1.96$), homogeneous dispersal (HD; $RC < -0.95$ and $|\beta NRI| \leq 1.96$), dispersal limitation (DL; $RC > 0.95$ and $|\beta NRI| \leq 1.96$), and drift (DR; $|RC| \leq 0.95$ and $|\beta NRI| \leq 1.96$) in governing microbial community assembly. This analysis was conducted with the iCAMP (version 1.3.4) package (https://github.com/DaliangNing/iCAMP1; accessed on 5 Nov 2022). On the basis of the principle of the null models employed by iCAMP, dispersal limitation, homogenizing dispersal, and drift fractions were considered stochastic. Thus, the sum of their estimated relative importance can be used to estimate the stochasticity of community assembly (Ning et al. 2020).

Data availability

The sequence data have been deposited in the NCBI Sequence Read Archive (SRA) under the accession number PRJNA1058805 https://www.ncbi.nlm.nih.gov/sra/PRJNA1058805.

Results

Bacterial community variation between sympatric and allopatric species

Alpha diversity indices were calculated to clarify the differences in the richness and diversity of the bacterial communities from different geographic sources. We used the Chao 1, ACE, Shannon and Simpson indices to determine the alpha diversity of the bacterial microbiome at the ASV level. In terms of sympatric groups, both the chao1 index and ACE index of *M. meridianus* gut bacterial communities were greater than those of the *D. sagitta* bacterial communities in the D and H regions. Regarding allopatric species, the Chao 1 index and ACE index of the LD population were higher than those of the HD population, and they were also greater in the LM population than in the HM population ($P < 0.05$ for both; Figure 2A and Figure S2B). The Shannon and Simpson indices were similar between the groups ($P > 0.05$; Figure 2B and Figure S2C).

PCoA based on Bray-Crutis distance revealed that for both regions, the bacterial communities of *D. sagitta* and *M. meridianus* were significantly different from each other (LD vs. LM: PERMANOVA: $F = 1.91$, $R^2 = 0.07$, $P < 0.001$; HD vs. HM: PERMANOVA: $F = 1.87$, $R^2 = 0.15$, $P < 0.001$). In addition, among *D. sagitta*, the bacterial communities at different sites were significantly different (PERMANOVA: $F = 1.84$, $R^2 = 0.09$, $P < 0.001$). However, for *M. meridianus*, whereas the difference between regions was not significant, there was still a noticeable trend, which was also quite clear from the ordination, as the groups did not really overlap (PERMANOVA: $F = 1.28$, $R^2 = 0.07$, $P = 0.08$) (Figure 2C, Table S3).

On the basis of the 16S rRNA gene sequences, we also analyzed the bacterial taxonomic compositions, and the ASV obtained via16S rRNA amplicon sequencing were classified into 14 phyla, 23 classes, 43 orders, 88 families, and 221 genera. In addition to their overall bacterial properties, the core ASVs also differed between host species (Figure 2D). We defined the core ASVs as those observed at more than 50% of the samples in each group with a relative abundance > 0.5%. Among the 2,928 bacterial ASVs detected in this study, 7 and 5 ASVs were identified as the core ASVs of *D. sagitta* and *M. meridianus*, respectively (Table S4). The 7 core ASVs of *D. sagitta* were assigned to the genera *Kineothrix* (including 2 ASVs), *Paramuribaculum*, *Pseudescherichia*, *Ligilactobacillus*, *Clostridium_XIVa*, and *Akkermansia*; the 5 core ASVs of *M. meridianus* were assigned to the genera *Lactobacillus* (including 2 ASVs), *Paramuribaculum* (2), and *Barnesiella*. Among these core ASVs, 3 assigned to *Paramuribaculum* were shared by both host species (Figure 2D and Table S4).

In addition to the general bacterial features (e.g., diversity and composition), group-specific taxa were identified via LEfSe. Three phyla, 3 classes, 3 orders, 4 families, and 6 genera were dominant in distinct groups. The bacterial microbiome of the LD population contained the majority of the distinct species, which were classified individually into the following groups: Bacteroidales, Bacteroidia, Bacteroidetes, Muribaculaceae, *Paramuribaculum*, *Akkermansia*, Akkermansiaceae, Verrucomicrobiales, Verrucomicrobiae, and Muribaculaceae. The LM population contained 5 distinct species, which were significantly abundant: Bacilli, Lactobacillales, Lactobacillaceae, and 2 unidentified genera. Two significantly abundant taxa in the HD population were Firmicutes and *Clostridium XIVa*; in the HM population, only 1 taxonomic genus, *Lactobacillus*, was enriched ($P < 0.05$; logarithmic LDA score >4.0) (Figure 2F and G).

