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Effects of Arbuscular Mycorrhizal Fungi and Corn Stover Application on Corn Growth and Nutrient Uptake

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Abstract: Arbuscular mycorrhizal fungi (AMF) are beneficial to crop nutrient absorption. This study investigates the effects of corn stover returned to the field and AMF inoculation, either individually or in combination, on soil fertility, post-harvest corn leaf chlorophyll fluorescence parameters, and nutrients, providing new insights into the ecological function of AMF in controlling crop nutrient acquisition. Compared with AMF-only treatment, AMF+MS (corn stover) inoculation increased the AMF infection rate in corn roots by 11.8% and the number of spores at root surfaces by 16.1%. MS treatment significantly increased the content of available nutrients in the soil and the potassium content in corn plants, but did not significantly affect corn biomass. Inoculation with AMF or MS+AMF significantly improved the contents of available nitrogen, phosphorus, and potassium in the soil, increased the nitrogen, phosphorus, and potassium contents in corn plants, and significantly enhanced corn leaf chlorophyll fluorescence parameters such as Fv/Fm, ΦPSII, ETR, and qP. Inoculation with AMF showed promising potential in increasing soil nutrient content, promoting corn nutrient uptake, and improving corn growth. Although there was no significant interaction between inoculation with AMF and corn stover application, corn stover returned to the field increased the infection rate of AMF in corn roots, suggesting that corn stover returned to the field should be considered when using AMF inoculants. This study highlights the potential value of AMF-corn stover integrated management strategy in promoting sustainable corn cultivation and enhancing agricultural ecosystem services, providing scientific evidence for the green intensification of crop production.

1. Introduction

Corn, as one of the most important food crops globally, its yield and quality improvement have significant implications for food security and promoting agricultural sustainable development. However, over-reliance on chemical fertilizers has led to soil degradation, environmental pollution, and increased costs [1]. In this background, biological technologies and organic material recycling in natural agriculture have attracted widespread attention, among which the application of arbuscular mycorrhizal fungi (AMF) and corn stover returned to the field as two eco-friendly technologies have shown tremendous potential.

AMF significantly enhances the plant's absorption efficiency of difficult-to-obtain nutrients in the soil through symbiotic relationships with plant root systems [2]. The hyphal network formed by AMF not only expands the plant's nutrient absorption range but also promotes the formation of soil aggregates, improves soil physical structure, and facilitates water and air permeability [3]. Moreover, AMF can regulate the plant's immune system and enhance its resistance to diseases and pests.

Corn stover, as agricultural waste, has been burned or discarded in the past, leading to resource waste and environmental pollution. However, using it as organic fertilizer can replenish soil organic matter, promote soil microbial activity, and improve soil structure and water retention capacity [4]. Research shows that organic acids produced by corn stover decomposition can activate fixed phosphorus, potassium, and other elements in the soil, increasing their biological availability [5]. Additionally, corn stover returned to the field can reduce the dependence on chemical fertilizers, lower agricultural production costs, and promote the positive cycle of the agricultural ecosystem. Combining corn stover returned to the field with AMF inoculation can potentially optimize the soil microenvironment and improve the nutritional effectiveness of soil nutrients, ultimately promoting corn healthy growth.

However, despite the significant effects of AMF and corn stover returned to the field on crop growth and soil health, the comprehensive benefits and mechanisms of their combined use are still lacking systematic studies. A few existing studies suggest that AMF and organic material co-application may further enhance plant growth and nutrient absorption efficiency through complementary mechanisms [6], but most of these

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studies focus on specific regions or crops. Therefore, this study uses red soil from Guangzhou, Guangdong Province, as the experimental soil and locally produced corn stover as the research material to conduct pot experiments adding corn stover and inoculating AMF. The study investigates the effects of AMF inoculation and corn stover addition on soil nutrient release and post-harvest corn growth and nutrient acquisition to clarify the ecological function of AMF in controlling plant growth and nutrient acquisition. The expected results will provide scientific evidence for optimizing agricultural management practices and improving corn production sustainability.

2. Materials and Methods

2.1. Test Materials

The test soil is red soil from southern China, which was collected from the tree garden of South China Agricultural University, Guangzhou, Guangdong Province (E113.359917, N23.153562). The soil was dried naturally after passing through a 2 mm screen and being sterilized at 121°C for 2 h. The basic physical and chemical properties of the soil are as follows: pH value is 5.54, organic matter content is 27.12 g/kg, available nitrogen, available phosphorus, and available potassium contents are 30.56, 18.17, and 112.13 mg/kg, respectively.

