

Figure S1. Experimental design of the study. The numbers on the timeline indicate soil CO_2 emission and C isotopic signature ($\delta^{13}\text{C}$) of CO_2 . Soil and microbial properties were detected after a 90-day incubation period.

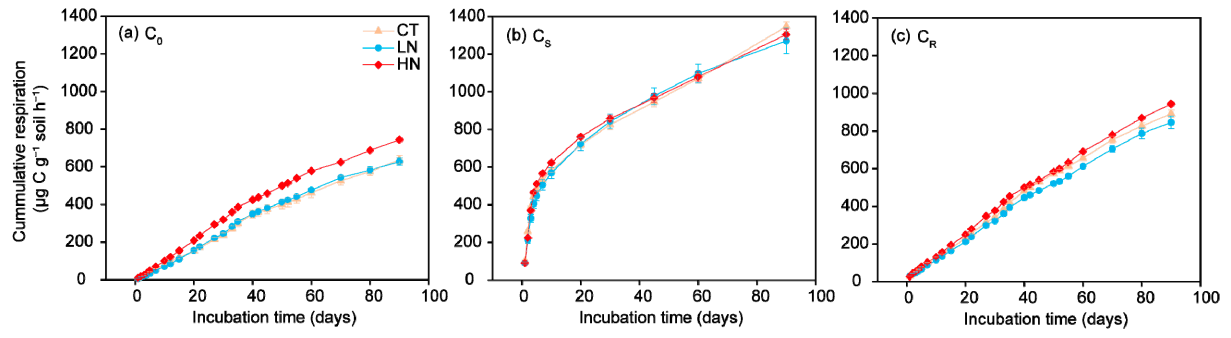


Figure S2. Temporal changes in CO₂ emission in different C-addition treatments. **(a)** C₀, no C addition; **(b)** C_s, single C addition; **(c)** C_R, repeated C additions. Abbreviations: CT, control; LN, low-level N addition; HN, high-level N addition.

