Maize/soybean intercropping improves stability of soil aggregates driven by arbuscular mycorrhizal fungi in a black soil of northeast China

Shu Zhang · Lingbo Meng · Jian Hou · Xiaodan Liu · Abiola O. Ogundeji · Zeyu Cheng · Tengjiao Yin · Nicholas Clarke · Baozhong Hu · Shumin Li

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Abstract

Background and aims It is demonstrated that intercropping improves soil fertility, but its effect on deep soil is still unclear. The major objective of this study was to determine the distribution of arbuscular mycorrhizal fungi (AMF) and soil aggregates and their interrelationship across soil depths in intercropping systems.

Methods A three-year positioning experiment based on a two-factor experimental design at two N application levels (N0 and N2) and different cropping systems (maize/soybean intercropping and corresponding monocultures) was started in 2017. Soil samples were collected from 0–15 cm and 15–30 cm for analyzing soil aggregates and from 0–15 cm, 15–30 cm, 30–5 cm, and 45–60 cm for determining the AMF composition.

Results It was observed that intercropping improved the macro-aggregate (>5 mm) content at 0–15 cm and 15–30 cm depths for maize soil and only 0–15 cm depth for soybean soil without N treatment. The application of N decreased the macro-aggregate content in the intercropping soil at 0–15 cm and 15–30 cm depths. Moreover, intercropping significantly improved the AMF diversity of maize and soybean soils across soil depths, while the application of N reduced the AMF diversity of soil across depths.

Conclusions The structural equation modeling analysis indicated that the intercropping system influenced the stability of soil aggregates and promoted the formation of large aggregates by altering soil nutrients and the diversity of AMF. The results further revealed the reasons behind soil fertility improvement by adopting crop diversification.

Keywords Maize/soybean intercropping · Soil aggregates · Arbuscular mycorrhizal fungi · Soil aggregate stability

L. Meng · B. Hu
School of Geography and Tourism, Harbin University, Harbin, Heilongjiang, China
e-mail: menglingbo2004@126.com

N. Clarke
Norwegian Institute of Bioeconomy Research, P.O. Box 115, N-1431 Aas, Norway
Abbreviations

AMF Arbuscular mycorrhizal fungi
M Monoculture maize
S Monoculture soybean
IM Intercropping maize
IS Intercropping soybean
TN Total nitrogen
AN Available nitrogen
AP Available phosphorus
AK Available potassium
TOC Total organic carbon
MWD Mean weight diameter
GMD Geometric mean diameter
PAD Percentage aggregate destruction
SEM Structural equation modeling

Introduction

Intensive agriculture, as a major method of agriculture, is facing several challenges such as soil degradation and environmental pollution due to the large-scale application of fertilizers, pesticides, and monoculture (Bélanger and Pilling 2019), which is not conducive to sustainable agriculture. Intercropping systems with crop diversity not only increase grain crop yield but also improve soil fertility (Yong et al. 2015; Liang and Shi 2021). Therefore, this is considered to be an important approach to sustainable agriculture. It holds great significance for improving soil fertility and promoting agroecosystem health management.

A maize (Zea mays L.)/soybean (Glycine max L.) intercropping system has been widely practiced for enhancing crop productivity in China. Studies have indicated that the maize/soybean intercropping system can significantly increase the yield of maize and soybean by improving nutrient utilization efficiency (Stoltz and Nadeau 2014). Yong et al. (2015) and Chen et al. (2017) found that soybean intercropping can improve N fixing efficiency and total N accumulation in the soil. Moreover, He et al. (2013) indicated that maize/soybean intercropping also increased plant P uptake. Although the effects of maize/soybean intercropping on nutrient uptake, land/light utilization, and crop yield have been investigated in previous studies, the mechanisms underlying the effect of the maize/soybean intercropping system on soil fertility are unknown.

