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Effects of Pruning on Tea Tree Growth, Soil Enzyme Activity and Microbial Diversity

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Abstract: In order to investigate the effect of pruning on the soil environment in which tea trees grow and the growth of tea trees, this study used Wuyi Meizhan (*Camellia sinensis*) as a research object and measured its growth indexes, soil physicochemical indexes, soil enzyme activity and microbial functional diversity to analyze the effects of pruning treatments on the growth of tea trees, soil enzyme activity and soil microbial functional diversity and the correlation between them**.** The results of the analysis of tea tree growth indexes showed that the hundred-bud weight, leaf area and yield in the pruning treatment were significantly higher than those in the unpruned treatment. The results of soil physicochemical index analysis showed that pH, available phosphorus, available potassium and organic matter were significantly higher in the pruning treatment than in the unpruned treatment ($p < 0.05$), while available nitrogen and total phosphorus were significantly lower than in unpruned treatment ($p < 0.05$). The results of soil enzyme activities showed that only polyphenol oxidase and catalase activities were significantly higher in the pruning than in the unpruned treatment, while urease, protease, acid phosphatase, asparaginase and glutaminase activities were significantly lower than in the unpruned treatment ($p < 0.05$). Biolog analysis showed that the utilization of microbial carbon sources, especially amino acid and amine, increased in the rhizosphere soil of the pruned tea tree, while there was a significant decrease ($p < 0.05$) in microbial diversity. It is evident that pruning promoted tea tree growth and some enzyme activity, while inhibiting the activity of enzymes associated with the nitrogen cycle, and the utilization of microbial carbon sources increased, but their diversity decreased. This study provides a theoretical basis for the daily management of tea plantation after pruning.

Keywords: pruning; Wuyi Meizhan; soil enzyme activity; growth index; microbial diversity; Biolog

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1. Introduction

Tea tree pruning is an important management method in the process of tea tree planting. Tea tree pruning can stimulate the growth of lateral buds, and the increase in lateral branches, which in turn can be beneficial for the increase in tea yield. At the same time, tea tree pruning could reduce the labor force and improve the efficiency of tea harvesting [1–3]. In the process of tea tree growth, light pruning generally consists of cutting off 3–5 cm of the green leaf layer on the crown surface of a tea tree, which can promote bud tip germination and reduce leaf pinching; heavy pruning consists of pruning branches with aging trees, more dead branches, diseases and pests, and weak bud-rearing ability, and to cut 1/3–1/2 of the original tree height according to the degree of aging;

generally, the older the trees, the more they are cut off [4,5]. Regardless of the degree of pruning, pruned tea trees need to be subjected to greater physical stress than pruning tea trees due to the large loss of plant biomass [6]. Relevant research showed that pruned treatment led to changes in root secretions of tea trees [7]. Therefore, this study hypothesized that after being subjected to pruning stress, tea trees would produce a series of responses to adapt to pruning stress, and these defense mechanisms could be transmitted to the soil at the roots of tea trees.

Soil enzymes are important biocatalysts in the soil and are involved in all material circulation in the soil [8]. Relevant research showed that the degradation of organic matter and animal and plant residues in soil required the involvement of a large number of soil enzymes, including soil urease, sucrase, acid phosphatase, protease and catalase [9,10]. The pruning of tea trees produced large amounts of litter, the degradation of which required the involvement of a large number of enzymes. At the same time, tea litter led to the accumulation of allelochemicals such as polyphenols, flavonoids, and alkaloids in the soil, so the entire tea plantation contained a large number of secondary metabolites, which in turn affected soil health, soil microbial activity, and biogeochemical cycles [11].

Soil microorganisms play an important role in soil nutrient metabolism and affect soil physicochemical properties [12–14]. Relevant studies have shown that root secretions induced microorganisms to constantly change and adapt to the changing root soil environment [15]. The plant-soil feedback mechanism affected the composition of root secretions and metabolites in the soil, which in turn regulated the structure and function of the soil microbial community. Relevant research showed that soil microorganisms played an important role in promoting tea tree growth, improving tea quality, and protecting tea trees from pathogens [16]. Li et al. [17] showed that soil bacterial diversity tended to decrease with increasing years of tea planting, especially decreasing the relative abundance of some beneficial bacteria. However, there are few reports of changes in rhizosphere soil enzymes and soil microbial communities of perennial plants such as tea trees affected by physical stress from pruning [18,19]. In this study, the effects of pruning on tea tree growth index, soil physicochemical index, soil enzyme activity, and microbial functional diversity were analyzed with Wuyi rock tea Meizhan as the research object, aiming to reveal the effects of pruning stress on the soil environment of the root system of tea trees and provide a theoretical basis for the healthy management of tea plantations.

2. Materials and Methods

2.1. Experimental Site and Soil Sample Collection

The experimental site was located in the area of Foguo Yan in the Wuyi Mountain Scenic Area (117.99° E, 27.72° N) at an altitude of 239 m (Figure 1). The experimental tea cultivar was Meizhan (*Camellia sinensis* cv. Meizhan), with a planting area of about 0.7 ha. The tea plants were nine years old and had not been pruned before. In August 2021, the test area was evenly divided into six areas, three of which had tea trees pruned for treatment (MX), i.e., three replicates, and the other three areas had tea trees unpruned, i.e., controls (MC). Tea trees were pruned by trimming 3–5 cm from the tea tree canopy, and pruning litter was left on the surface of the original tea tree soil. Each year in late October, 700 kg/ha of compound fertilizer (N: P: K = 21:8:16) was applied to tea plantations in each treatment. Other tea plantation management measures, such as weeding and watering, were the same for both pruned and unpruned treatments.

