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Effect of Two Exogenous Organic Acids on the Excitation Effect of Soil Organic Carbon in Beijing, China

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Abstract: Significance: The study of the effects and pathways of catechol and pyrogallic acid on soil organic carbon mineralization has a positive effect on mastering soil carbon transformation. Methods and objectives: In this study, we took 0–20 cm soil from *Pinus tabulaeformis* forest as an object to investigate the effects of catechol and pyrogallic acid with different concentrations on soil organic carbon mineralization through a 60-day mineralization incubation test. The soil active carbon content and changes in soil microbial diversity and community composition were analyzed by using single exponential fitting, quantitative PCR, and high-throughput sequencing to explore the influencing mechanisms of catechol and pyrogallic acid on soil organic carbon excitation. Results: Catechol and pyrogallic acid had the effect of enhancing the soil organic carbon mineralization and soil active carbon content, and the higher the concentration, the stronger the enhancement effect. Catechol reduced the Ace index, Chao1 index, and Shannon index of bacteria and fungi, and further changed the relative abundance of two dominant groups (*Proteobacteria* and *Acidobacteriota*) in bacteria and *Basidiomycota* in fungi at high concentrations. There was no obvious regularity in the effects of pyrogallic acid on bacteria and fungi, but the Ace index and Chao1 index of bacteria underwent drastic and disordered changes. Conclusions: Catechol and pyrogallic acid can trigger positive excitation of the soil organic carbon through two pathways: increasing the soil active carbon content and modulating soil microorganisms, but the way they modulate soil microorganisms are different. Catechol regulates soil microorganisms by reducing the number, richness, and evenness of the bacteria and fungi species, as well as the community composition, while the way pyrogallic acid regulates only closely relates to the changes in the number, richness, and evenness of bacteria species.

Keywords: soil organic carbon excitation; catechol; pyrogallic acid; soil microorganisms

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1. Introduction

Soil is the largest pool of organic carbon in terrestrial ecosystems. The research on the input and output of soil organic carbon plays a positive role in understanding and grasping the carbon balance and carbon cycle of ecosystems. Soil organic carbon mineralization is an important method of carbon output of the soil organic carbon pool, and also an important factor affecting regional carbon balance and the carbon cycle. Therefore, some researchers have carried out in-depth studies on this issue in various aspects and have made good progress [1–4], among which the excitation effect of soil organic carbon is one of the important research results.

It has been found that organic acids are important control factors for soil organic carbon excitation. Some organic acids can regulate soil organic matter decomposition through abiotic pathways, such as complexation, solubilization of organic carbon in the

activated mineral-bound state, promotion of cellulose depolymerization, and participation in structural rearrangement of humic materials [5–7], but few studies have been conducted. Many studies have focused on biotic pathway regulation, exploring the interrelationship between organic acid–soil microbial–soil organic matter excitation. For example, a study found that a phenolic acid mixture of protocatechuic acid, *p*-coumaric acid, *p*-hydroxybenzoic acid, ferulic acid, and catechol as additional carbon sources can activate microorganisms to trigger positive excitation [8]. Low concentrations of 2,4-di-tert-butylphenol and coumaric acid as exogenous carbon sources can produce selective stimulatory effects on fungi, altering the structural diversity of fungal communities and causing positive excitation effects [9]. However, high concentrations can inhibit microbial activity and growth, leading to negative excitation [9]. High concentrations of benzoic acid, ferulic acid, vanillin, and *p*-hydroxybenzoic acid mixtures may inhibit soil microbial activity and reduce soil organic carbon mineralization processes due to their own biotoxicity or by causing a low soil pH [10].

It has been proven that organic acids can induce excitation effects and control the direction and intensity of excitation by affecting soil microorganisms [11–15], but some findings lack deeper explanations. One study argued that low concentrations of 2,4-di-tert-butylphenol and vanillic acid can alter the structural diversity of fungal communities, but have little effect on the relative abundance of bacteria [9], but another study suggested that the addition of oxalic acid can alter the community composition of bacterial flora [5]. Bengtson et al. [16] and Rouatt et al. [13] argued that root exudates containing high levels of organic acids induce excitation effects by altering microbial populations, inducing an excitation effect. In contrast, Graaff et al. [17] and Cheng et al. [18] suggested that root exudates only enhance soil organic carbon turnover by stimulating microbial activity. Therefore, more extensive research is needed regarding the effect of organic acids on the excitation effect and the pathways regulating microorganisms.