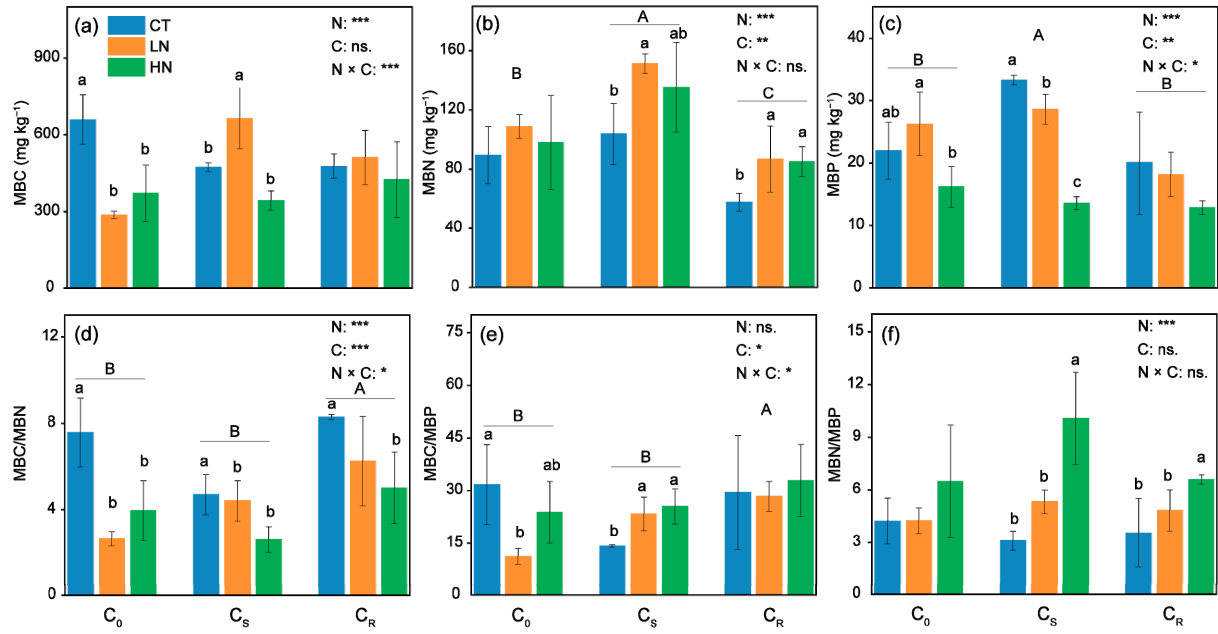


Figure S3. C, N, and P concentrations in soil microbial biomass (a–c) and their stoichiometry (d–f). Two-way analysis of variance (ANOVA) was used to determine the effect of N addition, C addition mode, and the interaction ($N \times C$) between N- and C-addition treatments. Asterisks (*) indicate a significant difference (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, ns. $p > 0.05$). Different lower-case and upper-case letters indicate significant differences among N- and C-addition treatments, respectively. Abbreviations: MBC, microbial biomass C; MBN, microbial biomass N; MBP, microbial biomass phosphorus; C₀, no C addition; C_s, single C addition; C_R, repeated C additions; CT, control; LN, low-level N addition; HN, high-level N addition.

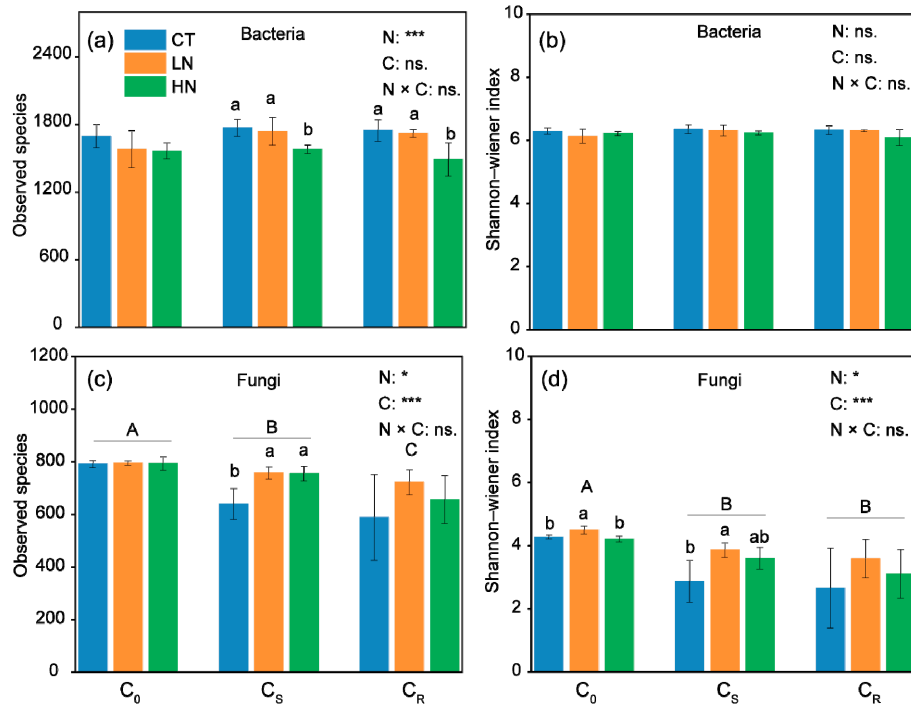


Figure S4. Diversity of soil microbial community. (a)–(b) and (c)–(d) indicate the observed species and Shannon–Wiener index of bacteria and fungi, respectively. Two-way analysis of variance (ANOVA) was used to determine the effect of N addition, C addition mode, and the interaction ($N \times C$) between N- and C-addition treatments. Asterisks (*) indicate a significant difference (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, ns. $p > 0.05$). Different lower-case and upper-case letters indicate significant differences among N- and C-addition treatments, respectively. Abbreviations: C₀, no C addition; C_S, single C addition; C_R, repeated C additions; CT, control; LN, low-level N addition; HN, high-level N addition.