According to the LMM analysis, there was a significant positive correlation between species and microbiome richness (Table 1). However, there was no strong or consistent correlation between the richness of the microbiome samples and the environment or species × environment interaction. Specifically, the marginal *R*-squared (R2m) of 0.2836 indicates that the fixed effects (species, environment, and their interaction) collectively explain approximately 28.36% of the variance in richness. This suggests that species and environmental factors individually contribute significantly to the variations in richness. Moreover, the conditional *R*-squared (R2c) of 0.5553, which considers both fixed effects and random effects (variations at the site level), indicated a more comprehensive model fit, explaining a substantial proportion of the total variance in the response variable. This higher R2c value underscores the model's robustness in capturing the overall variability in microbiome richness, accounting for both fixed and random effects across different environmental conditions and species interactions (Table 1).

Functional prediction of bacterial community

The functions of each group of bacterial communities were predicted using PICRUSt in the KEGG database, and the top 25 core functions at level 3 of each group are shown in Figure 3. The results revealed that while the extent of interindividual variation in microbiome composition differed between host species based on the identities of the bacterial taxa they included, there was no difference in the degree of interindividual variation in the predicted functional pathways between the groups. "Transporters" and "ABC transporters" were the dominant bacterial community proteins in all the groups.

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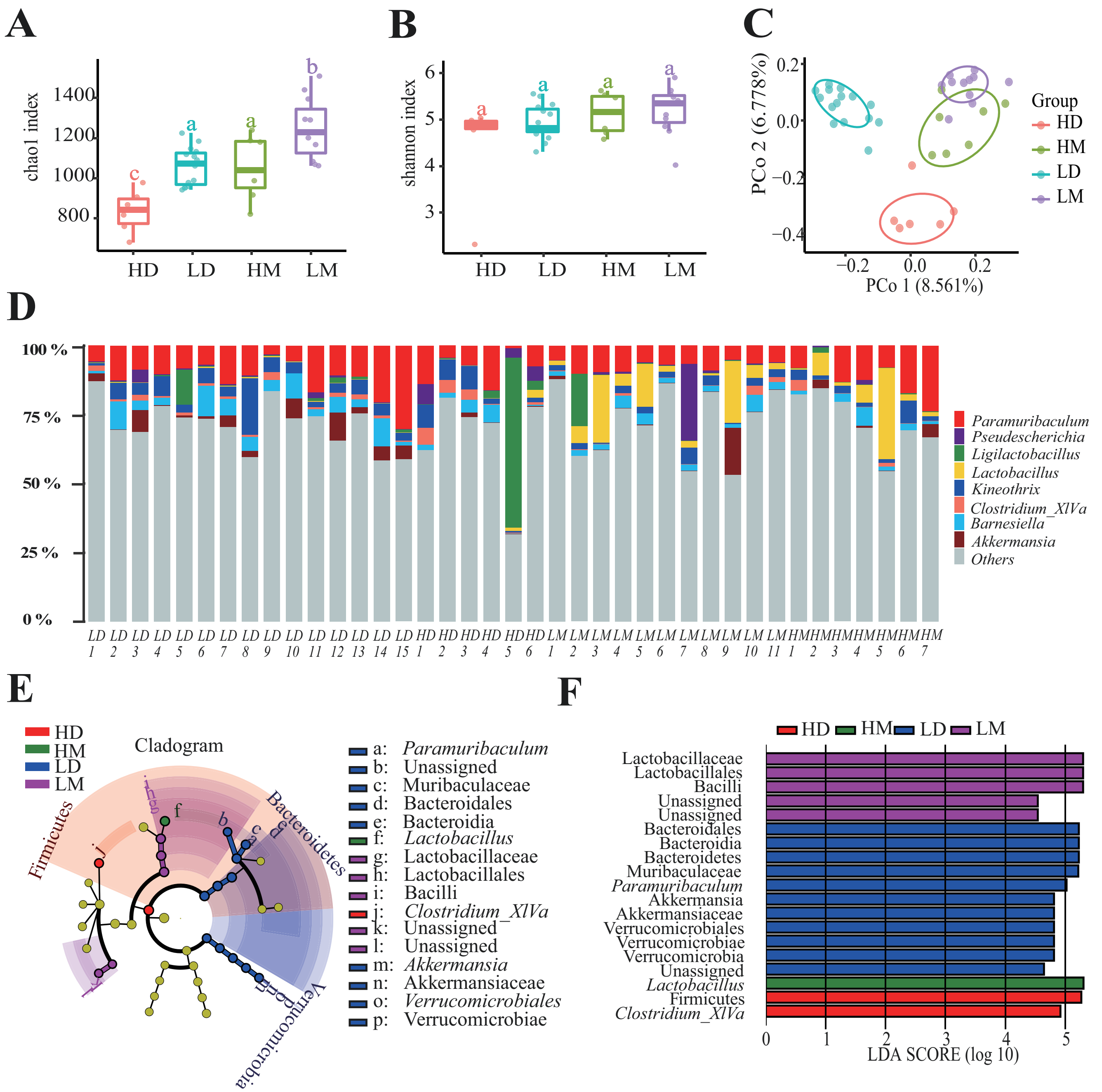