Pinus tabulaeformis is one of the main tree species for afforestation in northern China, with an area of 161 × 104 hm^2. Its soil organic carbon content and dynamics are important factors affecting the soil carbon cycle. Catecheol and pyrogallic acid are the main components of root exudates of *Pinus tabulaeformis*. However, the influencing mechanism, regulation pathway, and action intensity of catecheolic acid and pyrogallic acid on soil organic carbon excitation in *Pinus tabulaeformis* forests are still unclear, which restricts the understanding and grasp of the soil organic carbon cycle in the *Pinus tabulaeformis* forest. Thus, in this study, we took 0–20 cm soil from *Pinus tabulaeformis* forest as an object to investigate the effects of catechol and pyrogallic acid, with different concentrations on the soil organic carbon excitation through indoor culture and a single index fitting method, and analyzed the changes in soil microbial quantity, abundance, and community structure, as well as the association between excitation effects and soil microorganisms by high-throughput sequencing to provide a basis for exploring the mechanisms driving the evolution of soil organic carbon in *Pinus tabulaeformis* forests.

2. Materials and Methods

2.1. Soil Sample Collection and Processing

Three 20 m × 20 m standard plots were set in an artificial pine forest in Beijing Songshan National Nature Reserve, and 0–20 cm mixed soil samples were collected according to the "S" sampling method. After air-drying, the samples were sieved through 2 mm sieve and used for an organic carbon mineralization culture experiment.

2.2. Indoor Analysis

2.2.1. Soil CO_2 Cumulative Release Amount Determination Method

First, 10 mL of 0 mol/L, 0.005 mol/L, 0.01 mol/L, 0.1 mol/L, 0.5, and 1 mol/L catechol and pyrogallic acid solutions were added into a culture flask containing 100 g soil, respectively, and the soil CO_2 cumulative release amount was determined by the soil indoor culture method and alkali absorption method [19].

2.2.2. Soil Microbial Diversity Determination Method

Soil samples were collected from mineralization bottles on the 30th and 60th day of the mineralization culture to determine the soil microbial diversity and community composition.

Soil microbial DNA samples were extracted by the Fast DNA SPIN (MP Biomedicals, Santa Ana, CA, USA) method, the DNA mass was extracted using 2% agarose gel electrophoresis, and the DNA was quantified by NanoDrop 2000 ultra-micro spectrophotometer (Themo Fisher Scientific, Inc., Waltham, MA, USA). Quantification was performed. The bacterial genome V3-V4 region was amplified using primer 338F (5′-ACTCCTACGGGAGGCAGCAG-3′) and reverse primer 806R (5′-GGACTACHVGGGTWTCTAAT-3′) was amplified, and the fungal ITS1F region was amplified using the forward primer (5′-CTTGGTCATTTAGAGGAAGTAA-3′) and the reverse primer ITS2R (5′-GCTGCGTTCTTCATCGATGC-3′), which were processed for amplification, and the amplification products were recovered and purified, fluorescently quantified, and sequenced on the Illumina MiSeq platform. QIIME3 (v1.9.1, http://qiime.org/install/index.html (accessed on 21 December 2020)) was applied to determine the valid sequences for identification. The most abundant sequences in OTU were selected as representative sequences, and BLAST was applied to classify the representative sequences. Bacteria and fungi were identified using the GreenGenes database (v135, http://greengenes.secondgenome.com/ (accessed on 21 December 2020)) and UNITE database (v8.0, https://unite.ut.ee/ (accessed on 21 December 2020)), respectively. Community diversity indicators of bacteria and fungi included the Ace index, Chao1 index, and Shannon index.

2.3. Fitting the Equations for the Soil Organic Carbon Mineralization Process

The process of soil organic carbon mineralization was fitted using Stanford and Smith's first-order kinetic equation [20]:

$$C_{min} = C_0 (1 - e^{-kt}) \quad (1)$$

where C_{min} means the cumulative release of soil CO_2 after t time (g/kg), C_0 means the soil active carbon content (g/kg), and k means the active carbon turnover rate (d^{-1}).