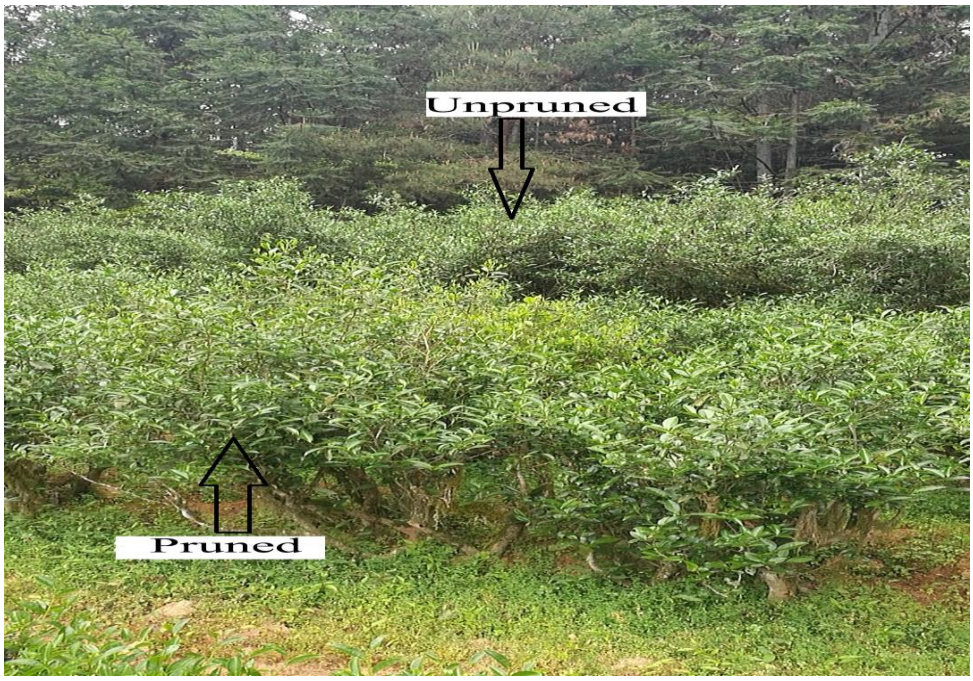

Figure 1. Tea plantations of Meizhan (*Camellia sinensis* cv. Meizhan). The upper part shows unpruned treatment and the lower part shows pruned treatment.

2.2. Determination of the Growth Index of Tea Trees

Growth indexes were measured on 27 April 2022. Leaf area, hundred-bud weight, chlorophyll content, nitrogen content of tea leaves, and yield of pruned and unpruned treatments were measured [20]. Leaf area: 20 mature shoots were randomly selected to measure the leaf length and width, and leaf area was calculated based on length × width × 0.7. One hundred (100)-bud weight: 100 standard bud tips with three leaves were randomly selected, weighed and replicated three times. Chlorophyll and nitrogen content: chlorophyll and leaf nitrogen content of leaves were determined by chlorophyll meter (TYS-N), and the second functional leaf of new shoots was repeated eight times. Tea yields: three rows of tea trees were randomly selected to harvest fresh leaves, respectively [20]. Three leaves of standard standing buds were picked and weighed. Tea yield = tea weight per row/area per row.

2.3. Soil Sample Collection and Determination of Physicochemical Indexes

Soil sampling was conducted on 27 April 2022. Equidistant soil sampling was used to divide the strip planting area into five equal divisions. Five tea trees from each equal division center were taken as one sample, and a total of three soil samples were taken [20]. The roots were dug up near the roots of the tea bushes, and the fine roots were collected at a depth of 5–20 cm of soil. Rhizosphere soil on fine roots was collected by shaking off. Soil samples collected from the pruned region were denoted MX and those collected from the unpruned region were denoted MC. Fresh soil samples were used for enzymatic and microbial functional diversity analyses, while air-dried soil samples were analyzed for soil physicochemical indexes.

Part of the air-dried soil was taken, crushed, screened by 2 mm, and sampled according to the quartering method. Soil pH, total nitrogen (TN), total phosphorus (TP), total potassium (TK), available nitrogen (AN), available phosphorus (AP), available potassium (AK), and organic matter (OM) were determined using the Soil Agricultural Chemical Analysis method [21]. Analyses were repeated three times.

2.4. Determination of Soil Enzyme Activity

The soil enzymes measured in this study mainly include urease, sucrase, polyphenol oxidase, catalase, acid phosphatase protease, asparaginase, glutaminase, and nitrate reductase. The determination methods all refer to the experimental manual of 'Soil and environmental microbiology research method' [22]. Briefly, urease activity was estimated colorimetrically by sodium phenol-sodium hypochlorite. Sucrase activity was determined colorimetrically by nitrosalicylic acid. Polyphenol oxidase activity was determined colorimetrically by pyrogallol. Catalase activity was determined by titration of potassium permanganate. Acid phosphatase activity was determined colorimetrically by disodium phenyl phosphate. The protease activity was determined colorimetrically by ninhydrin. The activities of asparaginase and glutaminase were determined colorimetrically by Nye's reagent. Nitrate reductase activity was determined colorimetrically by phenol disulfonic acid.

2.5. Determination of Functional Diversity of Microorganisms

Soil microbial functional diversity was determined by BIOLOG ECO microplate method [23]. Briefly, 10 g of fresh soil was taken, placed in a 250 mL sterilized conical bottle, filled with 90 mL sterilized normal saline, sealed, shaken at 120 r/min for 10 min and left to stand for 2 min. In an ultra-clean workbench, 5 mL of supernatant was taken and put into 45 mL of sterile water to obtain a 1:100 dilution; the dilution was repeated once to obtain 1:1000 extract to be set aside. One hundred and fifty (150) μL of the diluted soil suspension was transferred to each well, and 150 μL of sterile water was added to wells A1, A5 and A9 as control. After scanning the absorbance at 590 nm on a microplate reader (Nanjing Detie Experimental Equipment Co., Ltd., Nanjin, Jiangsu province, China) (recorded as the absorbance of 0 h), incubation was carried out in the dark at 28°C. The absorbance at 590 nm of each well was then scanned at regular intervals each day for 7 days. Average well color development (AWCD) in BIOLOG ECO plate was used to express the results of ELISA reaction of soil microbial communities. AWCD = $[\sum(C - R)]/31$, where C is the absorbance of 31 carbon source wells and R is the absorbance of the control well. According to the classification of 31 single carbon sources in BIOLOG ECO microplate, carbon sources are divided into six categories: sugar, amino acids, carboxylic acids, amines, polymers and phenols.

2.6. Data Analysis

Excel 2017 software was used for data sorting; IBM SPSS Statistics 21.0 software (Chicago, IL, USA) was used for data T-text and correlation analysis; Excel 2017 and Rstudio 3.3 software (Boston, MA, USA) were used for OPLS-DA simulation, box diagram, heat map and redundancy analysis.