Figure 2 Diversity, structure, and composition in *D. sagitta* and *M. meridianus*. Box and whisker plots of 2 alpha diversity indices (A) Chao1 and (B) Shannon indices in each group. Different letters above the whiskers denote significant differences between each group determined with analysis of variance tests ($P < 0.05$). (C) PCoA plots showing group variation in the bacterial microbiome based on Bray–Curtis dissimilarity. The ellipses indicate the 95% confidence intervals, and the percentages in parentheses are the proportions of variation explained by the PCoA. (D) The 8 core genera (present in more than 50% of samples in each group with a relative abundance of more than 0.5%.) across all samples. (E) Cladogram depicting the phylogenic relationships between microbial taxa, which were significantly different in each group. Yellow nodes represent no significant differences. (F) The LDA histogram represents the bacterial groups with significant differences between groups ($P < 0.05$, Kruskal–Wallis test; logarithmic LDA score > 4.0).

Gut bacterial community assembly process

To further evaluate the ecological processes of the gut bacterial community assembly among sympatric desert rodents from different geographic sources, null model analyses based on taxonomic and phylogenetic metrics were performed (Ning et al. 2020). According to the analysis, dispersal limitation was more important than other processes in bacterial community assembly, with average relative importance values of 52.89% in HD, 67.83% in LD, 60.59% in HM, and 64.79% in LM (Figure 4B). On the basis of the principle of the null models employed by iCAMP, the dispersal limitation, homogenizing dispersal, and drift fractions were stochastic. Thus, the sum of their estimated relative importance can be used to estimate the stochasticity of the community assembly. The relative importance of stochastic processes were 83.10%, 87.79%, 83.01%, and 81.14% in the HD, LD, HM, and LM groups, respectively (Figure 4A). These results revealed that the

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biogeographic patterns of the microbiome were driven by dispersal limitation and then drift. This might be due to long geographic distances limiting host dispersal in various desert ecosystems.

Table 1. Liner mixed model (LMM) analysis testing the effects of species, environment, and the species × environment interaction on microbial community measured using richness

Variable	Estimate	Std. error	df	*t* Value	Pr(>|*t*|)
Intercept	791.747	114.290	0.394	6.928	0.310
Species	163.835	64.892	34.881	2.525	0.016
Environment	−0.770	1.126	0.797	−0.684	0.640
Species: environment	0.194	0.771	34.909	0.251	0.803
Model fit	R^{2m}	0.284	R^{2c}	0.555	

Discussion

This study was conducted to explore the microbial community ecology of 2 sympatric desert rodents (*D. sagitta* and *M. meridianus*) from different geographic sources, with a special interest in microbial community assembly processes. Contrary to our initial expectations, this study revealed that the microbiome differed significantly both between sympatric and allopatric species (Figure 2A–C). In addition, we detected a significant positive correlation between species and microbiome richness (Table 1). These findings suggest that, across sympatric rodents of different families, species have a greater influence on bacterial communities than do environmental factors. Unexpectedly, our results also do not support our second hypothesis: as allopatric species, both *D. sagitta* and *M. meridianus* presented similar patterns. Rodents in the L region, where low precipitation and high temperatures are present, harbor bacterial communities with relatively high