2.4. Data Analysis

Excel 2007 and SPSS 17.0 were used for the statistical analysis of the data. Pearson correlation coefficient was used to evaluate the correlation between CO_2 cumulative release and the soil microbial diversity index. The process of soil organic carbon mineralization was fitted by Origin 8.6 software.

3. Results

3.1. Effect of Organic Acids on Soil Organic Carbon Mineralization

The mono-exponential model fit the mineralization curve of the soil organic carbon well and was used to calculate the soil active carbon content [21–24]. The R^2 of the fitting results in this study reached 0.969–0.995, which was a good fit (Figure 1).

The cumulative CO_2 release of both organic acids showed a trend of rapid increase at the early stage and a moderate change at the later stage, and an increase with the increasing concentration and incubation time. At the early stage of mineralization incubation (1–6 days), the cumulative CO_2 release at low concentrations (0.005–0.01 mol/L) showed an increasing trend, but the increase was not significant, and thereafter, there was a significant concentration change. However, at the early stage of mineralization at high concentrations (0.1–1.0 mol/L), the cumulative CO_2 release was already significantly higher than that of the no-addition and low concentration treatments.

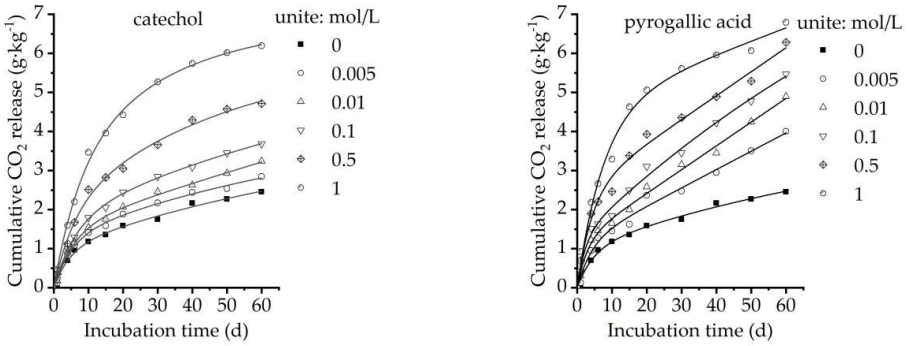


Figure 1. Effect of organic acids on soil organic carbon mineralization.

The cumulative CO_2 release of pyrogallic acid was greater than that of catechol at the same concentration for the same incubation time. The cumulative CO_2 release of pyrogallic acid was 3.26 g/kg, 4.08 g/kg, 4.85 g/kg, 5.99 g/kg, and 6.80 g/kg for each concentration after 60 days of incubation, all higher than that of catechol at the same concentration (Figure 1).

The mineralization rates of the two organic acids showed the same trend. The peak was reached rapidly around 1–5 days of incubation and decreased thereafter. The mineralization rate tended to be the same for all concentrations on day 60, and the mineralization rate increased with the concentration. The effect of pyrogallic acid on the mineralization rate was greater than that of catechol at the same concentration, with peak mineralization rates of 64.60–252.00 g/(kg × d) for pyrogallic acid in the range of 0.005–1.0 mol/L, higher than that of catechol at 59.38–127.20 g/(kg × d) (Figure 2), especially at high concentrations, which could reach more than twice that of catechol.

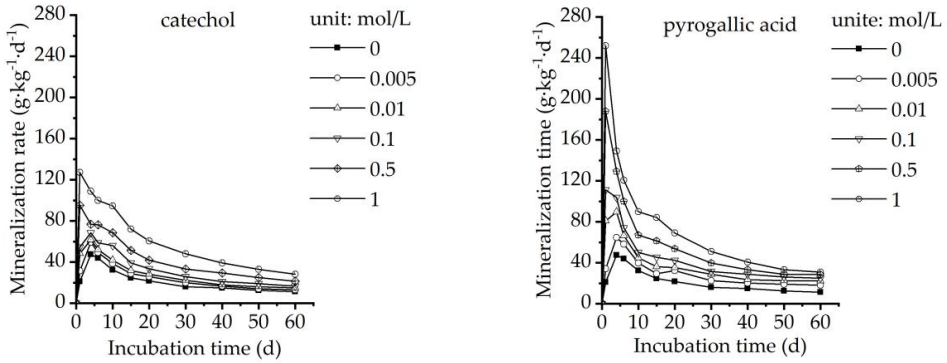


Figure 2. Effect of organic acids on the mineralization rate of the soil organic carbon.