Soil aggregates, an ensemble of heterogeneous particulates composed of minerals and organic substances, influence soil properties and play an important role in maintaining soil fertility (Li et al. 2019). The high content of macro-aggregates in the soil increases soil carbon sequestration and influences soil nutrient circulation. Soil aggregates can enhance the absorption of minerals and improve the stability of the soil structure by increasing the tightness of the bond between organic carbon and mineral particles (Tan et al. 2017). Rabot et al. (2018) found that soil aggregates play a key role in enhancing soil carbon storage and can retain approximately 90% of organic carbon. Previous studies have confirmed that the cropping systems (e.g., intercropping system) and the agricultural practices (e.g., fertilization) affect soil aggregates. For example, Garland et al. (2017) found that the soil macro-aggregates in maize/pigeon pea intercropping increased by 52% compared to those in the corresponding monocultures (Garland et al. 2017). Šimanský et al. (2019) reported that NPK application slightly decreased the proportion of water-stable macro-aggregates in the soil of the 41-year-old experiment. However, till now, the effects of a maize/soybean intercropping system on the soil aggregates across soil depths at different N application levels remain unclear.

Arbuscular mycorrhizal fungi (AMF), an important component of soil microflora, are essential for enhancing the soil quality, increasing soil microbial diversity, influencing the soil biological environment, and reducing soil erosion (Hamel and Strullu 2006), thus resulting in improved soil ecosystem function. AMF, an important group of plant symbionts, rely on host plants to obtain carbon supply. Meanwhile, AMF can stimulate the growth and reproduction of crops through enhanced absorption of nutrients (P and N) and water uptake from the soil by the mycelium (Pilíarova et al. 2019). These processes can promote the secretion of various organic compounds (e.g., glomalin-related protein) by AMF, which contributes to the soil carbon content. Moreover, previous studies have indicated that AMF can promote the production of root exudates (e.g., extracellular polysaccharides and root mucilage), which can stick soil particles together, thereby leading to the macro-aggregate formation and soil stability (Rillig and Mummey 2006). A study by Miller and Jastrow (1990) has highlighted that AMF may also affect the stability of soil structure by
facilitating the formation of soil aggregates through the interaction between roots and mycorrhizal fungi. Zhang et al. (2019) found that AMF colonization significantly increased the content of macro-aggregates (>2 mm) compared to those without AMF colonization in a karst soil system (Zhang et al. 2019). In addition, Tian et al. (2019) reported that the soil macro-aggregates (>2 mm) in intercropping systems increased by 15.5–58.6%, partly due to an increase in the relative abundance of soil Sordariales (an order of fungi) and enhanced arbuscular mycorrhizal fungal biomass. Generally, the structure and diversity of AMF communities are affected by N fertilizer and cultivation systems (Zhang et al. 2020). Liu (2022) reported that the diversity of AMF in a wheat/soybean intercropping system was higher than that in the soybean monoculture. Moraes et al. (2019) observed that the intercrops of grasses with legumes positively influenced the mycorrhizal colonization and diversity of AMF species. Similarly, a higher AMF diversity and changes in the soil microbial communities in a maize/soybean intercropping system were observed in our previous study (Zhang et al. 2020). In addition to the effects of intercropping, it was observed that different N inputs also influenced the soil AMF diversity in a study, i.e., negatively affecting the diversity and abundance of AMF (Zhang et al. 2021). Ma et al. (2021) reported that the long-term N application significantly decreased the AMF alpha diversity in the rhizosphere. This may be due to the decrease in soluble C distributed in the plant roots with the increase in N, thereby inhibiting the diversity and abundance of AMF in rhizosphere soil, and roots (Schwab et al. 1991). Although the latest research has shown that intercropping systems have significant effects on rhizosphere soil, AMF, and soil aggregates, the changes in the composition and diversity of AMF communities and their dynamic interactions with soil aggregates across soil depths in the intercropping systems under different N application levels are still poorly understood and require further investigation.

To address this knowledge gap, we conducted a three-year positioning experiment with black soil in northeast China to assess the stability of soil aggregates and their relationship with AMF communities and soil properties across soil depths at two N application levels. The major objectives of this study were to (i) assess the distribution and stability of soil aggregates across soil depths, (ii) evaluate the differences in AMF communities of the soil profile (0–60 cm), and (iii) identify the relationship between the AMF communities and the stability of soil aggregates in the maize/soybean intercropping system using a structural equation modeling (SEM) analysis.