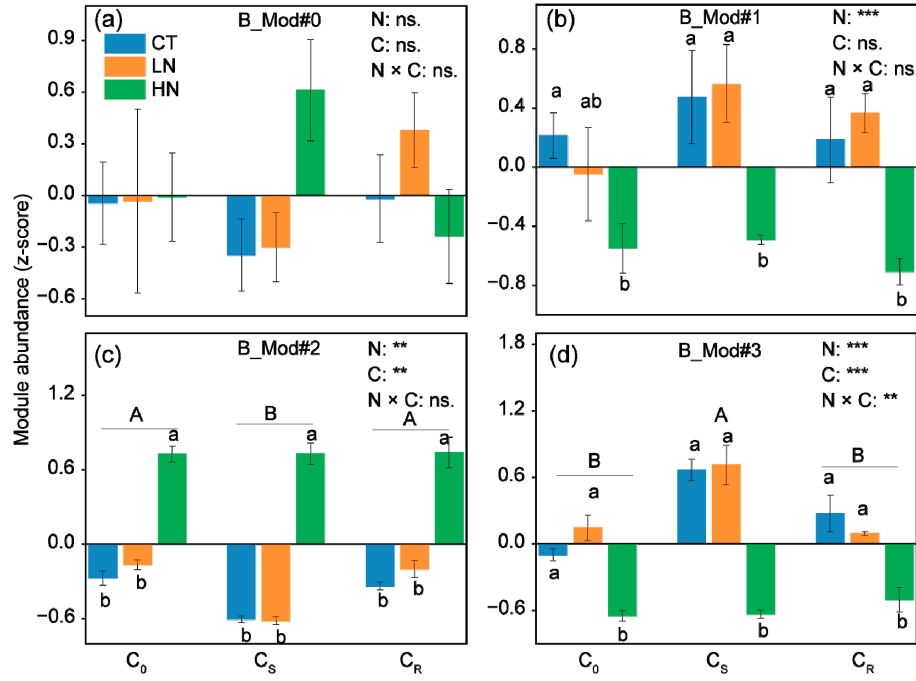


Figure S5. Relative abundance of the bacterial modules in different treatments. Two-way analysis of variance (ANOVA) was used to determine the effect of N addition, C-addition mode, and the interaction ($N \times C$) between N- and C-addition treatments. Asterisks (*) indicate a significant difference ($**p < 0.01$, $***p < 0.001$, ns. $p > 0.05$). B_Mod#0–3 were the major bacterial modules (a–d). Different lower-case and upper-case letters indicate significant differences among N- and C-addition treatments, respectively. Abbreviations: C₀, no C addition; C_S, single C addition; C_R, repeated C additions; CT, control; LN, low-level N addition; HN, high-level N addition.