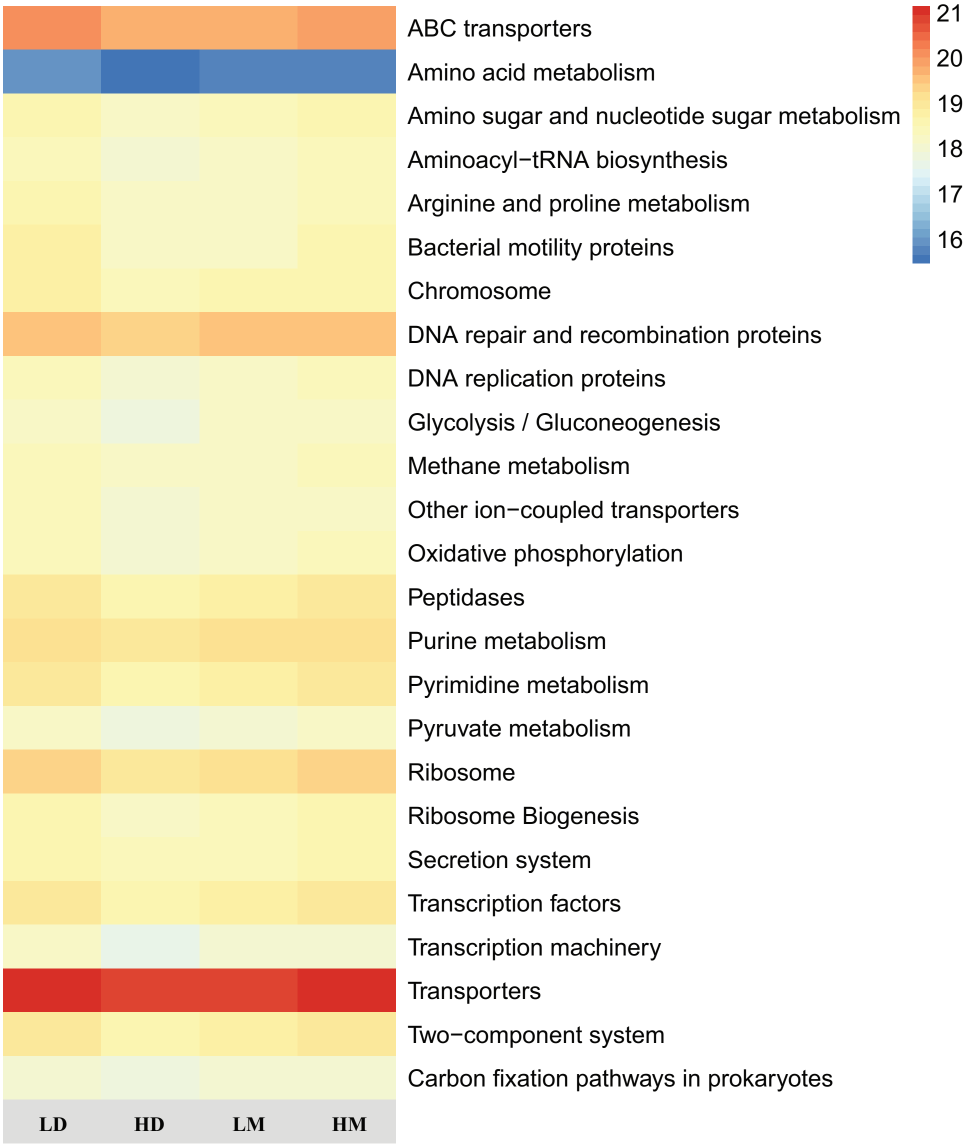

Figure 3 Heatmap illustrating the function traits (top 25 core functions at level 3) of bacteria community among each group. The abundance of functional traits was normalized, and the scale bar referred to the log10 value.