3.2. Effect of Organic Acids on the Active Carbon Content of the Soil

The active carbon content of soil increased with the increasing concentration. The active carbon content of catechol and pyrogallic acid soil increased from 2.35 g/kg (no addition) to 6.12 g/kg and 6.43 g/kg, and both organic acids served to increase the soil active carbon content.

At day 60 of incubation, the ratios of soil active carbon content to cumulative CO_2 release were 0.93–0.99 (catechol) and 0.95–1.11 (pyrogallic acid), respectively, and the active carbon in the soil was basically consumed completely, and microorganisms were forced to start decomposing the refractory organic carbon, the decomposition rate decreased [25], and the mineralization process began to enter a slow decomposition stage (Table 1 and Figure 2).

Table 1. Single exponential fitting equation for the soil mineralization process.

Concentrations (mol/L)	Catechol				Pyrogallic Acid			
	Equations	C_0 (g/kg)	k_0	R^2	Equations	C_0 (g/kg)	k_0	R^2
0	$Y = 2.35\ (1 - e^{-0.063x})$	2.35	0.063	0.969	$Y = 2.35\ (1 - e^{-0.063x})$	2.35	0.063	0.969
0.005	$Y = 2.66\ (1 - e^{-0.069x})$	2.66	0.069	0.975	$Y = 3.62\ (1 - e^{-0.039x})$	3.62	0.039	0.984
0.01	$Y = 3.05\ (1 - e^{-0.063x})$	3.05	0.063	0.977	$Y = 4.47\ (1 - e^{-0.041x})$	4.47	0.041	0.983
0.1	$Y = 3.58\ (1 - e^{-0.063x})$	3.58	0.063	0.982	$Y = 4.99\ (1 - e^{-0.050x})$	4.99	0.050	0.973
0.5	$Y = 4.65\ (1 - e^{-0.064x})$	4.65	0.064	0.985	$Y = 5.89\ (1 - e^{-0.062x})$	5.89	0.062	0.977
1.0	$Y = 6.12\ (1 - e^{-0.072x})$	6.12	0.072	0.995	$Y = 6.43\ (1 - e^{-0.082x})$	6.43	0.082	0.985

3.3. Effect of Catechol on Soil Microorganisms

The Ace index and Chao1 index of bacteria showed similar trends with the increasing concentration and incubation time. It was shown that, firstly, both indices decreased with increasing concentrations at the same incubation time. The decrease was not significant in the concentration range of 0–0.1 mol/L, but in the range of 0.5–1 mol/L, the decrease was obvious. Secondly, the Ace and Chao1 indices also showed different degrees of decrease with the increasing mineralization time for each concentration, which is 0 mol/L < 0.005–0.1 mol/L < 0.5–1 mol/L. The Shannon index also showed a decreasing trend with increasing concentration, but the effect of mineralization time was not strong (Table 2).

Table 2. Effect of catechol on bacterial diversity.

Concentrations (mol/L)	Ace			Chao1			Shannon		
	30 d	60 d	Differences	30 d	60 d	Differences	30 d	60 d	Differences
0	3014.88	2980.23	−34.65	2966.15	2950.20	−15.96	6.39	6.38	−0.02
0.005	3018.22	2873.40	−144.82	3009.06	2839.56	−169.50	6.34	6.18	−0.16
0.01	2936.62	2751.88	−184.74	2931.71	2754.11	−177.60	6.31	6.24	−0.07
0.1	2819.87	2671.37	−148.50	2884.99	2636.00	−158.99	5.99	5.66	−0.13
0.5	2264.41	2039.64	−224.78	2657.18	2092.78	−564.40	5.32	5.20	−0.11
1	2476.23	2209.31	−266.92	2610.06	2200.85	−409.21	5.70	5.56	−0.15

Note: The difference is the difference between the indices on day 60 and day 30 of incubation at the same concentration. The same below.