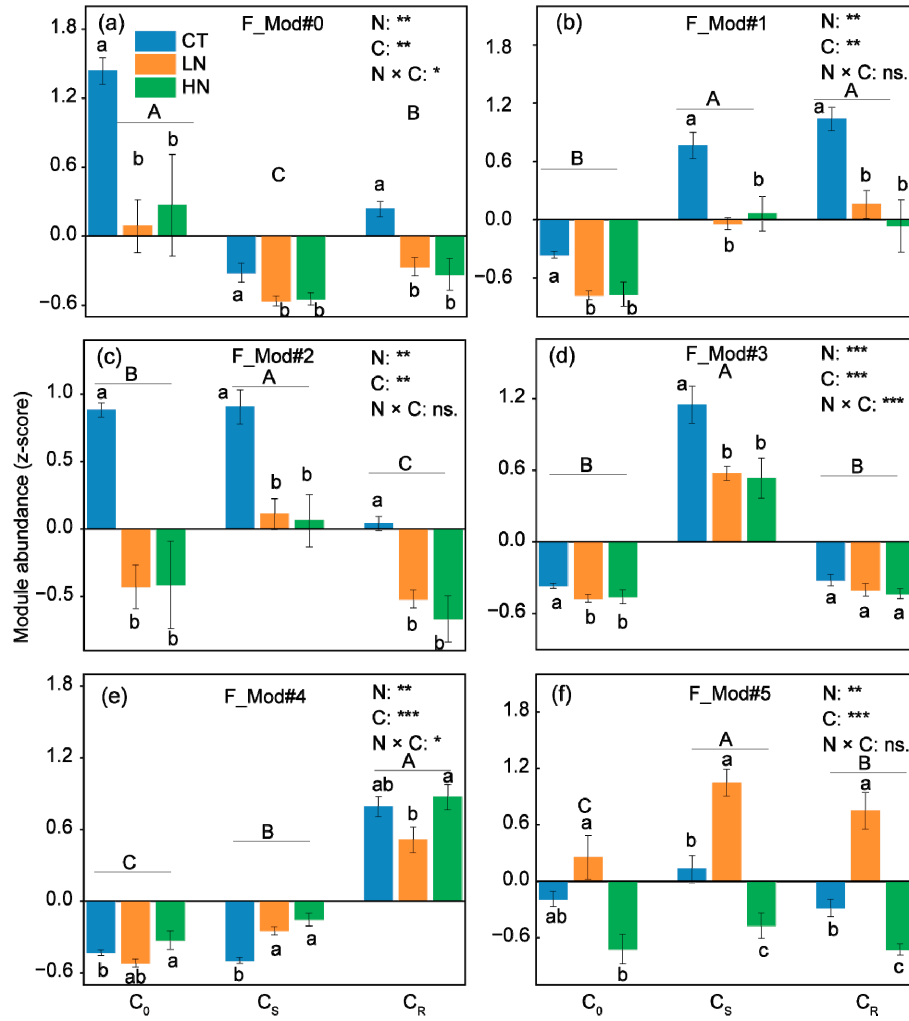


Figure S6. Relative abundance of the fungal modules under different treatments. Two-way analysis of variance (ANOVA) was used to determine the effect of N addition, C-addition mode, and the interaction ($N \times C$) between N- and C-addition treatments. Asterisks (*) indicate a significant difference ($*p < 0.05$, $**p < 0.01$, $***p < 0.001$, ns. $p > 0.05$). F_Mod#0–5 were the major fungal modules (a–f). Different lower-case and upper-case letters indicate significant differences among N- and C-addition treatments, respectively. Abbreviations: C₀, no C addition; C_s, single C addition; C_R, repeated C additions; CT, control; LN, low-level N addition; HN, high-level N addition.

Table S1 Effect of long-term N addition on physicochemical properties of soil before incubation. Different lowercase letters indicate significant differences between different N-addition treatments. Abbreviations: TC, total carbon; TN, total N; TP, total phosphorus; DOC, dissolved organic carbon; MN, mineral N; AP, available phosphorus; CT, control; LN, low-level N addition; HN, high-level N addition.

Treatments	pH	TC (g kg ⁻¹)	TN (g kg ⁻¹)	TP (g kg ⁻¹)	DOC (mg kg ⁻¹)	MN (mg kg ⁻¹)	AP (mg kg ⁻¹)
CT	3.80	51.84	4.26	0.82	170.39a	17.90b	8.25a
LN	3.82	52.21	4.15	0.80	134.02b	18.44ab	6.06b
HN	3.76	53.52	4.45	0.81	176.28a	25.14a	5.89b

Table S2 Correlation coefficients between the relative abundances of dominant bacterial and fungal taxa at the phylum level and cumulative priming effect. * $p < 0.05$; numbers are partial correlation coefficients (r) of Spearman's correlation.

Dominant Phyla		Under different N addition gradients		Under different C addition modes	
		r	<i>p</i>	r	<i>p</i>
Bacteria	Actinobacteria	−0.40		−0.41	
	Bacteroidetes	−0.39		−0.31	
	Chloroflexi	−0.41		−0.16	
	Firmicutes	−0.12		0.06	
	Planctomycetes	0.25		0.06	
	Proteobacteria	0.51	*	0.43	*
	Verrucomicrobia	−0.03		−0.07	
	Unassigned	0.23		0.03	
Fungi	Ascomycota	−0.37		0.20	
	Basidiomycota	0.39		−0.10	
	Mortierellomycota	−0.10		0.07	
	Rozellomycota	0.09		0.22	
	Unassigned	−0.40		−0.08	

Table S3 Correlations between the relative abundances of dominant bacterial and fungal modules and cumulative priming effect (PE). Mixed linear-model regressions were used to analyze to the relationship between cumulative PE and bacterial and fungal dominant modules under different N-addition addition treatments or C-addition modes, whereby variance of one treatment was eliminated by selecting either N- or C-addition treatment as a random factor. B_Mod#0, B_Mod#1, B_Mod#2, B_Mod#3, and B_Mod#4 were the key bacterial modules; F_Mod#0, F_Mod#1, F_Mod#2, F_Mod#3, F_Mod#4, and F_Mod#5 were the key fungal modules. *p* values represent significant levels; *F* values represent the proportion of total variance explained for the dependent variable of cumulative PE. Microbial ecological modules significantly related to PE are indicated in bold text.