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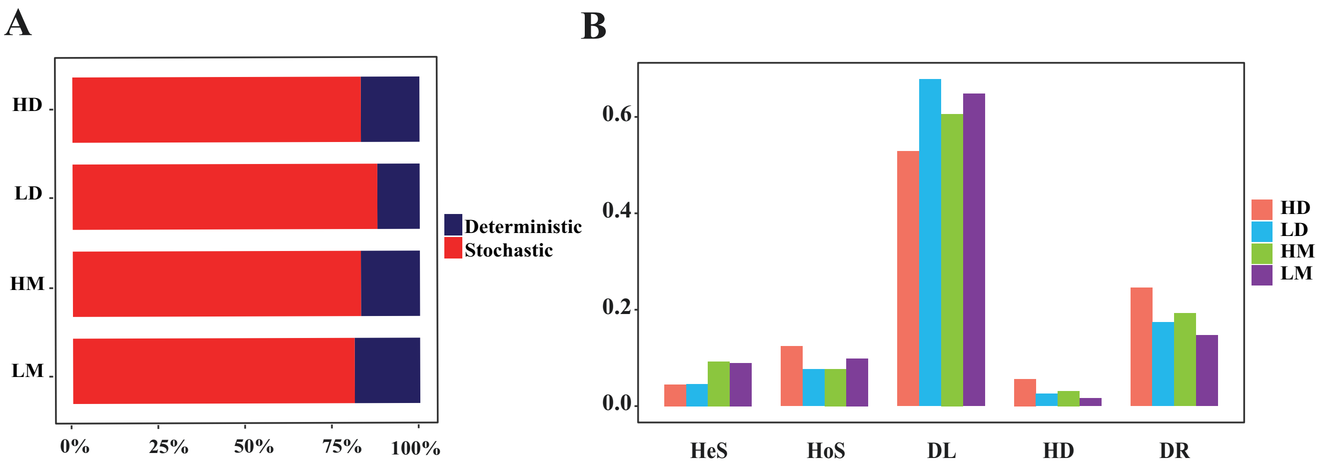

Figure 4 Relative importance of different ecological processes. (A) The relative importance of deterministic and stochastic processes on bacterial community assembly in each group. (B) Bar plot of relative contributions of different ecological processes in each group (HeS: heterogeneous selection; HoS: homogeneous selection; HD: homogenizing dispersal; DL: dispersal limitation; and DR: drift).

alpha diversity (Figure 2A–C). The data also revealed evidence contrary to our third hypothesis we found that stochastic processes (e.g., dispersal limitation) dominated in shaping desert rodent microbiota composition (Figure 3). Although there are several reports on the soil microbiota composition, diversity and assembly process in different desert ecosystems (Feng et al. 2018; Bay et al. 2021; Dong et al. 2024), we are the first to study the microbial community assembly process in desert rodents across a large geographic span. These results expand our understanding of bacterial community assembly patterns in mammals.

We detected significant differences in alpha and beta diversity within the bacterial communities of different rodent species. Moreover, LMM analysis was carried out (Table 1), which revealed that microbial richness can be affected mostly by species. These results are similar to those of other studies in wild rodents (Knowles et al. 2019; Anders et al. 2021), in which species identity dominated over environment in shaping the microbiota of small mammals. In our study, the 2 sympatric species (*D. sagitta* and *M. meridianus*) were from different rodent families, which may have contributed to the differences observed across these rodent hosts. In addition, we found that, compared with *D. sagitta*, *M. meridianus* presented greater microbial diversity in each region. Previous studies have shown that herbivores have greater microbial diversity in their guts than mammals with other feeding strategies (Ley et al. 2008). It is hypothesized that this greater diversity is related to the complexity of fibers present in plant material, and so the diversity of bacterial communities in herbivorous rodents may assist them in digestion. Interestingly, there are also relationships between body size and microbial diversity, such that larger animals tend to have greater diversity, and this relationship is exaggerated in herbivorous species (Nishida and Ochman 2018). In addition, we identified different core genera in each host species, and the core genus *Lactobacillus* has been reported to perform essential functions in herbivore adaptation (Yu et al. 2018). Compared with *D. sagitta*, which consumes small amounts of insects, *M. meridianus* is entirely herbivorous. *M. meridianus* are also considerably larger than *D. sagitta*, which may also contribute to the differences observed across these rodent hosts. However, these ideas require validation and further study. In our future research, we will analyze their phylogenetic relationships, dietary habits, and morphological data in detail.