3. Results

3.1. Analysis of Tea Tree Growth Indexes

The results of pruning on tea tree growth index showed (Figure 2) that there was no significant difference between MC and MX in chlorophyll content and leaf nitrogen content. The leaf area and yield of MX were significantly higher than MC ($p < 0.01$), which were 1.32 and 1.84 times higher, respectively. The hundred-bud weight of MX was significantly higher than that of MC ($p < 0.05$), which was 1.40 times higher. Overall, pruning resulted in more vigorous growth and higher tea yields.

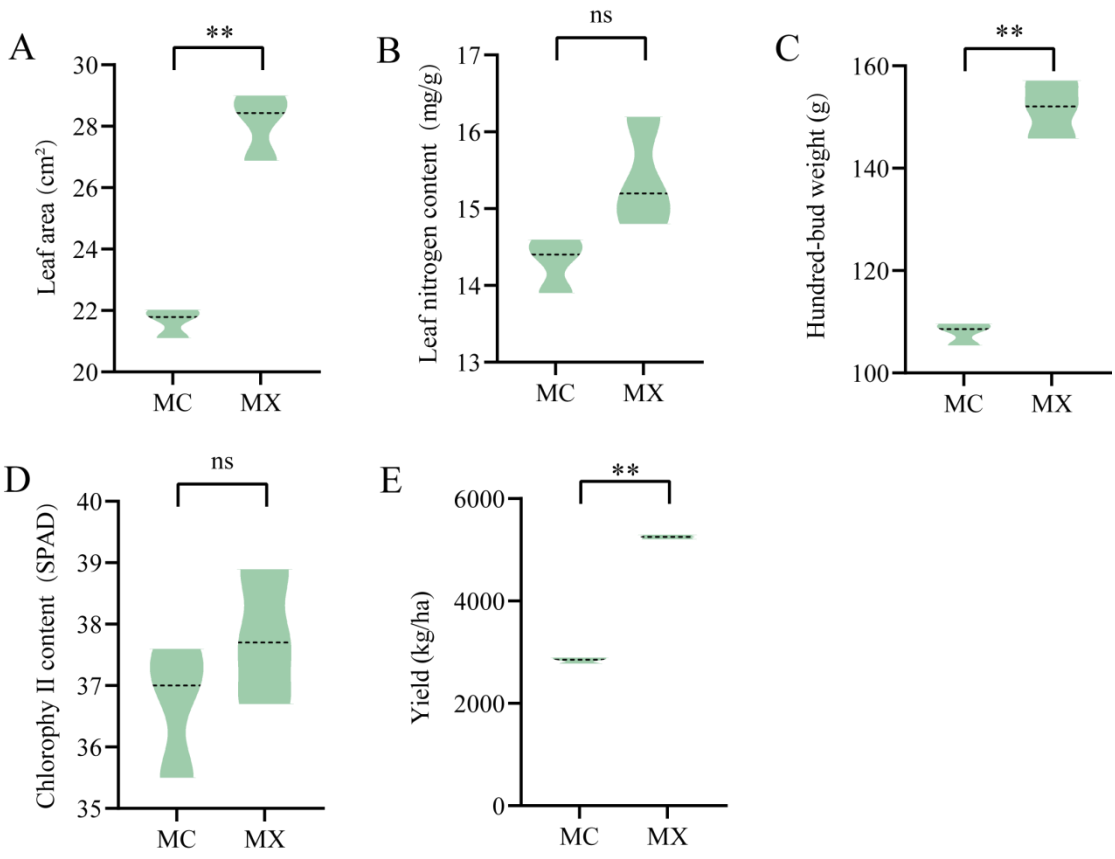

Figure 2. Effect of pruning on growth indexes of Meizhan tea trees**.** MC: Meizhan unpruned; MX: Meizhan pruned; the dotted line is average value; the green delineation shows the range of values; **: significant difference at 0.01 level; ns: not significant. (**A**) Effect of pruning treatment on tea leaf area; (**B**) effect of pruning treatment on tea leaf nitrogen content; (**C**) effect of pruning treatment on tea hundred-bud weight; (**D**) effect of pruning treatment on tea Chlorophy II content; (**E**) effect of pruning treatment on tea yield.

3.2. Analysis of Soil Physicochemical Index

The contents of AN and TP in MX were significantly lower than those in MC ($p < 0.05$), by 6.15% and 29.13%, respectively (Table 1). After pruning, the pH, AP, AK and OM contents in tea soil were significantly higher than those in MC ($p < 0.05$), by 10.94%, 17.22%, 9.74% and 5.65%, respectively. The content of TN and TK in MX was not significantly different from MC. This suggests that pruning could affect the available nutrient cycle in the rhizosphere soil of tea trees, especially N and P.

Table 1. Effect of pruning on soil physicochemical indexes of tea trees ($n = 3$).

Index	MC	MX	Index	MC	MX
pH	4.02 ± 0.01 ^{b}	4.46 ± 0.01 ^{a}	TN (g/kg)	1.15 ± 0.02 ^{a}	1.10 ± 0.04 ^{a}
AN (mg/kg)	122.10 ± 1.03 ^{a}	114.59 ± 1.82 ^{b}	TP (g/kg)	1.03 ± 0.03 ^{a}	0.73 ± 0.02 ^{b}
AP (mg/kg)	13.24 ± 0.48 ^{b}	15.52 ± 0.62 ^{a}	TK (g/kg)	5.62 ± 0.33 ^{a}	5.84 ± 0.47 ^{a}
AK (mg/kg)	101.41 ± 2.81 ^{b}	111.29 ± 1.10 ^{a}	OM (g/kg)	20.01 ± 0.02 ^{b}	21.14 ± 0.09 ^{a}

Note: AN: available nitrogen, AP: available phosphorus, AK: available potassium, TN: total nitrogen, TP: total phosphorus; TK: total potassium, OM: organic matter. MC: Meizhan unpruned, MX: Meizhan pruned, lowercase letter represents the significant difference at the 0.05 level.

3.3. Analysis of Soil Enzyme Activity

Soil enzymes are important for nutrient cycle in soil. The analysis of soil enzyme activity showed (Figure 3) that the activities of polyphenol oxidase and catalase were significantly higher in MX than in MC ($p < 0.05$ or $p < 0.01$), 1.29 and 1.43 times higher, respectively; the activities of acid phosphatase, protease, urease, asparaginase and glutaminase were significantly higher in MC than in MX ($p < 0.05$ or $p < 0.01$), by 1.49, 1.05, 2.47, 1.10 and 1.11 times, respectively. This revealed that pruning could affect the activity of some enzymes in rhizosphere soil of tea trees, particularly those related to the nitrogen cycle.