Under different N addition treatments	<i>F</i>	<i>p</i>	Under different C addition modes	<i>F</i>	<i>p</i>
B_Mod#0	3.211	0.088	B_Mod#0	0.73	0.402
B_Mod#1	6.507	0.019	B_Mod#1	3.115	0.091
B_Mod#2	11.21	0.003	B_Mod#2	4.231	0.052
B_Mod#3	14.093	0.001	B_Mod#3	6.353	0.019
F_Mod#0	0.133	0.7195	F_Mod#0	0.003	0.955
F_Mod#1	4.554	0.045	F_Mod#1	0.346	0.563
F_Mod#2	0.313	0.582	F_Mod#2	0.266	0.612
F_Mod#3	0.012	0.913	F_Mod#3	0.017	0.897
F_Mod#4	6.495	0.019	F_Mod#4	3.536	0.073
F_Mod#5	0.14	0.712	F_Mod#5	61.053	< 0.001

Table S4 Effects of long-term nitrogen (N) addition and carbon (C) addition mode on bacteria and fungi in different phylogenetic levels (%).

Taxonomy	C ₀			C _S			C _R			Two-way ANOVA analysis			Among group			Among group		
	CT	LN	HN	CT	LN	HN	CT	LN	HN	N	C	N × C	CT	LN	HN	C ₀	C _S	C _R
Bacteria_Phylum-level																		
Actinobacteria	25.93	27.65	31.08	23.75	23.25	29.70	26.68	26.15	33.68	*	***	ns.	a	b	a	B	B	A
Bacteroidetes	0.29	0.48	2.29	0.19	0.31	2.24	0.52	0.55	3.88	*	***	ns.	b	b	a	B	B	A
Chloroflexi	7.27	5.57	8.72	5.07	4.89	8.90	6.03	7.82	6.09	ns.	ns.	ns.	a	a	a	A	A	A
Firmicutes	19.55	18.25	20.90	16.35	17.70	20.00	17.38	18.53	17.25	ns.	ns.	ns.	a	a	a	A	A	A
Planctomycetes	1.24	0.97	0.94	1.26	1.32	1.06	1.30	1.58	0.66	ns.	ns.	ns.	a	a	a	A	A	A
Proteobacteria	40.55	42.15	31.53	47.98	47.15	33.30	42.65	39.05	34.95	*	***	ns.	ab	a	b	A	A	B
Verrucomicrobia	0.82	1.09	0.79	0.92	1.08	1.20	1.38	1.25	0.62	ns.	ns.	ns.	a	a	a	A	A	A
Unassigned	3.62	3.14	3.21	3.56	3.33	3.02	3.25	4.16	2.50	ns.	ns.	ns.	a	a	a	A	A	A
Others	0.74	0.70	0.54	0.94	0.97	0.58	0.83	0.92	0.38	*	***	ns.	b	a	ab	A	A	B
Bacteria_Genus-level																		
Acidiferrimicrobium	1.01	1.20	1.26	1.02	1.04	1.04	0.91	1.05	1.18	ns.	ns.	ns.	a	a	a	A	A	A
Actinoallomurus	1.36	1.43	1.80	1.11	1.06	1.62	1.36	1.23	1.81	*	***	ns.	a	b	ab	B	B	A
Actinocorallia	1.30	1.36	1.88	1.20	1.24	1.95	1.52	1.35	2.06	ns.	***	ns.	a	a	a	B	B	A
Actinomadura	7.65	8.65	9.76	5.95	5.88	8.71	7.92	7.77	11.29	***	**	ns.	a	b	a	B	B	A
Aliidongia	5.16	5.59	3.38	6.03	5.69	3.47	4.60	4.88	3.68	ns.	***	ns.	a	a	a	A	A	B
Azospirillum	1.48	1.56	1.43	1.36	1.45	1.33	1.42	1.17	1.47	ns.	ns.	ns.	a	a	a	A	A	A
Bradyrhizobium	10.71	10.58	9.11	8.99	9.25	7.93	9.98	9.44	8.87	*	*	ns.	a	a	b	A	B	AB
Desulfofundulus	3.96	2.28	4.54	2.48	2.28	3.81	2.09	2.58	2.93	ns.	*	ns.	a	a	a	AB	B	A
Dictyobacter	3.11	2.26	3.90	2.30	2.03	3.32	2.66	3.14	2.52	ns.	ns.	ns.	a	a	a	A	A	A
Methylopila	2.22	2.22	1.53	2.47	2.25	1.34	2.18	2.10	1.70	ns.	***	ns.	a	a	a	A	A	B
Mycobacterium	2.67	2.84	2.86	2.63	2.59	2.58	2.82	2.78	2.93	ns.	ns.	ns.	a	a	a	A	A	A
Rhodoplanes	2.66	3.67	1.57	3.19	4.24	1.60	2.73	3.31	1.50	ns.	***	ns.	a	a	a	B	A	C