As allopatric species, both *D. sagitta* and *M. meridianus* presented similar patterns. Rodents in the L region, where low precipitation and high temperatures are present, harbor bacterial communities with relatively high richness. However, these results are not consistent with the findings of Li et al. (2020) and Brown et al. (2024). The former demonstrated that there was no significant difference in the alpha diversity of rodent bacterial communities under different precipitation manipulation conditions. The latter showed that high rainfall was associated with greater microbial richness in co-occurring rodent species. The metabolic diversity harbored by microbial communities facilitates an array of unique functions that may directly benefit hosts thus these communities may be routes of rapid ecological and evolutionary adaptation for their hosts (Alberdi et al. 2016; Henry et al. 2021). These microbial communities can be especially important for the degradation of complex biopolymers that animal hosts cannot metabolize themselves. For example, plant material contains cell walls and structural components made of carbohydrate monomers linked by β-1,4 glycosidic bonds, which cannot be broken down by vertebrate enzymes (Karasov and Martínez 2007). Therefore, many herbivorous mammals rely on the activity of microbial symbionts to conduct fermentative processes and then they absorb the microbial byproducts of this metabolism. We speculate that the bacterial communities of low-rainfall desert rodents with greater alpha diversity probably exhibit greater adaptation to desert habitats. The functional prediction results revealed that the bacterial community functions in transport were the highest across groups, but the differences between groups were not significant (Figure 3). In the future, metagenomic sequencing approaches will be needed to specifically elucidate their specific functions.

To further explore bacterial community turnover in different ecological desert niches and hosts, null model analysis was carried out using the framework described by Stegen et al. (2013). Selection is the result of biotic and abiotic pressures that cause variation in reproductive success across species. Dispersal is the degree to which individuals move between communities, and drift results from population size. In this study, our results suggested the predominant importance of

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dispersal limitation and ecological drift in shaping bacterial microbiome variation among groups (Figure 3A and B), which indicated that dispersal between the sympatric and allopatric species was very limited and involved undominated processes (Dini Andreote et al. 2015; Zhou and Ning 2017). The bacterial communities were affected by both biotic (species) and abiotic (environment) pressure, but overall, the null modelling results suggested that the bacterial communities were dominated by stochastic processes rather than deterministic processes, indicating that random birth, death, and dispersal events can strongly affect the distribution of the bacterial communities of desert rodents, resulting in a species composition that is indistinguishable from patterns arising randomly. Furthermore, when a response of the microbiome to environmental or physiological variation is observed, deterministic processes must not be assumed as the sole causal process.

In summary, deserts present formidable challenges for desert mammal life because of their different arid conditions. Therefore, this study was conducted with the hypothesis that habitat specialization due to climate change-related aridity levels drives the differentiation of bacterial diversity patterns, community functions, and assembly processes across various ecosystems within the vast deserts of China. The results revealed that the microbiomes of both sympatric and allopatric species significantly differ. In addition, *M. meridianus* had greater microbiome diversity than *D. sagitta*, and both rodent microbiome diversity indices were greater in low-rainfall regions, as confirmed by the alpha and beta diversity indices. The correlations among species, the environment, and sites with microbiome richness revealed that the species highly influences microbial richness. The bacterial community composition revealed that each group had specific core genera, and the species in the low-rainfall regions presented greater specific bacterial abundances. Moreover, microbial community assembly process analysis revealed that gut bacterial diversity followed the dispersal limitation pattern (which involves undominated processes). These results elucidated the diversity patterns and assembly processes of microorganisms in small desert mammals and provided fundamental information necessary to understand the microbiome composition of different desert mammals in diverse desert ecosystems exposed to severe climate change. However, there are still some limitations in the microscopic-level functional analysis (metagenomics and metatranscriptomics) that need to be addressed in future research. To further explore microbial functions in desert rodents, we recommend conducting metagenomic, meta-transcriptomic, and metabolomic studies. Furthermore, it would be beneficial to compare the patterns of desert rodent microbial communities with those of soil and plant microbial communities. By comparing the microbial communities across desert mammals, soil, and plants, we can identify unique adaptations and interactions, which occur in these environments. This comparative analysis enhances our understanding of the functional dynamics of desert microbial communities.

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Conflict of Interest statement

The authors declare that they have no conflict of interest.

Supplementary material

Supplementary material can be found at https://academic.oup.com/cz.

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