**Materials and methods**

**Description of the study site**

The field experiment was conducted in Acheng District, Harbin City, Heilongjiang Province, northeastern China (44°04’N, 125°42’E). The soil type in the region is black soil, which is Mollisol according to the USDA soil classification. The climate in the region is a continental monsoon climate of the cold temperate zone, with an annual mean temperature of 3.4 °C, mean annual precipitation of 553.2 mm, mean 162 frost-free days annually, and mean 2442 h of sunshine annually. Before starting the three-year positioning experiments, the basic soil properties were measured. Briefly, the soil was collected and analyzed in 0–20 cm depth using soil auger from five equidistant points. The average basic soil physical-chemical properties were as following: The soil total organic carbon (TOC) content of the cultivation layer was 17.2 g kg⁻¹, the total nitrogen (TN) was 1.47 g kg⁻¹, the available nitrogen (AN) was 125 mg kg⁻¹, the available phosphorus (AP) was 29.1 mg kg⁻¹, and the available potassium (AK) was 123 mg kg⁻¹. Soil pH was 6.1. The soil texture was 9.8% sand, 64.4% silt, and 25.8% clay.

**Design of the experiment**

In this study, a three-year positioning experiment (i.e., the location, planting pattern, and field management were fixed for each year) started in 2017. A two-factor experimental design based on the N application level and cropping system was considered, and the plots were completely randomly arranged with three replicates. The experimental plots were described in detail in our previous study (Zhang et al. 2020). In this experiment, maize (Xianyu-335) and soybeans (Dongnong-252) were selected as the planting crops. The cropping systems included monoculture maize (M), monoculture soybean (S), and maize/soybean
intercropping (intercropping maize (IM)), intercropping soybean (IS)). Two N application levels (N0 and N2) were applied to all the cropping systems. The application rates of N0 and N2 in the maize cropping systems were 0 kg ha⁻¹ and 240 kg ha⁻¹, respectively, in the maize cropping systems. In the N2 treatment, 80 kg ha⁻¹ urea was applied as the basal N fertilizer, and the remaining 160 kg ha⁻¹ urea was applied in the 12th leaf stage of the maize. In the soybean monocropping and intercropping systems, the application rates of N0 and N2 were 0 kg ha⁻¹ and 80 kg ha⁻¹, respectively. In this case, all the 80 kg ha⁻¹ urea in the N2 treatment was applied as the basal N fertilizer before sowing. Moreover, 52 kg P ha⁻¹ P and 83 kg K ha⁻¹ K were applied as the basal fertilizers in all the treatment plots before the maize and soybean sowing.

The maize/soybean intercropping patterns were two rows of maize intercropped with two rows of soybean. Each plot was of 4.8 m (width) × 10 m (length), and included eight rows with a row spacing of 0.6 m. In the monoculture plots, maize or soybean was grown throughout the eight rows. Two rows of maize intercropped with two rows of soybeans constituted one strip in the intercropping plots, and each intercropping plot contained two strips. After the crop was harvested, the field was plowed. Further, the fields were directly ridged and seeds were sowed in the following spring.

Sampling methods

Undisturbed soil samples were collected at maturity for both maize and soybean crops after harvest in October 2019, which marked the conclusion of the third year of the experiment. The above-ground crop portions were removed after harvesting, and the soils were collected from five equidistant points on each plot using a ring knife for the analyses of soil aggregates, soil nutrients, and AMF. Soil profiles were dug to a depth of 60 cm, and the soil samples were collected from four equidistant points on each plot using a ring knife for the analyses of soil aggregates, soil nutrients, and AMF. Soil profiles were dug to a depth of 60 cm, and the soil samples were collected from four equidistant points on each plot using a ring knife for the analyses of soil aggregates, soil nutrients, and AMF. Soil profiles were dug to a depth of 60 cm, and the soil samples were collected from four equidistant points on each plot using a ring knife for the analyses of soil aggregates, soil nutrients, and AMF. Soil profiles were dug to a depth of 60 cm, and the soil samples were collected from four equidistant points on each plot using a ring knife for the analyses of soil aggregates, soil nutrients, and AMF. Soil profiles were dug to a depth of 60 cm, and the soil samples were collected from four equidistant points on each plot using a ring knife for the analyses of soil aggregates, soil nutrients, and AMF.