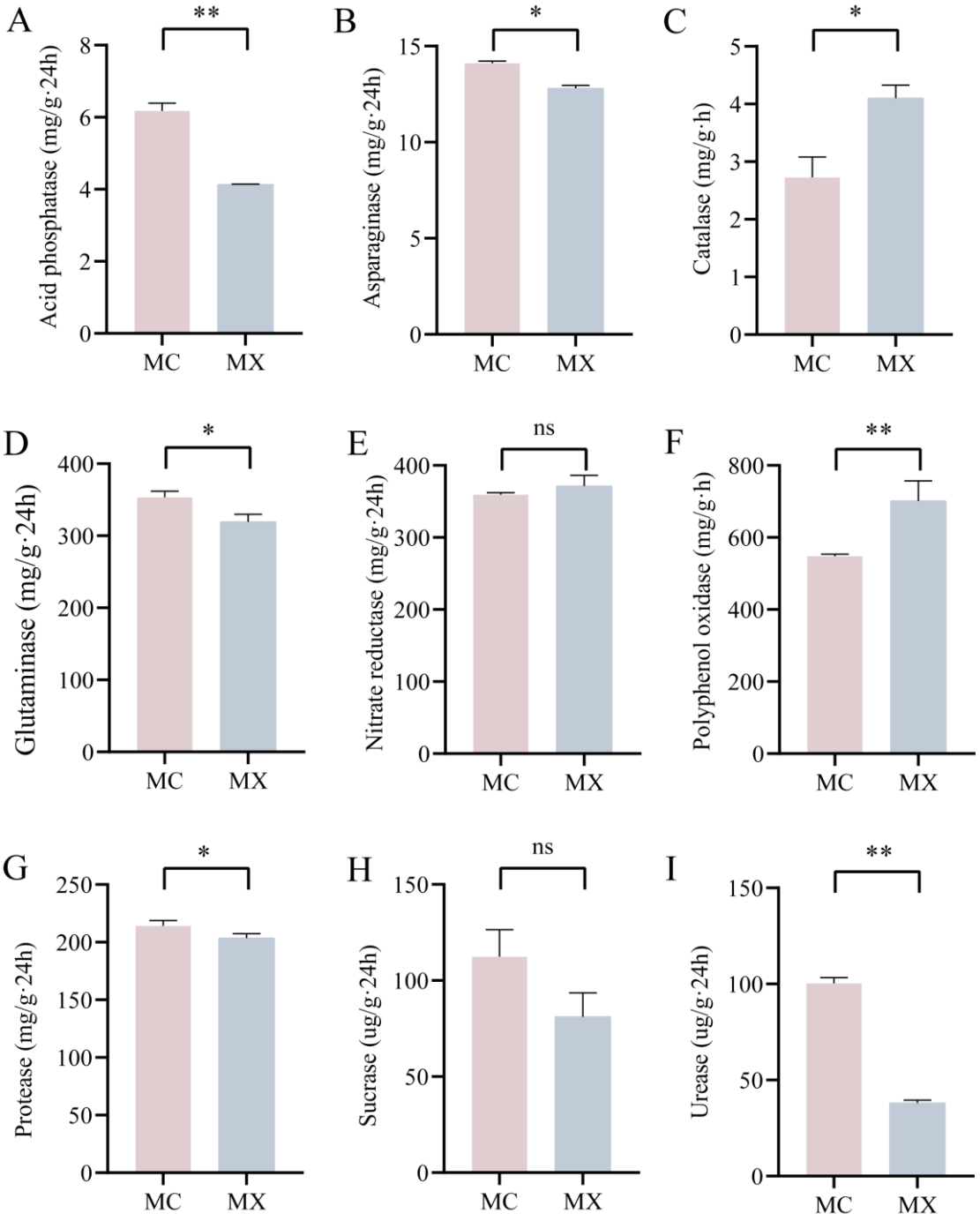

Figure 3. Effect of pruning treatment on soil enzyme activity of Meizhan tea trees (n = 3). MC: Meizhan unpruned; MX: Meizhan pruned; *: significant difference at 0.05 level; **: significant difference at 0.01 level; ns: not significant. (**A**) Effect of pruning treatment on acid phosphatase in

rhizosphere soil of tea trees; (**B**) Effect of pruning treatment on asparaginase in rhizosphere soil of tea trees; (**C**) Effect of pruning treatment on catalase in rhizosphere soil of tea trees; (**D**) Effect of pruning treatment on glutaminase in rhizosphere soil of tea trees; (**E**) Effect of pruning treatment on nitrate reductase in rhizosphere soil of tea trees; (**F**) Effect of pruning treatment on polyphenol oxidase in rhizosphere soil of tea trees; (**G**) Effect of pruning treatment on protease in rhizosphere soil of tea trees; (**H**) Effect of pruning treatment on sucrase in rhizosphere soil of tea trees; (**I**) Effect of pruning treatment on urease in rhizosphere soil of tea trees.

3.4. Correlation Analysis

The correlation results showed that tea tree growth indexes were significantly and positively correlated with soil physicochemical indexes, except for available nitrogen (Figure 4). Tea tree growth indexes were positively correlated with polyphenol oxidase, catalase and nitrate reductase, and negatively correlated with other enzymes. With the exception of total nitrogen and available nitrogen, soil physical and chemical indexes were positively correlated with polyphenol oxidase, catalase and nitrate reductase, but negatively correlated with other enzymes. Total nitrogen and available nitrogen were negatively correlated with polyphenol oxidase, catalase and nitrate reductase, but positively correlated with other enzymes.