With the increasing concentration and incubation time, the Ace index and Chao1 index of the fungus showed the same changing trend. The changes of Ace index and Chao1 index were not obvious in the concentration range of 0–0.1 mol/L for the same incubation time, but after increasing to 0.5–1 mol/L, a more significant decrease occurred. With the prolongation of mineralization time, both the Ace index and Chao1 index showed a decrease, but the decrease was different for different concentrations: 0–0.1 mol/L < 0.5–1 mol/L. The Shannon index showed a trend of decreasing with increasing concentration, but the pattern of time change was not obvious (Table 3).

Table 3. Effect of catechol on fungal diversity.

Concentrations (mol/L)	Ace			Chao1			Shannon		
	30 d	60 d	Differences	30 d	60 d	Differences	30 d	60 d	Differences
0	537.61	508.54	−29.07	526.86	505.93	−20.93	3.28	3.17	−0.11
0.005	562.19	498.95	−63.24	542.97	490.12	−52.85	3.14	2.42	−0.72
0.01	570.56	539.98	−30.58	565.63	485.41	−80.22	3.30	2.98	−0.32
0.1	527.94	493.98	−33.96	522.01	471.69	−50.32	2.86	2.61	−0.25
0.5	436.42	264.28	−172.14	441.62	242.29	−199.34	2.82	1.97	−0.85
1	401.75	243.61	−158.14	498.88	305.04	−193.83	2.77	2.53	−0.24

A total of 12 bacterial communities were detected (Figures 3 and 4), among which *Actinobacteriota*, *Proteobacteria*, *Acidobacteriota,* and *Chloroflexi* were the dominant groups, with a combined relative abundance of more than 80%. *Gemmatimonadota*, *Myxococcota*, *Firmicutes*, *Bacteroidota*, *Methylomirabilota*, *Verrucomicrobiota*, *Bdellovibrionota,* and *Planctomycetota*, as well as other communities, accounted for about 20%. The relative abundance of *Proteobacteria* was significantly higher at high concentrations than at low concentrations (0–0.1 mol/L), while *Acidobacteriota* showed a decrease at high concentrations (Figures 3 and 4).

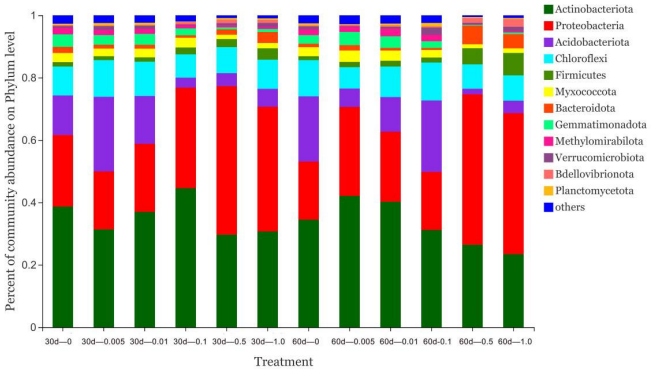


Figure 3. Effect of catechol on bacterial community composition. Note: In the figure, 30 d and 60 d indicate 30 days and 60 days of culture, respectively. 0, 0.005, 0.01, 0.1, 0.5, and 1.0 respectively, indicate the concentration of exogenous acid (in mol/L). The same below.

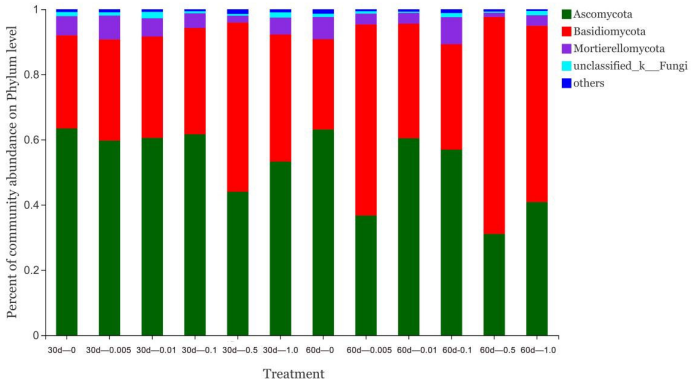


Figure 4. Effect of catechol on fungal community composition.