Soil aggregate stability was comprehensively evaluated by dry sieving and wet sieving methods given by Guo et al. (2020). Briefly, air-dried soil sample (100 g) was placed on a set of five sieves with diameters of 0.25, 0.5, 1.0, 2.0, and 5 mm. Further, the set of sieves was oscillated for 2 min with 150 cycles min⁻¹, at left and right amplitude of 10 cm, and the weights of different particle size fractions of the dry sieve aggregates were determined. 50 g of a mixed soil sample containing the five sieved fractions in the ratio of each particle size fraction of the dry aggregates was determined. Soil aggregates

Soil nutrient analysis

Soil nutrient analysis included pH; total nitrogen (TN); available N, P, and K (AN, AP, and AK); and total organic carbon (TOC). The soil pH was measured in water using an acidometer (water/soil ratio of 2.5:1, PHS-3C Meter, Toledo). TN was analyzed using the Kjeldahl distillation method (Hou et al. 2007). AP was extracted and determined according to the method given by Bray and Kurtz (1945). The diffusion adsorption method and flame photometry were used to analyze AN and AK, respectively (Lu 2000). TOC of the soil samples was measured using a TOC analyzer (Multi N/C 2000, Germany) following the method of Xiao et al. (2019).

Soil aggregates

In this study, the soil aggregate stability was comprehensively evaluated by dry sieving and wet sieving methods given by Guo et al. (2020). Briefly, air-dried soil sample (100 g) was placed on a set of five sieves with diameters of 0.25, 0.5, 1.0, 2.0, and 5 mm. Further, the set of sieves was oscillated for 2 min with 150 cycles min⁻¹, at left and right amplitude of 10 cm, and the weights of different particle size fractions of the dry sieve aggregates were determined. 50 g of a mixed soil sample containing the five sieved fractions in the ratio of each particle size fraction of the dry aggregates was weighed and placed in the soil aggregate analyzer for measurement based on the method given by Zhou et al. (2020). Furthermore, the soil aggregate stability was analyzed in terms of the mean weight diameter (MWD; mm) and geometric mean diameter (GMD; mm) according to the method given by Okolo et al. (2020). The equations for MWD and GMD are expressed below.

\[
MWD = \sum_{i=1}^{N} M_i \times X_i
\]

\[
GMD = \exp \left[ \sum_{i=1}^{N} M_i \times \ln(X_i) \right]
\]

where \(X_i\) is the average diameter of aggregates \(i\), and \(M_i\) is the mass ratio of aggregate \(i\) (Kemper and Rosenau 1986).

The equation for calculating the percentage aggregate destruction (PAD) was given by:
where PAD is the percentage of aggregate destruction (%), and \( M_d \) and \( M_w \) represent the aggregate mass fractions of the dry and wet sieves with particle sizes > 0.25 mm, respectively.

Composition of AMF communities in the soil profile

Illumina MiSeq sequencing of 18S rRNA gene amplicons was performed to analyze the composition of fungal communities. DNA extraction and sequencing analyses were performed according to the method described by Zhang et al. (2020). Briefly, the V4-V5 hypervariable region of the 18S rRNA gene was amplified using the universal primers AML1F (5’-ATCAACTTTCAGTGAGGATAGA-3’) and AML2R (5’-GAACCCAAACA CTITGGTTTCC-3’). The AMF-specific primers AMV4-5NF (5’-AAG CTGTAAGTGAATTTCG-3’) and AMDGR (5’-CCCACACTCCCTATT AATCAT-3’) were used in the second amplification. Gene sequencing and analysis were performed by Shanghai Majorbio Bioinformatics Technology Co., Ltd., China. Initially, the resulting amplicons were purified and pooled in equimolar ratios for paired-end sequencing on an Illumina MiSeq platform. Then, the sequencing data were processed and merged by FLASH. Finally, OTUs were assigned to different fungal taxa by the basic local alignment search tool (BLAST), with a percentage similarity of 97%. The OTU table was clustered by the rarefied sequences and used to calculate the AMF alpha diversity indices and the relative abundance of every AMF taxon. Moreover, the AMF diversity indices, including Shannon, Simpson, Chao, and ACE, were calculated according to the methods described by Keylock (2005) and Chao and Lee (1992).