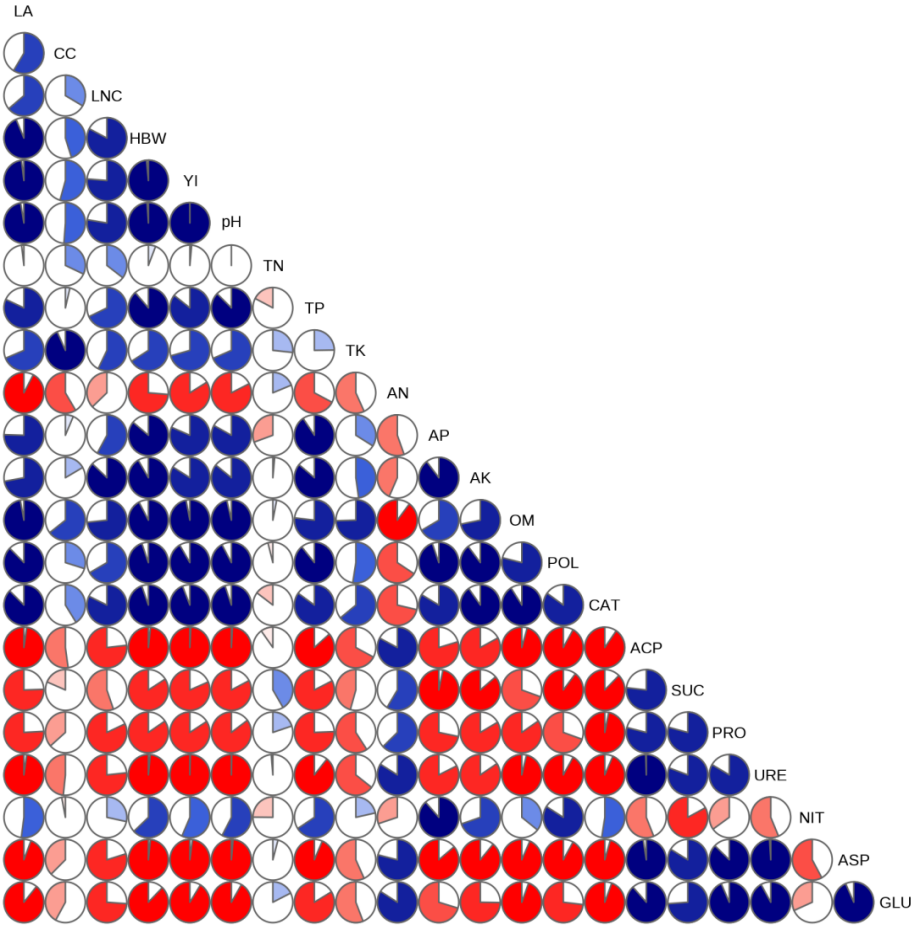

Figure 4. Correlation analysis between growth index, soil physicochemical index and soil enzyme activity. LA: leaf area, CC: chlorophyll content, LNC: leaf nitrogen content, HBW: hundred-bud weight, YI: yields, TN: total nitrogen, TP: total phosphorus, TK: total potassium, AN: available nitrogen, AP: available phosphorus, AK: available potassium, OM: organic matter, POL: polyphenol oxidase, CAT: catalase, ACP: acid phosphatase, SUC: sucrase, PRO: protease, URE: urease, NIT: nitrate reductase, ASP: asparaginase, GLU: glutaminase. Red is negatively correlated and blue is positively correlated. The color area is the correlation coefficient.

3.5. Analysis of Microbial Functional Diversity in Rhizosphere Soil Microorganisms of Tea Trees

The results of carbon source metabolism characteristics analysis of rhizosphere soil microorganisms of tea trees showed (Figure 5) that the degree of carbon source utilization by microorganisms in tea rhizosphere soil increased with the increase in culture time. From 24 h onwards, microbial carbon source utilization in tea rhizosphere soil was higher in MX than in MC.

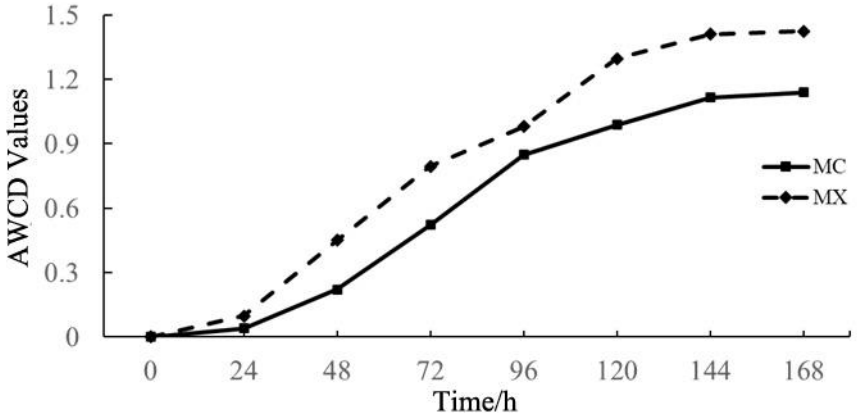

Figure 5. Changes in AWCD values of carbon sources utilized by soil microorganisms. MC: Meizhan unpruned; MX: Meizhan pruned.

The results of the utilization degree of six carbon sources categories showed that (Figure 6) the utilization degree of amino acids and amines by soil microorganisms was significantly higher in MX than in MC ($p < 0.01$), by 1.64 and 2.15 times, respectively, whereas the utilization of carboxylic acids, carbohydrates, polymers and phenols did not differ significantly.