The test crop is Zea mays L., Guangdong 25 Sweet Corn, which was obtained from the Crop Research Institute of Guangdong Academy of Agricultural Sciences. The seeds with uniform size and full grains were selected, disinfected with 75% ethanol and 10% sodium hypochlorite solution, washed with distilled water, and cultured at 28°C for 3 days until the seeds germinated. Some of the seeds were sown in pots containing quartz sand for seedling cultivation, and water and nutrients were supplemented in time. The seedlings were transplanted into pots when the root system grew to 8-10 cm.

The test AMF inoculant is Glomus mosseae, which was provided by the Crop Research Institute of Guangdong Academy of Agricultural Sciences. The inoculum was obtained by culturing the fungus on corn in the laboratory, and the inoculum contained 20-30 spores per gram.

The corn stover was prepared by selecting corn stover from southern crops, washing it with deionized water, drying it at 75°C to a constant weight, grinding it, and passing it through a 0.5-2.0 mm sieve. Some of the corn stover was used to prepare samples for the nylon bag experiment, and 2.0 g of corn stover was weighed and placed in a nylon bag with a pore size of 75 μm, length of 9 cm, and width of 5 cm. The entire bag was weighed, and the weight was recorded to four decimal places. The total nitrogen, total phosphorus, and total potassium contents of corn stover are 12.5%, 0.96%, and 0.55%, respectively.

2.2. Experimental Design of Pot Cultivation and Inoculation

The experiment consisted of four treatments: control (CK), addition of corn stover (MS), inoculation of AMF (AMF), and addition of corn stover + inoculation of AMF (AMF+MS), with five replicates for each treatment, totaling 20 pots. Each pot contained 3 kg of soil, and the inoculation treatment involved adding 100 g of AMF inoculum to 2.5 kg of sterilized soil in the pot, mixing well, and sowing germinated corn seeds. For the non-inoculation treatment, an equal amount of sterilized AMF inoculum was added, and germinated corn seeds were sown. Finally, 0.5 kg of sterilized soil was added on top of each pot, and three germinated corn seeds were sown in each pot. The corn stover was added at a ratio of 1:20 with soil and mixed thoroughly. No fertilizers or pesticides were used during the experiment, and natural light was used. The soil was watered with deionized water according to the soil moisture content, and the plants were grown for 60 days before the plant and soil samples were collected.

2.3. Sample Collection and Determination

The corn plants were harvested after 65 days of growth, and the plant height was measured with a ruler. The aboveground and root systems of the corn plants were separated, washed with distilled water, and dried. Except for some fresh root samples used for AMF colonization rate determination, the remaining parts were dried at 105°C for 30 minutes and then at 75°C for 72 hours until constant weight. The dried samples were ground and stored for further analysis.

The maximum photochemical efficiency of photosystem II (Fv/Fm), actual photochemical efficiency of photosystem II (ΦpsII), electron transport rate (ETR), photochemical quenching coefficient (qP), and non-photochemical quenching coefficient (NPQ) of corn leaves were determined using a Li-6800 portable photosynthesis system [7].

Soil samples were collected using the shaking method, air-dried, ground, and passed through 1.00 and 0.15 mm sieves for further analysis. Soil pH was determined using a pH meter, total nitrogen was determined using the Kjeldahl method, total phosphorus was determined using the NaOH melting-molybdenum antimony colorimetric method, total potassium was determined using the NaOH melting-flame photometric method, available nitrogen was determined using the alkali diffusion method, available phosphorus was determined using the 0.5 mol/L $NaHCO_3$ extraction-molybdenum antimony colorimetric method, and available potassium was determined using the 1 mol/L NH_4Ac extraction-flame photometric method.

The nitrogen (N), phosphorus (P), and potassium (K) contents of corn stover were determined by weighing 0.1 g of aboveground and underground plant samples, digesting them with H_2SO_4-H_2O_2, and determining the total nitrogen using the Kjeldahl method, total phosphorus using the molybdenum antimony yellow colorimetric method, and total potassium using the flame photometric method.

Some fresh root samples were randomly selected, washed with distilled water, and the AMF colonization rate was determined using the acid fuchsin staining method. Some corn roots were randomly selected, and the soil attached to the roots within 2 mm of the root surface was collected as rhizosphere soil. The spore number in the rhizosphere soil was determined using the wet sieving-centrifugation method [8].