Data analysis

First, the normal distribution parameters and homogeneity of variances were used to analyze the effects of monoculture and intercropping on soil fertility and AMF. A three-way (considering N application, cropping system, and soil depth as the three variables) analysis of variance (ANOVA) was used to determine the interaction between the variables that met these assumptions of normality and homogeneity (IBM SPSS 22.0, Chicago, IL, USA). The Duncan test was used to compare the means at 5% and 1% significance levels. The correlations between the indicators in this experiment were analyzed using SPSS 22.0 (Chicago, IL, USA). Principal coordinates analysis (PCoA) was carried out in Rstudio version 3.3.0 using the Bray–Curtis dissimilarity matrices of OTUs at 97% cutoff to reveal the differences in the structure of AMF communities at different treatments. All the figures were plotted using Origin 8.5 (Origin Lab, USA).

The structural equation modeling (SEM) was used to evaluate the multivariate effects of the cultivation system and N application rates on soil nutrients, AMF communities, and soil fertility. Similarly, the direct and indirect interrelationships between AMF communities, soil nutrients, and soil properties were analyzed using SEM analysis. The SEM analysis was conducted as given in the study by Wang et al. (2021). Briefly, before SEM analysis, autoregressive correlation structures in IBM SPSS AMOS 23.0 were used to identify potential correlations. The a priori model was then established based on the previous studies. Finally, the data from this study were fitted to the a priori model using the maximum-likelihood estimation method in AMOS 23.0 software. The chi-square test in the SPSS software was used to verify the quality of the fit. SPSS AMOS 23.0 (SPSS Inc., Chicago, IL, USA) was used to analyze and verify the potential correlations among the indicators based on the maximum-likelihood estimation method.

Results

Distribution of the soil aggregate fractions

This study revealed that the planting patterns and N application levels had a significant influence on the soil aggregate content at two depths (0–15 cm and 15–30 cm) for maize and soybeans \((P \leq 0.05)\). For maize, the soil macro-aggregate content (> 5 mm) in the intercropping system at 0–15 cm soil depth (49% and 39% for N0 and N2, respectively) was significantly higher than those in the corresponding monocultures (25.6% and 27% for N0 and N2, respectively) (Table 1). The macro-aggregate (> 5 mm) content in the intercropping systems at 0–15 cm soil depth was increased by 91% and 45% compared to those in the
Table 1 Relative proportion of soil aggregate fractions in maize monoculture (M) and intercropping (IM) systems at 0–15 cm and 15–30 cm soil depths under two N treatments (N0 and N2)