Skermanella	1.62	1.56	1.02	1.77	1.54	0.91	1.47	1.15	1.01	ns.	***	ns.	a	a	a	A	A	B
Thermosporothrix	3.97	3.12	4.57	2.54	2.65	5.46	3.17	4.54	3.43	ns.	ns.	ns.	a	a	a	A	A	A
Unassigned	28.95	27.90	28.53	28.98	29.25	27.23	29.43	29.83	27.13	ns.	ns.	ns.	a	a	a	A	A	A
Others	23.17	23.82	23.14	27.00	26.72	27.72	25.76	22.94	25.75	***	ns.	ns.	c	a	b	A	A	A
Bacteria_Order-level																		
Acidimicrobiales	1.82	1.98	2.01	1.69	1.82	1.82	1.57	1.82	1.96	ns.	ns.	ns.	a	a	a	A	A	A
Bacillales	5.01	6.04	4.8	3.78	4.63	4.61	4.11	5.14	4.29	*	*	ns.	b	a	ab	A	B	B
Burkholderiales	0.42	0.5	0.19	4.16	3.88	1.05	2.9	1.25	1.32	**	***	*	a	a	b	C	A	B
Chromatiales	2.89	1.94	1.51	2.59	1.99	1.82	2.93	1.47	1.79	***	ns.	ns.	a	b	b	A	A	A
Clostridiales	10.58	8.9	11.65	8.79	9.03	11.2	9.42	9	9.27	ns.	ns.	ns.	a	a	a	A	A	A
Ktedonobacterales	7.09	5.42	8.49	4.85	4.72	8.78	5.84	7.69	5.96	ns.	ns.	ns.	a	a	a	A	A	A
Mycobacteriales	2.79	2.91	2.91	2.79	2.75	2.65	2.95	2.9	2.99	ns.	ns.	ns.	a	a	a	A	A	A
Myxococcales	2.06	1.7	1.49	2.54	2.47	1.46	2.08	2.45	1.1	***	ns.	ns.	a	a	b	A	A	A
Planctomycetales	1.23	0.97	0.94	1.25	1.32	1.06	1.3	1.58	0.66	ns.	ns.	ns.	a	a	a	A	A	A
Rhizobiales	17.83	20.28	14.48	19.78	20.78	13.05	18.75	19.13	15.5	***	ns.	ns.	a	a	b	A	A	A
Rhodospirillales	11.75	12.1	8.56	12.98	11.98	8.52	10.54	9.84	9.06	**	ns.	ns.	a	a	b	A	A	A
Solirubrobacterales	2.51	2.96	2.96	2.48	2.5	2.85	2.36	2.65	3.31	*	ns.	ns.	b	ab	a	A	A	A
Streptosporangiales	12.73	14.33	16.58	10.16	10.17	15.1	13.25	13.13	18.75	***	**	ns.	b	b	a	A	B	A
Unassigned	10.92	9.48	10.42	11.22	10.78	9.72	10.47	11.9	8.7	ns.	ns.	ns.	a	a	a	A	A	A
Xanthomonadales	0.61	1.71	1.01	0.62	1.45	3.23	0.48	0.82	2.04	***	*	**	c	b	a	B	A	B
Others	9.79	8.79	12.01	10.34	9.74	13.09	11.06	9.27	13.33	***	ns.	ns.	b	c	a	A	A	A
Fungi_Phylum-level																		
Ascomycota	45.00	45.33	43.35	24.13	38.95	36.43	23.66	32.83	30.05	***	*	ns.	a	b	b	B	A	A
Basidiomycota	16.25	16.80	16.78	59.00	37.75	41.05	59.85	41.33	48.28	***	*	ns.	b	a	a	A	B	AB
Mortierellomycota	21.78	15.18	11.73	8.40	9.83	7.31	8.79	9.79	5.25	***	***	ns.	a	b	b	C	B	A
Rozellomycota	2.72	4.05	1.66	0.98	2.31	0.84	1.19	2.60	0.94	***	***	ns.	a	b	b	B	A	B
Unassigned	14.00	18.43	26.23	7.36	10.99	14.23	6.43	13.28	15.41	***	***	ns.	a	b	b	A	A	B
Others	0.26	0.22	0.27	0.13	0.17	0.15	0.08	0.18	0.08	**	ns.	ns.	a	b	b	A	A	A