2.4. Data Processing and Analysis

The data were processed using Excel 2010, and the mean and standard deviation were calculated. The data were analyzed using SPSS 23.0 and SNK test at the 0.05 significance level. Two-way ANOVA was used to analyze the significance of inoculation of AMF and addition of corn stover. OriginPro 8 was used to draw charts.

3. Results and Discussion

3.1. AMF colonization characteristics of corn roots

As shown in Figure 1, when corn grew to 50 days, the root colonization rate of the non-inoculated treatment was relatively low, and the number of spores was small. Compared with the control, the addition of straw had no significant effect on the root colonization rate and the number of spores ($P < 0.05$). However, the AMF+MS treatment significantly increased the root colonization rate of corn by 11.8%, and the number of root-soil spores increased significantly by 16.1%. This indicates that inoculation with AMF can establish a symbiotic relationship with corn roots.

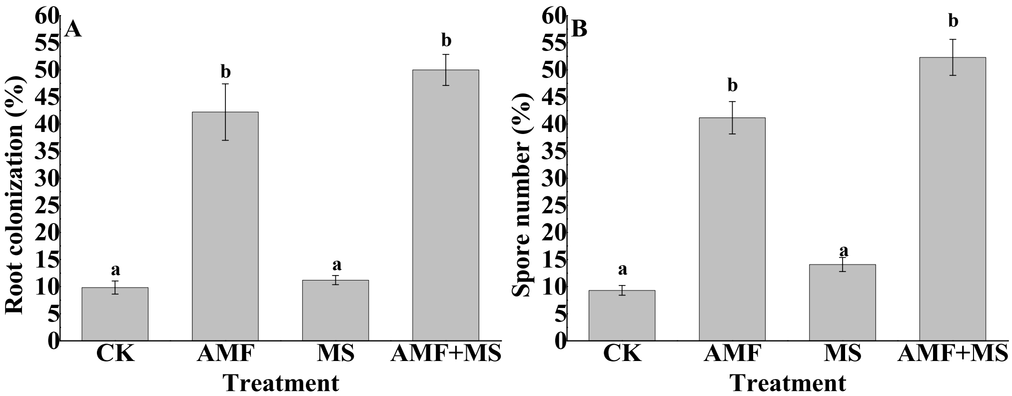


Figure 1. Effect of adding straw and inoculating AMF on AMF colonization characteristics of corn roots (%)
Note:The handling of different lowercase letters shows significant processing differences ($P < 0.05$).

3.2. Effect of adding straw and inoculating AMF on soil pH and available nutrient content

As shown in Table 1, compared with CK, the pH of the soil treated with MS, AMF, and AMF+MS increased significantly by 12.25%, 3.03%, and 7.26%, respectively. The addition of straw and inoculation of AMF significantly affected soil pH ($P < 0.05$). Compared with CK, the increase in available nitrogen, available phosphorus, and available potassium in the soil treated with MS, AMF, and AMF+MS was 1.36%-20.66%, 12.47%-44.99%, and 5.44%-25.77%, respectively. The increase in available nitrogen, available phosphorus, and available potassium in the soil treated with AMF+MS was significantly higher than that of the other treatments. Compared with inoculation of AMF alone, the AMF+MS treatment significantly increased the available phosphorus in the soil by 21.90%, and both were significantly higher than CK and MS treatments. Two-factor analysis showed that inoculation with AMF and the addition of straw significantly affected the available nitrogen, phosphorus, and potassium in the soil, but their interaction was not significant.

Table 1. Effect of adding straw and inoculating AMF on soil pH and available nutrient content

Treatment	pH	available nitrogen	available phosphate	available potassium
CK	6.61 ± 0.01 a	16.17 ± 0.94 a	16.36 ± 3.02 a	72.42 ± 5.02 a
MS	7.42 ± 0.03 c	16.39 ± 2.03 a	18.39 ± 4.03 a	78.47 ± 4.01 b
AMF	6.81 ± 0.04 b	17.53 ± 3.02 b	19.45 ± 3.43 ab	90.51 ± 6.63 c
AMF+MS	7.09 ± 0.03 c	19.51 ± 4.01 c	23.71 ± 4.34 b	93.60 ± 8.71 c

Note:The processing difference between different lowercase letters in the same column is statistically significant ($P < 0.05$).

3.3. Effect of adding straw and inoculating AMF on corn plant height and biomass

As shown in Table 2, compared with CK, the plant height of corn treated with AMF and AMF+MS increased significantly by 40.33% and 45.86%, respectively; the aboveground and belowground biomass increased significantly by 52.07% and 37.33%, 34.84% and 46.15%, respectively. There was no significant difference in plant height and biomass between the AMF and AMF+MS treatments. The inoculation of AMF had an extremely significant effect on the plant height, aboveground and belowground biomass of corn, while the addition of straw had no significant effect on the plant height, aboveground and belowground biomass of corn. Therefore, the good growth of corn seedlings is mainly caused by the inoculation of AMF.