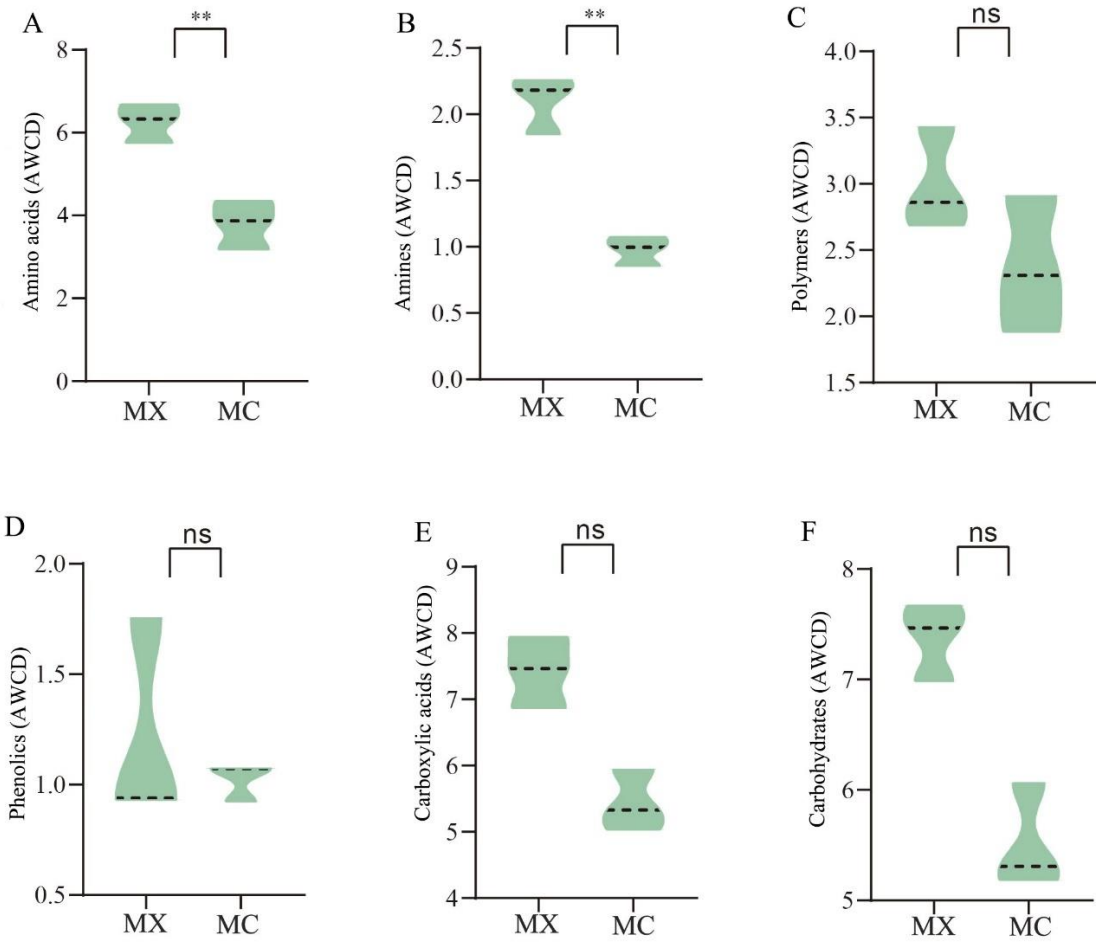

Figure 6. Utilization rate of carbon sources by soil microorganisms**.** MC: Meizhan unpruned; MX: Meizhan pruned; **: significant difference at 0.01 level; ns: not significant; the dotted line is average value; the green delineation shows the range of values; (**A**) Utilization rate of amino acids by soil microorganisms; (**B**) Utilization rate of amines by soil microorganisms; (**C**) Utilization rate of polymers by soil microorganisms; (**D**) Utilization rate of phenols by soil microorganisms; (**E**) Utilization rate of carboxylic acids by soil microorganisms; (**F**) Utilization rate of carbohydrates by soil microorganisms.

The diversity analysis showed (Table 2) that the Simpson diversity index of soil microorganisms was significantly lower in MX than in MC ($p < 0.05$). However, the Shannon diversity index and Chao1 in MX were not significantly different from MC.

Table 2. Microbial diversity index in rhizosphere soil of tea tree.

Treatment	Simpson Index	Shannon Index	Chao 1
MC	0.999 ± 0.001 ^{a}	4.525 ± 0.009 ^{a}	0.913 ± 0.002 ^{a}
MX	0.983 ± 0.001 ^{b}	4.510 ± 0.028 ^{a}	0.910 ± 0.006 ^{a}

Note: MC: Meizhan unpruned; MX: Meizhan pruned; lowercase letter represents a significant difference at 0.05 level.

3.6. Carbon Source Screening for Microbial Differences in Rhizosphere Soil of Tea Trees

OPLS-DA can be used to model the correlation between carbon source utilization and sample type, and to screen key carbon sources that can characterize sample differences by variable importance projection value (VIP value). Meanwhile, in order to test the reliability of the OPLS-DA model, permutation testing was usually used to verify the model, so as to evaluate the accuracy of the model. The results of the analysis of the OPLS-DA model showed that the goodness of fit R^2Y value of the OPLS-DA model for pruned

and unpruned samples was 0.999 ($p < 0.01$), and the predictive Q^2 value for predictability was 0.892 ($p < 0.01$) (Figure 7A). It can be seen that both R^2Y and Q^2 values of the model reached significant levels, and the fit of the model was good and credible for further analysis. The results of the OPLS-DA score map showed (Figure 7B) that OPLS-DA could effectively distinguish pruned and unpruned samples in different areas. In conclusion, there were significant differences in the utilization degree of carbon sources by soil microorganisms between pruned and unpruned treatments. The S-plot analysis showed (Figure 7C,D) that 15 key carbon sources (VIP > 1) distinguished MC and MX samples, among which pruned treatment increased the utilization of 13 carbon sources, and both amino acids and carboxylic acids accounted for more than 30% of the 15 key carbon sources.