Three dominant groups of fungi were detected, namely, *Ascomycota*, *Basidiomycota,* and *Mortierellomycota*, as well as unclassified_k__Fungi, which accounted for only about 1%. Among them, the relative abundance of *Basidiomycota* showed a significant increase at high concentrations.

3.4. Effect of Pyrogallic Acid on Soil Microorganisms

The Ace, Chao1, and Shannon indices of the bacteria did not show temporal and concentration variation patterns, but the difference in Ace and Chao1 indices for all five concentrations changed dramatically and disorderly on days 30 and 60 of mineralization compared to no addition (Table 4). The temporal and concentration variations of the three fungal indices were not strong and did not show a clear pattern (Table 5).

Here, 12 and 4 communities of bacterial and fungal were detected (Figures 5 and 6), respectively. Four dominant bacterial communities accounted for more than 83% in total and three dominant fungal communities accounted for more than 98%. The comparison revealed that there was no obvious regular variation in the relative abundance of both bacteria and fungi at different concentrations and different mineralization times.

Table 4. Effect of pyrogallic acid on bacterial diversity.

Concentrations (mol/L)	Ace			Chao1			Shannon		
	30 d	60 d	Differences	30 d	60 d	Differences	30 d	60 d	Differences
0	3174.11	3173.50	−0.61	3074.97	3125.72	50.74	6.442	6.426	−0.016
0.005	562.19	498.95	−63.24	542.97	490.12	−52.85	3.14	2.42	−0.72
0.01	3141.12	3416.40	275.27	3143.80	3462.27	318.47	6.311	6.459	0.148
0.1	3225.59	3105.74	−119.85	3230.44	3082.39	−148.05	6.361	6.365	0.005
0.5	2801.83	3331.09	529.26	2779.22	3331.06	551.84	6.101	6.476	0.375
1	3259.09	2963.23	−295.86	3252.48	3045.26	−207.23	6.276	6.169	−0.106

Table 5. Effect of pyrogallic acid on fungal diversity.

Concentrations (mol/L)	Ace			Chao1			Shannon		
	30 d	60 d	Differences	30 d	60 d	Differences	30 d	60 d	Differences
0	537.71	504.10	−33.61	531.35	512.40	−18.95	3.287	3.181	−0.106
0.005	493.03	471.99	−21.04	479.00	501.11	22.11	2.971	1.344	−1.627
0.01	534.50	524.47	−10.03	522.92	556.33	33.41	3.038	3.043	0.006
0.1	511.95	536.28	24.32	514.74	492.69	−22.04	2.999	2.672	−0.327
0.5	416.10	477.52	61.42	453.25	506.71	53.46	2.814	3.237	0.423
1	449.74	478.46	28.72	430.74	475.06	44.32	2.965	2.980	0.015

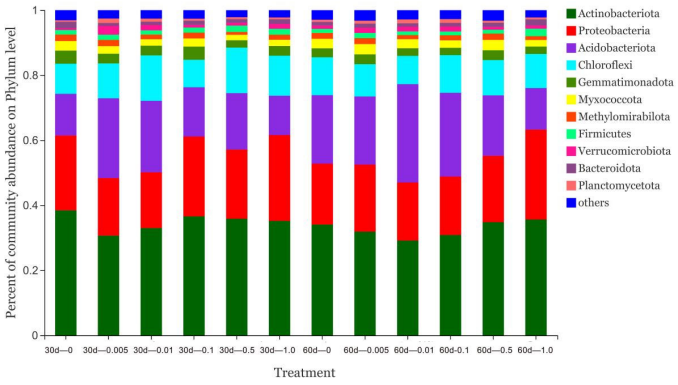


Figure 5. Effect of pyrogallic acid on bacterial community composition.