Fungi_Order-level

Agaricales	1.40	4.94	2.36	0.62	2.68	1.01	0.45	2.31	0.82	***	***	ns.	b	a	b	A	B	B
Archaeorhizomycetales	16.48	15.25	12.75	6.68	8.93	6.40	7.65	11.56	7.92	ns.	***	ns.	a	a	a	A	B	B
Chaetothyriales	0.78	1.47	0.83	1.18	2.95	2.60	1.41	2.47	2.54	***	***	ns.	b	a	a	B	A	A
Filobasidiales	1.62	1.07	1.13	5.51	6.05	5.42	4.95	3.90	3.49	ns.	***	ns.	a	a	a	B	A	C
GS11	2.38	3.40	1.31	0.85	1.98	0.65	0.98	2.19	0.70	***	***	ns.	b	a	c	A	B	B
Helotiales	2.92	3.65	3.69	1.29	2.45	2.82	1.42	2.32	2.23	***	***	ns.	b	a	a	A	B	B
Hypocreales	2.38	3.89	2.49	3.31	7.02	4.86	1.62	2.88	2.26	***	***	*	c	a	b	B	A	B
Mortierellales	21.78	15.18	11.73	8.40	9.83	7.31	8.79	9.79	5.25	**	***	*	a	a	b	A	B	B
Sebacinales	10.88	9.04	11.05	49.00	27.43	32.20	52.60	33.95	42.63	*	***	ns.	a	b	ab	B	A	A
Trichosporonales	1.78	1.19	1.51	3.60	1.17	2.11	1.64	0.84	1.12	***	***	**	a	c	b	B	A	B
Unassigned	33.45	35.40	45.55	16.53	24.45	27.68	16.14	24.38	28.18	**	***	ns.	b	a	a	A	B	B
Xylariales	0.71	1.11	1.74	0.88	1.77	3.54	0.49	0.73	0.92	***	***	***	c	b	a	B	A	C
Others	3.48	4.43	3.88	2.16	3.31	3.42	1.85	2.70	1.96	**	***	ns.	b	a	a	A	B	C

Fungi_Genus-level

Apiotrichum	1.75	1.13	1.46	3.55	1.15	2.08	1.62	0.83	1.08	***	***	***	b	a	b	A	C	B
Archaeorhizomyces	16.48	15.25	12.75	6.68	8.93	6.40	7.65	11.56	7.92	***	ns.	ns.	a	b	b	A	A	A
Cladophialophora	0.35	0.49	0.41	0.74	1.54	1.73	0.95	1.58	1.98	***	***	ns.	b	a	a	B	A	A
Microdochium	0.61	0.86	1.52	0.83	1.62	3.43	0.45	0.62	0.84	***	***	***	b	a	c	C	B	A
Mortierella	21.78	15.18	11.73	8.40	9.83	7.31	8.79	9.79	5.25	***	***	*	a	b	b	A	A	B
Oidiodendron	1.62	1.71	1.70	0.44	0.69	0.76	0.59	0.92	0.76	***	ns.	ns.	a	b	b	A	A	A
Paecilomyces	0.55	1.33	0.56	1.39	3.76	2.30	0.54	1.22	0.66	***	***	**	b	a	b	B	A	B
Sebacina	10.74	8.78	10.90	48.95	27.28	32.15	52.50	33.85	42.53	*	***	ns.	b	a	a	AB	B	A
Solicoccozyma	1.62	1.07	1.13	5.51	6.05	5.42	4.95	3.90	3.49	***	ns.	ns.	c	a	b	A	A	A
Unassigned	40.50	47.50	52.28	20.20	32.18	32.90	19.34	31.73	32.03	***	***	ns.	a	b	b	B	A	A
others	4.01	6.71	5.58	3.31	6.98	5.53	2.62	4.01	3.47	***	***	ns.	a	a	b	C	A	B