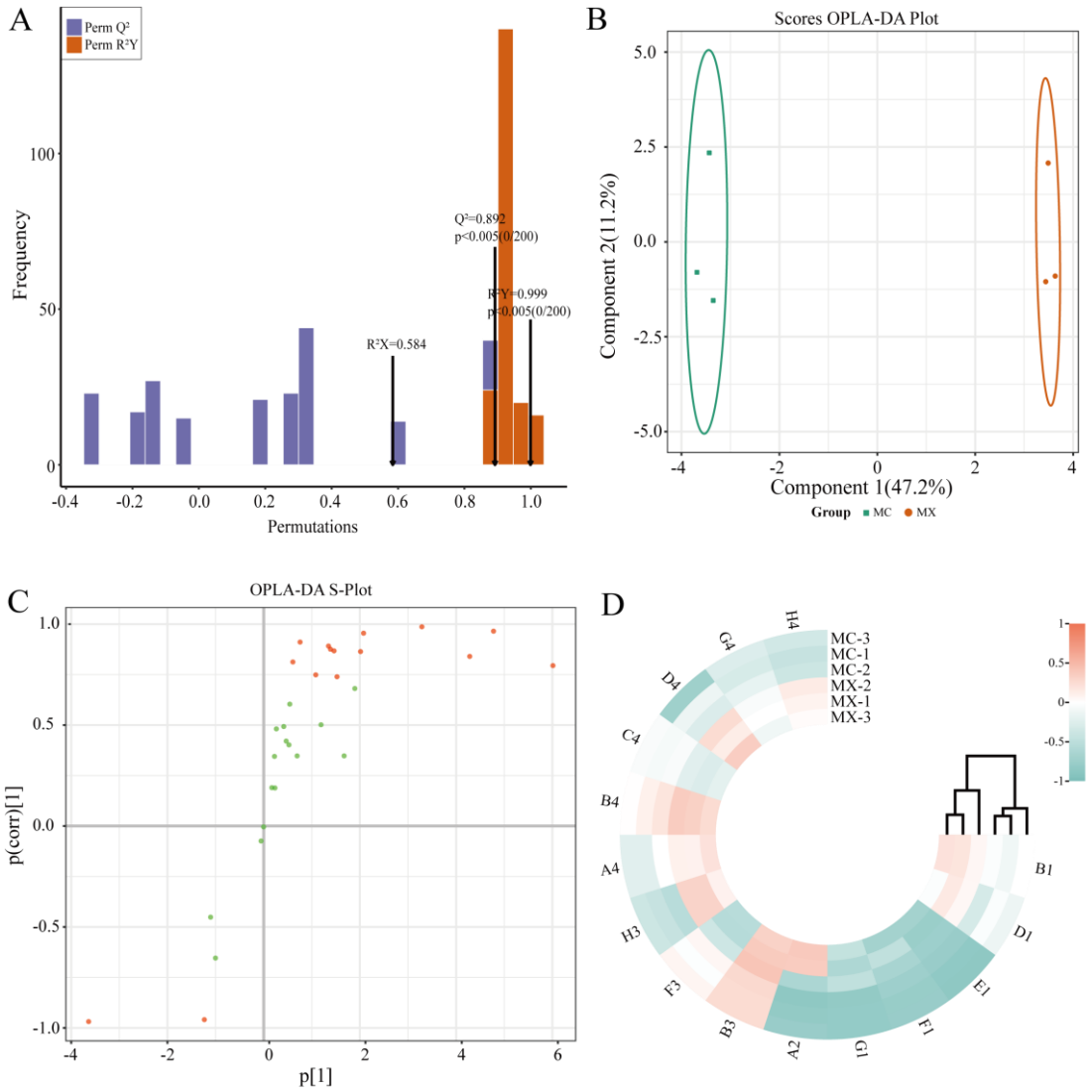

Figure 7. OPLS–DA model and S-plot of soil microorganisms after tea tree pruning. (**A**) OPLS–DA models of the fitting degree test of MX and MC; (**B**) analysis of carbon source utilization of MX and MC by OPLS–DA model; (**C**) OPLS–DA loading diagram for carbon source utilization of MX and MC, the red dots represent different substances of MX, and the green dots represent different substances of MC, p[1] represents the cocorrelation coefficient between the principal component and the index, p(corr)[1] represents the correlation coefficient between the principal component and the index; (**D**) heat maps for carbon source utilization of MX and MC, the red is up-regulated and the green is down-regulated; the darker the color, the greater the utilization rate.

3.7. Redundancy Analysis

The results of redundancy analysis (RDA) showed (Figure 8) that differences in soil microorganisms' use of carbon sources in rhizosphere soil of pruned and unpruned tea trees could be well distinguished using the AWCD 72 h values. RDA analysis of Biolog data and growth indicators showed (Figure 8A) that RDA1 could explain 63.16% of the variation and RDA2 could explain 23.81% of the variation. The carbon source with the highest positive correlation coefficient with RDA1 was β-methyl-D-glucoside, and the carbon source with the highest negative correlation coefficient with principal component 1 was itaconic acid. Further correlation analysis showed that the growth indexes were positively correlated with MX. RDA results of Biolog data and soil enzyme activities showed (Figure 8B) that RDA1 could explain 65.76% of the variation, and RDA2 could explain 22.79% of the variation. The carbon source with the highest positive correlation coefficient with RDA1 was β-methyl-D-glucoside and the carbon source with the highest negative correlation coefficient with RDA1 was itaconic acid. Further correlation analysis showed that polyphenol oxidase, catalase and nitrate reductase were positively correlated with MX, while protease, urease, sucrase, acid phosphatase, asparaginase and glutaminase were positively correlated with MC.

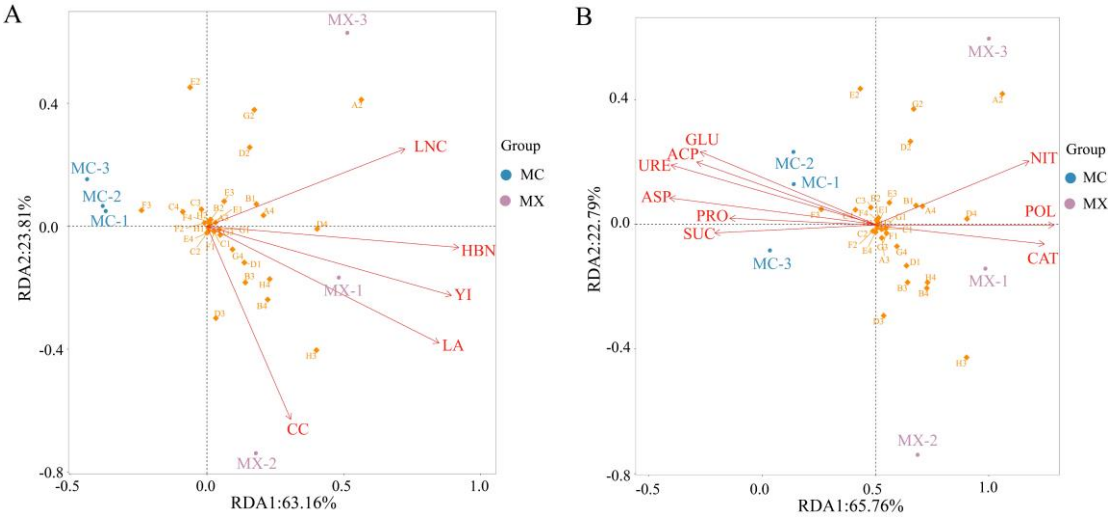

Figure 8. BIOLOG cluster and redundancy analysis of soil microorganisms of tea trees. (**A**) redundancy analysis of BIOLOG data and growth indices; (**B**) redundancy analysis of BIOLOG data and soil enzyme activity; MC: Meizhan unpruned; MX: Meizhan pruned; LA: leaf area, CC: chlorophyll content, LNC: leaf nitrogen content, HBW: hundred-bud weight, YI: yields, POL: polyphenol oxidase, CAT: catalase, ACP: acid phosphatase, SUC: sucrase, PRO: protease, URE: urease, NIT: nitrate reductase, ASP: asparaginase, GLU: glutaminase; 1, 2, 3: the mean with replicate experiments; the dotted line show the horizontal and vertical axes; the red lines show environmental factors.