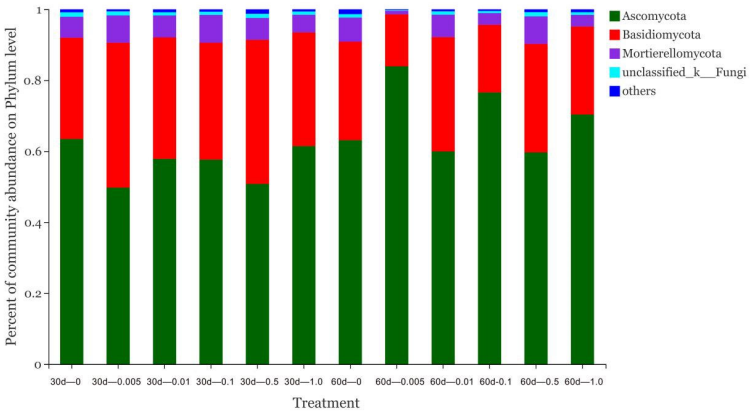


Figure 6. Effect of pyrogallic acid on fungal community composition.

3.5. Correlation Analysis between Cumulative CO_2 Release and Microbial Diversity

Cumulative CO_2 release of catechol soil showed a significant negative correlation with the Ace index, Chao1 index, and Shannon index of bacteria and fungi. Cumulative CO_2

release of pyrogallic acid soil did not reach a significant level with each index of bacteria and fungi, although it showed some correlations (Tables 6 and 7).

Table 6. Correlation analysis between cumulative CO_2 release and soil microbial diversity after catechol addition.

Bacteria	Ace Indices	Chao1 Indices	Shannon Indices
CO_2 accumulative delivery amount (g/kg)	−0.857 **	−0.880 **	−0.772 **
Fungi	**Ace Indices**	**Chao1 Indices**	**Shannon Indices**
CO_2 accumulative delivery amount (g/kg)	−0.889 **	−0.743 **	−0.660 **

Note: ** $p < 0.01$.

Table 7. Correlation analysis between cumulative CO_2 release and soil microbial diversity after pyrogallic acid addition.

Bacteria	Ace Indices	Chao1 Indices	Shannon Indices
CO_2 accumulative delivery amount (g/kg)	−0.062	0.095	−0.281
Fungi	**Ace Indices**	**Chao1 Indices**	**Shannon Indices**
CO_2 accumulative delivery amount (g/kg)	−0.370	−0.362	−0.073

4. Discussion

The two main pathways of soil organic carbon producing positive excitation include the additional input of exogenous substances to the soil carbon source and the regulation of soil microorganisms [26–28]. Both simple and complex carbon compound inputs can increase soil active carbon content directly or indirectly [29–34]. Both catechol and pyrogallic acid exhibited an increase in soil active carbon content from 2.35 g/kg (without addition) to 6.12 g/kg and 6.43 g/kg with increasing concentrations, one of the important factors triggering the organic carbon excitation (Table 1 and Figure 1). However, there was no significant difference in the increase of active carbon content between the two organic acids at the same concentration (Table 1). The molecular structure of the two organic acids was very similar, with only one hydroxyl difference, and the amount of carbon input was very similar at the same concentration and volume. For example, adding 10 mL pyrogallic acid and catechol at a concentration of 0.005 mol/L can input 0.034 gC and 0.037 gC for 1 kg soil, respectively. Due to the similar structural stability and carbon input between the two, there was no significant change in soil active carbon content at the same concentration. Therefore, it could be further inferred that the difference in excitation intensity between the two was mainly caused by the different regulatory pathways of the soil microorganisms.

The overall trends of soil mineralization rates after inputting both organic acids were the same, but the difference in peak value was obvious, especially at high concentrations, where the peak value of pyrogallic acid were more than twice that of catechol (Figure 2). As they had the same effect on the soil active carbon content, the differences in peak mineralization rates reflected the variability of the soil microbial characteristics. The soil pH value had important effects on microbial diversity, activity, and biomass [35,36]; bacteria are suitable to grow in neutral-alkaline soil environments [37]; and soil acidification can be stressful to bacteria, inhibiting its growth and reproduction [38]. The testing soil was slightly acidic, with pH values of 6.96–7.12, and the pH value of catechol and pyrogallic acid aqueous solutions were 6 and 4, respectively. The difference in the effect of the two organic acids on soil pH, causing different responses in soil microorganisms, should be the main reason for the difference in peak mineralization rates.