Table 2. Effect of adding straw and inoculating AMF on corn plant height and biomass

Treatment	plant height (cm)	biomass (g/pot)	
		Shoot	Root
CK	26.21 ± 5.21 a	2.17± 2.98 a	2.21 ± 5.21 a
MS	27.98 ± 7.87 a	2.31 ± 3.32 a	2.08 ± 7.87 a
AMF	36.78 ± 5.81 b	3.3± 2.98 b	2.98 ± 5.81 b
AMF+MS	38.23 ± 6.23 b	2.98± 2.98 b	3.23 ± 6.23 b

Note:The processing difference between different lowercase letters in the same column is statistically significant (P < 0.05).

3.4. Effect of adding straw and inoculating AMF on chlorophyll fluorescence parameters of corn leaves

As shown in Table 3, compared with CK, the Fv/Fm, ΦPSII, ETR, and qP of corn leaves treated with MS, AMF, and AMF+MS increased, while NPQ decreased. The increase in Fv/Fm, ΦPSII, and qP of corn leaves treated with AMF+MS was significantly higher than that of other treatments. Compared with the inoculation of AMF alone, the AMF+MS treatment increased the leaf Fv/Fm, ΦPSII, and qP by 12.82%, 4.11%, and 23.81%, respectively, and both were significantly different from the CK treatment (P < 0.05). Compared with the inoculation of AMF alone, the AMF+MS treatment decreased the leaf NPQ by 5.21%.

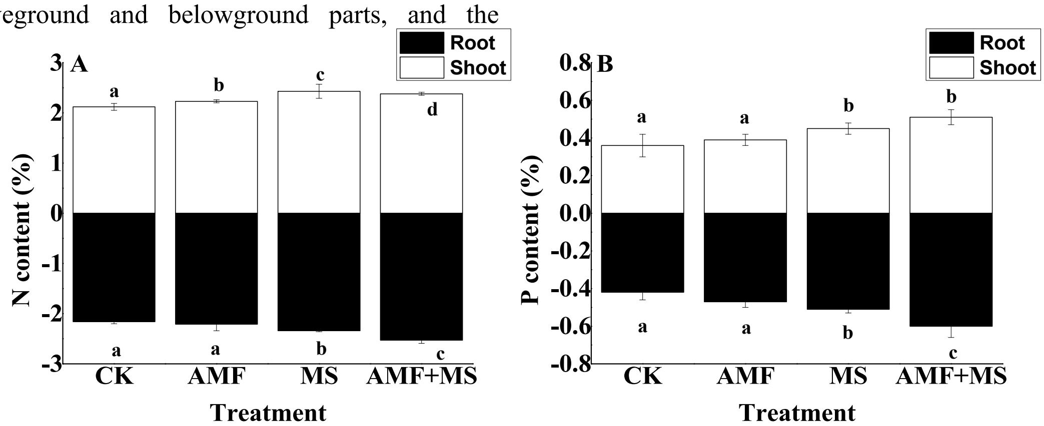
Table 3. Effect of adding straw and inoculating AMF on chlorophyll fluorescence parameters of corn leaves

Treatment	Fv/Fm	ΦPSII	ETR	qP	NPQ
CK	0.64±0.05a	0.51±0.05a	169.24±10.35a	0.24±0.03a	2.61±0.41c
MS	0.71±0.07b	0.65±0.07b	198.31±9.77b	0.33±0.02b	2.24±0.31bc
AMF	0.78±0.09c	0.73±0.04c	223.44±20.26bc	0.42±0.03c	1.91±0.11b
AMF+MS	0.88±0.02d	0.76±0.05d	241.81±9.65c	0.52±0.02d	1.82±0.39a

Note:The processing difference between different lowercase letters in the same column is statistically significant (P < 0.05).

3.5. Effect of adding straw and inoculating AMF on corn nutrient content

As shown in Figure 2, compared with CK, the MS treatment significantly increased the nitrogen and potassium content in the corn aboveground part and the potassium content in the corn belowground part; the AMF and AMF+MS treatments significantly increased the nitrogen, phosphorus, and potassium content in the corn aboveground and belowground parts, and the increase was 9%-29%. The AMF+MS treatment had a better effect than the AMF treatment. The inoculation of AMF had an extremely significant effect on the nitrogen, phosphorus, and potassium content in the corn aboveground and belowground parts, and the addition of straw had a significant or extremely significant effect on the nitrogen content in the corn aboveground part and the nitrogen, phosphorus, and potassium content in the corn belowground part.