| Treatments | >5 mm Content (%) | Mean (%) | 2-5 mm Content (%) | Mean (%) | 1-2 mm Content (%) | Mean (%) | 0.5-1 mm Content (%) | Mean (%) | 0.25-0.5 mm Content (%) | Mean (%) | <0.25 mm Content (%) | Mean (%) |
|------------|-------------------|----------|--------------------|----------|--------------------|----------|----------------------|----------|------------------------|----------|------------------------|----------|
| N0_M_15    | 25.62 ± 2.35b3    | 4.53 ± 0.42a | 4.65 ± 1.61a       | 14.49 ± 0.88a | 15.14 ± 4.11a      | 35.58 ± 4.4a |
| N0_IM_15   | 48.88 ± 6.40a     | 37.25A2   | 4.16 ± 1.60a       | 4.35A     | 3.72 ± 1.14a       | 4.19A     | 7.88 ± 2.87b         | 11.19A   | 13.62 ± 3.39a          | 14.38A   | 21.73 ± 4.69b          | 28.66A   |
| N2_M_15    | 26.92 ± 1.89b     | 2.19 ± 0.52b| 4.19 ± 1.98a       | 10.86 ± 1.82a| 24.60 ± 3.28a      | 30.23 ± 2.56a |
| N2_IM_15   | 39.07 ± 9.08a     | 32.99B    | 4.90 ± 0.70a       | 3.55A     | 6.25 ± 1.00a       | 5.22A     | 11.17 ± 2.44a        | 11.52A   | 13.97 ± 4.00b          | 19.29A   | 24.64 ± 4.51a          | 27.44A   |
| N0_M_30    | 2.80 ± 1.54b      | 0.30 ± 0.01a| 2.89 ± 0.57a       | 3.57 ± 2.31a| 22.83 ± 1.00a      | 67.63 ± 2.31a |
| N0_IM_30   | 4.99 ± 8.22a      | 3.85A     | 0.53 ± 0.33a       | 0.42B     | 1.72 ± 0.34a       | 2.31A     | 6.49 ± 1.83a         | 5.03B    | 19.51 ± 2.34a          | 21.17A   | 66.7 ± 10.27a          | 67.21A   |
| N2_M_30    | 0.73 ± 0.84b      | 2.75 ± 1.68a| 3.33 ± 0.75a       | 11.97 ± 3.47a| 21.62 ± 0.74a      | 59.60 ± 4.78a |
| N2_IM_30   | 1.90 ± 2.42a      | 1.32B     | 1.44 ± 0.68a       | 2.10A     | 2.51 ± 0.65a       | 2.92A     | 13.17 ± 0.66a        | 12.57A   | 18.69 ± 2.07b          | 20.16A   | 62.30 ± 2.03a          | 60.95A   |

ANOVA (P value)

| C  | N  | D  | C*N | N*D | C*D | C*N*D |
|----|----|----|-----|-----|-----|-------|
| 0.000 | 0.121 | 0.000 | 0.165 | 0.693 | 0.001 | 0.245 |

1 Different lower-case letters in the same column indicate significant differences between monoculture and intercropping treatments (P≤0.05)
2 Data are the means between monoculture and intercropping treatments followed by different capital letters in the same column indicating significant differences between the two nitrogen levels for the same soil aggregate fraction (P≤0.05)

C: Cropping systems; N: Nitrogen application level; D: Soil depths; C*N: Interaction between cropping systems and the nitrogen application level; N*D: Interaction between the nitrogen application level and soil depths; C*D: Interaction between the cropping systems and soil depths; C*N*D: Interaction among the above three factors
corresponding monocultures for N0 and N2, respectively. A similar trend was observed at 15–30 cm soil depth, and the macro-aggregate (> 5 mm) content in the maize intercropping (IM) system was significantly higher than those in the maize monoculture (M) for N0 and N2. Moreover, the results also highlighted that increasing the N application rate could significantly decrease the macro-aggregate content in the intercropping system at both 0–15 cm and 15–30 cm depths.

When compared with maize, soybean intercropping increased the content of macro-aggregates (> 5 mm) at 0–15 cm soil depth for N0 (Table 2). However, the macro-aggregate (> 5 mm) content at 15–30 cm soil depth for soybean intercropping (IS) was lower than that of the corresponding monoculture. In addition, the N fertilizer level exhibited a significant difference in the aggregate content between soybean monoculture and intercropping soils. In the soil under soybean monoculture (S), an increase in the N application rate increased the macro-aggregate content at 0–15 cm soil depth from 12 to 50%, whereas that at 15–30 cm soil depth decreased from 9.6% to 1.6%. When compared with that in the S system, the macro-aggregate content in intercropping systems at 0–15 cm and 15–30 cm soil depths decreased from 57% and 3.8% to 38% and 1.8%, respectively with increasing N application. Generally, the macro-aggregate (> 5 mm) content in the 0–15 cm layer of the intercropping system was significantly higher than that of the monoculture system.