Catechol with low concentrations exerted regular effects on both bacteria and fungi, promoting soil organic carbon mineralization by reducing the number of species, the richness and evenness of species composition, and community diversity (Tables 2, 3 and 6). In addition to the above effects, organic acids at high concentrations could significantly increase or decrease the relative abundance of some bacterial groups, as shown by an increase in the relative abundance of *Proteobacterias* and *Basidiomycota* of fungi and a decrease in the relative abundance of *Acidobacteriota* (Figures 3 and 4). After 60 days, the *Proteobacteria* and *Basidiomycete* relative soil richness increased by about 26%, and *Acidobacteria* decreased by about 17%. Keiluweit et al. found that oxalic acid addition changed the composition of the bacterial community, with a significant increase in the relative abundance of *Proteobacteria* and *Bacteroid*, and a decrease in the relative abundance of *Acidobacteria*, *Firmicutes,* and *Verrucomicrobia* [5], and the same change was shown in this study for *Proteobacteria* and *Acidobacteriota*. *Proteobacteria* are eutrophic bacteria that can grow rapidly under environmental conditions with a high soil organic matter and nutrient elements [39]. Here, 0.5–1 mol/L catechol resulted in a 2–3 times increase in soil active carbon content compared to no-addition soil, which, combined with the fact that the testing soil was 0–20 cm forest soil and was rich in nutrients, triggered the rapid growth of *Proteobacteria,* leading to rapid carbon mineralization [28]. *Proteobacteria* are nutrient-poor bacteria that are enriched in environments with a low nutrient content and mainly degrade complex organic matter [40], and good soil nutrient conditions and high concentrations of catechol trigger an increase in active carbon content, which becomes a limiting factor for the growth of *Proteobacteria*.

There were no significant regular changes in the effects of pyrogallic acid at different concentrations on the number of species, the richness and evenness of species composition, community diversity, and community composition of bacteria and fungi (Tables 4 and 5, Figures 5 and 6), and no significant correlations with cumulative CO_2 release (Table 7), but there were dramatic and disorderly changes in the Ace and Chao1 indices of bacteria compared to the no-addition treatment (Table 4). Although the changing mechanism is still unclear, pyrogallic acid may promote the mineralization process by altering the soil pH and strongly affecting the number of bacterial species, the richness, and homogeneity of species composition (Figure 2). However, the correlation needs further study.

It has been suggested that some high concentrations of organic acids, which are both carbon sources and biotoxic, such as benzoic acid, ferulic acid, vanillin, and *p*-hydroxy benzoic acid mixtures, for the promotion of soil organic matter decomposition and inhibition mechanisms may exist at the same time, however, their biological toxicity or low soil pH value, the influence of suppress the soil microbial activity, the inhibition of organic carbon decomposition masked the promoting effect of carbon sources, and the mineralization intensity of soil organic carbon decreased [10]. However, in this study, the two high concentrations of organic acids did not show inhibition of the biological toxicity. Although the low pH value of pyrogallic acid may be an important reason for the drastic and disorderly change of bacteria, it did not change the direction of the soil organic carbon mineralization, and the promoting effect was strong.

5. Conclusions

Both catechol and pyrogallic acid can increase the soil active carbon content and modulate the positive excitation effect of the soil microorganisms producing soil organic carbon. The excitation intensity of pyrogallic acid at the same concentration was greater than that of catechol at the same concentration, and the mineralization rate at a high concentration was more than two times that of catechol. The difference in excitation intensity between pyrogallic acid and catechol was mainly caused by the different regulation pathways of the soil microorganisms.

Catechol mainly regulates soil microorganisms by reducing the number of species, the richness and evenness of species composition, and even the community composition of bacteria and fungi. However, the pyrogallic acid regulating pathways are unrelated to

fungi, but may be closely related to the changes in the number of species and richness, and in the evenness of species composition of bacteria.

At high concentrations, neither of the two organic acids showed their own biological toxicity or the inhibition induced by reducing the soil pH, and the soil organic carbon mineralization promoted strongly.

According to the results of this research, we think that the influence of root exudates must be fully considered when researching the dynamic process of soil organic carbon in the future, which will also help guide us to discover a new research idea for forest soil rhizosphere carbon turnover.

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