4. Discussion

Pruning is a cultivation management method that is usually performed as a routine step in tea cultivation to maintain the height of the leaf-picking table, increase the branches of tea trees, and improve tea productivity [24,25]. Zhang et al. [3] reported that summer pruning improved branch growth and tea quality. Yin et al. [26] reported that light pruning after spring could improve the yield and quality of the following spring tea. Lu et al. [5] reported that heavy pruning of tea trees could increase hundred-buds weight and thicker and shorter stems of tea leaves. The results of this study showed (Figure 2) that the hundred-bud weight, leaf area and yield of pruned treatment were significantly higher than those of unpruned treatment, but there were no significant differences in chlorophyll content and leaf nitrogen content. This suggested that tea yield increased by increasing leaf area after pruning.

Plant growth depends on the presence and availability of water and nutrients in the soil [7]. Agricultural management measures and other factors, including abiotic conditions (pH, light, temperature) and biological conditions (varieties, planting years) affected soil physicochemical indexes [27,28]. Tea trees absorb a large amount of nutrients from the environment, especially the soil, during growth. In this study, it was shown that pruning significantly increased soil pH (Table 1). After pruning, the absorption of nitrogen by tea trees increased, resulting in a significant decrease in the nitrogen content of rhizosphere soil. The tea tree was stimulated by exogenous factors after pruning, and absorbed mineral elements efficiently, resulting in increased yield. For example, Mg or Mn could promote the enhancement of photosynthesis in tea trees. Cao et al. [29] indicated that in low-yield and inefficient tea plantations, applying organic fertilizer to tea trees after pruning could improve tea quality. The results suggested that the application of nitrogen fertilizer should be increased after pruning, which could increase the yield of tea.

Soil enzyme activity is a characterization and indicator of its biological condition and fertility [30,31]. Changes in the soil environment can lead to differences in soil enzyme activity. Correlation results showed that polyphenol oxidase and catalase activities were positively correlated with pH, and total phosphorus, available phosphorus, available potassium, and organic matter contents (Figure 4). Pruning increased soil polyphenol oxidase activity and favored the degradation of soil acidic substance and the increase in soil pH. Sun et al. [32] showed that soil pH and nitrogen were the main factors affecting catalase activity. Therefore, the increase in catalase activity after pruning may be due to an increase in soil pH. In this study, tea trees were treated with fertilizer after pruning to provide sufficient nitrogen sources for their growth, when there were sufficient nutrients in the soil and it was the soil environment that tea trees needed to improve in order to facilitate their own growth. Further research found that the increased activity of rhizosphere soil polyphenol oxidase and catalase after pruning provided a good environment for nitrogen uptake by tea trees, ensuring their own growth.

In general, inadequate fertilizer quantity stimulated and increased soil enzyme activity and promoted soil nutrient cycle, while enzyme activity was relatively low when the fertilizer was adequate. In this study, urease, protease, glutaminase and asparaginase activities related to the nitrogen cycle were significantly lower in the pruned treatment than in the unpruned treatment (Figure 3). On the one hand, the reduction in soil available nitrogen content after pruning proved that tea trees have a high demand for nitrogen (Table1), while the content of available nitrogen in the soil was sufficient for the growth of tea trees (up to the Chinese national standard for soil fertility level 1), therefore, the activity of enzymes related to nitrogen cycle in the soil did not need to be increased. On the other hand, Arafat et al. [7] showed that degradation of tea litter led to the accumulation of allelochemicals such as polyphenols, flavonoids and alkaloids in the soil, which might inhibit the activity of these enzymes, as shown by the decrease in enzyme activity after pruning.

Soil microorganisms are highly sensitive to changes in the surrounding environment, and are often considered biological indicators for assessing soil health [33]. Relevant research showed that organic compounds secreted by roots trigger soil microorganisms to constantly change their surroundings [15,34]. Pramanik et al. [18] showed that pruning increased the number of bacteria, fungi and actinomycetes in the rhizosphere soil of tea trees, but reduced the number of denitrifying bacteria. The results of this study showed that Simpson diversity index was significantly lower in pruning treatment than in unpruned treatment, and the efficiency of carbon source utilization by tea rhizosphere microorganisms was higher than that in unpruned treatment, indicating that pruning treatment reduced the functional diversity of tea rhizosphere microorganisms, but enhanced the functions of specific species (Table 2, Figure 6). At the same time, litter degradation due to pruning increased amino acids and carboxylic acids of carbon sources in the soil, and promoted the growth of microorganisms that used the relevant carbon sources (Figure 7). The research of Bora et al. [35] showed that long-term pruning reduced

the richness of microorganisms in the rhizosphere soil of tea trees, while soil polyphenols accumulation promoted the growth and abundance of some microorganisms. This may be the reason pruned treatment reduced rhizosphere microbial diversity, but the utilization rate of carbon source was higher than that of unpruned treatment.

5. Conclusions

In conclusion, the pruned treatment stimulated the growth of tea trees and increased tea yield. The activity of some enzymes in tea rhizosphere soil was inhibited by the effect of tea leaf litter, especially those related to nitrogen metabolism, and microbial diversity was reduced. In order to prevent the accumulation of allelochemicals in the soil and to promote the growth of core microorganisms, the carbon source utilization of soil microorganisms was higher in the pruned treatment than in the unpruned treatment. As tea trees grow vigorously after pruning, they required large amounts of soil nitrogen, but soil enzyme activities related to the nitrogen cycle were affected by allelopathic substances. Therefore, it is recommended to add nitrogen fertilizer after pruning to meet the growth needs of tea trees and increase tea yield. Microbial diversity in the soil is reduced. Tea trees promote the growth of core microorganisms in defense against the accumulation of chemosensitive substances in the soil, resulting in a higher utilization of carbon sources by soil microorganisms between the roots of tea trees in pruning treatment than in unpruned treatment. As a result of pruning, tea trees grow more strongly and therefore require large amounts of soil nitrogen, but soil enzyme activities associated with nitrogen cycling are affected by chemosensitive substances. It is recommended that nitrogen fertilization should be supplemented in time after pruning to meet the growth requirements of tea trees in order to improve tea production.

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