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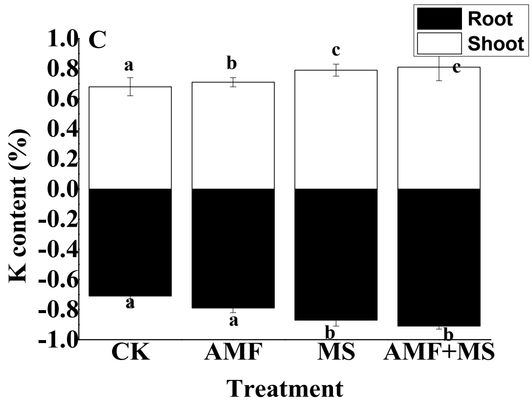

Figure 2. Effect of adding straw and inoculating AMF on corn nutrient content

Note:The handling of different lowercase letters shows significant processing differences ($P < 0.05$).

4. Discussion

Corn stover is rich in nutrients such as nitrogen, phosphorus, and potassium, and is an important nutrient source for promoting crop growth. Therefore, improving the utilization of fertilizer has significant importance for crop growth and development. The results of this study indicate that the addition of corn stover has no significant effect on the stem height and biomass of post-maize, which is consistent with the research results of Li et al. [9]. The observed inhibition of plant growth may be attributed to the excessive production of organic acids during the decomposition of corn stover, which can exert a significant inhibitory effect on plant development by altering soil pH and nutrient availability[10]. Additionally, it may also be due to the differences in the type, application amount, and chemical composition of corn stover [11]. However, the inoculation of AMF can significantly promote corn growth and increase biomass; at the same time, it can significantly increase the content of nutrients in the rhizosphere soil. AMF can infect corn roots, and the increase in the contact area between roots and soil due to the increase in external hyphae reduces the loss of nutrients and improves their utilization efficiency, thereby promoting corn growth.

The combined treatment of adding corn stover and inoculating AMF significantly increases the content of alkali-hydrolyzable nitrogen, available phosphorus, and quick-acting potassium in the rhizosphere soil of corn, which improves the absorption of mineral nutrients by corn and promotes its growth [12]. Prasad et al. [13] also found that the application of corn stover and inoculation of AMF increased soil fertility and promoted the growth of rice. Synergistic interactions between adding corn stover and inoculating arbuscular mycorrhizal fungi (AMF) facilitate the release of nitrogen, phosphorus, and potassium from corn stover, which is then colonized by AMF hyphae. AMF promotes soil enzyme activity, increases microbial biomass and density, and accelerates corn stover decomposition, thereby releasing mineral nutrients into the soil. The transferred nutrients enhance the utilization efficiency of nutrients, improving crop mineral nutrient supply. Specifically, AMF hyphae facilitate the transfer of nitrogen during corn stover decomposition, and promote nutrient absorption by the root system of post-maize crops.

The decomposition of corn stover produces organic matter, which has a significant effect on the extension of AMF hyphae and the increase in the number of spores [14]. The extension of hyphae can expand the absorption area of crops for mineral nutrients in the soil, which is beneficial to improving the growth ability of crops. The improvement of chlorophyll fluorescence parameters in corn leaves reflects the improvement of photosynthetic efficiency. This is not only due to sufficient nutrients in the plant, but also possibly due to the optimization of root structure and the enhancement of function by AMF, which improves the transport efficiency of water and nutrients to the above-ground part, and enhances the synthesis and capture ability of photosynthetic pigments [15].

5. Conclusion

Inoculation of arbuscular mycorrhizal fungi (AMF) in soil has been shown to enhance the availability of essential nitrogen, phosphorus, and potassium nutrients, thereby facilitating the uptake of these nutrients by corn and promoting increased biomass production. The concurrent application of corn stover and AMF inoculation has been found to exhibit a synergistic effect in optimizing mineral nutrient supply and promoting the growth of post-maize crops. This combined approach has significant implications for the restoration and fertilization of soil by AMF and corn stover, and highlights the regulatory role of AMF in modulating crop mineral nutrient supply. The findings suggest that AMF plays a crucial role in optimizing soil fertility and crop productivity, and underscores the importance of considering the interplay between AMF, soil nutrients, and crop growth in agricultural systems.

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