Soil aggregate stability

The indicators including MWD and GMD were derived based on the aggregate content to assess the stability of the soil structure. The MWD and GMD are indicators that generally reflect the size and distribution of aggregates and are positively correlated with aggregate stability. As shown in Fig. 1, the MWDs and GMDs in the intercropping systems (0–15 cm soil depth) for N0 were significantly higher than those in the two monocultures and followed the order intercropping system > soybean monoculture > maize monoculture. In general, the soil aggregate stability (expressed in terms of MWD and GMD) of the intercropping system was significantly higher than that of the monoculture systems. However, there were no significant differences between the intercropping and monoculture systems at 15–30 cm soil depth. Moreover, compared with N0, N2 significantly increased the MWD and GMD at 0–15 cm soil depth for M and S and reduced the MWD and GMD at 0–15 cm soil depth for IM and IS systems.

In addition, the PAD is an important indicator of soil structure stability. For N0, the PAD at 0–15 cm soil depth in the intercropping system (63% and 60% for IM and IS, respectively) was lower than that in the corresponding monoculture systems (70% and 75% for M and S, respectively). A similar trend was observed at 15–30 cm soil depth for both the monoculture and intercropping systems. In addition, when compared with N0, N2 treatment decreased the PAD of soil at 0–15 cm for the M and S systems but led to an increase in PAD of the soil at 0–15 cm for the intercropping system.

Changes in the AMF diversity at different soil profiles

For N0, the Shannon indices of the IM across soil depths were higher than those of the M by 15%, 8.5%, 1.8%, and 31%, at depths 0–15 cm, 15–30 cm, 30–45 cm, and 45–60 cm, respectively (Fig. 2). However, the AMF diversity in the IM system was lower than that in the M system by 9.1%, 35%, and 9.3%, respectively at 15–30 cm, 30–45 cm, and 45–60 cm soil depths for N2. Moreover, the Shannon indices of the M and IM highlighted different trends after the N application. The Shannon index increased in the M system at 0–15 cm and 15–30 cm soil depths, while the Shannon index decreased at 30–45 cm and 45–60 cm soil depths with N application. At the same time, the Shannon index in the IM for N0 was higher than that after N application, i.e., for N2. However, the changes in Simpson and Shannon indices revealed opposite trends in the maize soil (Table 3).

When compared with the maize soil, the Shannon index of the soybean soil was affected by the cropping system and N application, as well as by the interaction of these two factors (Fig. 3). The Shannon index of each layer of IS was significantly higher \((P \leq 0.01)\) when compared with that of the S system for N0 and N2. This result indicated that the AMF diversity in each layer of the IS system was higher than that of the S system, e.g., for N0, the AMF diversity in each intercropped layer was increased by 19%, 46%, 74%, and 46% at depths 0–15 cm, 15–30 cm, 30–45 cm,
Table 2 Relative proportion of soil aggregate fractions in soybean monoculture (S) and intercropping (IS) systems at 0–15 cm and 15–30 cm soil depths under two N treatments (N0 and N2)

| Treatments | > 5 mm Content (%) | Mean (%) | 2–5 mm Content (%) | Mean (%) | 1–2 mm Content (%) | Mean (%) | 0.5–1 mm Content (%) | Mean (%) | 0.25–0.5 mm Content (%) | Mean (%) | < 0.25 mm Content (%) | Mean (%) |
|------------|-------------------|----------|-------------------|----------|-------------------|----------|-------------------|----------|-------------------|----------|-------------------|----------|
| N0_S_15    | 12.09 ± 2.24b1    | 5.93 ± 0.56a | 5.26 ± 2.35a    | 13.91 ± 1.62a | 19.29 ± 1.97a | 43.52 ± 8.29a |
| N0_IS_15   | 56.72 ± 14.28a    | 3.02 ± 0.35b | 4.48A            | 4.18A     | 6.13 ± 1.59b     | 10.02A   | 11.15 ± 3.86b     | 15.22A   | 19.88 ± 8.54b      | 31.70A   |
| N2_S_15    | 50.40 ± 6.01a     | 4.83 ± 1.06a | 10.66 ± 3.41a   | 8.12 ± 1.94a | 9.66 ± 0.59a     | 16.33 ± 4.45b |
| N2_IS_15   | 38.34 ± 3.85b     | 2.98 ± 1.91a | 4.61 ± 2.59a    | 11.99 ± 3.76a | 14.87 ± 5.06a | 12.26A   | 27.21 ± 2.99a     | 21.77A   |
| N0_S_30    | 9.58 ± 2.78a      | 0.28 ± 0.22a | 1.32 ± 1.03a    | 3.95 ± 1.65a | 16.09 ± 1.63a   | 68.78 ± 4.46a |
| N0_IS_30   | 3.75 ± 5.32b      | 0.53 ± 0.36a | 0.41A            | 1.79 ± 0.68a | 8.34 ± 2.50a    | 6.15A   | 18.13 ± 1.24a     | 17.11A   | 67.47 ± 8.48a      | 68.12A   |
| N2_S_30    | 1.58 ± 2.46a      | 0.99 ± 0.87a | 2.55 ± 1.64a    | 6.82 ± 2.28b | 20.19 ± 5.54a   | 67.8 ± 11.02a |
| N2_IS_30   | 1.80 ± 3.03a      | 1.69A      | 0.33 ± 0.25a    | 0.66A     | 1.11 ± 0.25a    | 1.83A   | 11.09 ± 1.02a     | 8.95A    | 17.24 ± 2.71a      | 18.72A   | 68.43 ± 5.49a      | 68.15A   |

ANOVA (P value)

|     | C     | N     | D     | C*N  | N*D   | C*D  | C*N*D |
|-----|-------|-------|-------|------|-------|------|-------|
| C   | 0.018 | 0.341 | 0.000 | 0.000| 0.002 | 0.002| 0.000 |
| N   | 0.002 | 0.034 | 0.000 | 0.013| 0.091 | 0.005| 0.000 |
| D   | 0.019 | 0.131 | 0.005 | 0.140| 0.109 | 0.071| 0.004 |
| C*N | 0.010 | 0.140 | 0.003 | 0.112| 0.109 | 0.071| 0.004 |
| N*D | 0.008 | 0.109 | 0.005 | 0.013| 0.109 | 0.071| 0.004 |
| C*D | 0.002 | 0.112 | 0.003 | 0.112| 0.109 | 0.071| 0.004 |

1 Different lower-case letters in the same column indicate significant differences between monoculture and intercropping treatments (P ≤ 0.05)

2 Data are the means of monoculture and intercropping treatments followed by different capital letters in the same column indicating significant differences between the two nitrogen levels for the same soil aggregate fraction (P ≤ 0.05)

C: Cropping systems; N: Nitrogen application level; D: Soil depths; C*N: Interaction between cropping systems and the nitrogen application level; N*D: Interaction between the nitrogen application level and soil depths; C*D: Interaction between the cropping systems and soil depths; C*N*D: Interaction among the above three factors
Fig. 1 Mean weight diameter (MWD), geometric mean diameter (GMD), and percentage of aggregate destruction (PAD) values in maize (left) and soybean (right) monocropping (M or S) and intercropping (IM or IS) systems at soil depths 0–15 cm (a, c, e) and 15–30 cm (b, d, f). * means significant difference at P ≤ 0.05, ** means significant difference at P ≤ 0.01, *** means significant difference at P ≤ 0.001, ns means no significant difference. C: cropping systems; N: Nitrogen application level (N0 or N2); C*N: Interaction between the nitrogen application level and cultivation method, a and b represent the significance level at P ≤ 0.05 between monoculture and intercropping at the same nitrogen level.
Fig. 2 AMF alpha-diversity in maize monocropping (M) and intercropping (IM) systems at different soil depths with different nitrogen application rates (N0 or N2). The three-way ANOVA results between cropping systems (C), nitrogen application level (N) and soil depth (D) are listed in Table 3.

Table 3 Statistical results of three factorial ANOVA of AMF alpha-diversity in maize/soybean intercropping (IMS) and corresponding monoculture (M and S) systems at different soil depths with two different nitrogen application rates (N0 and N2) (Figs. 2 and 3)

| ANOVA (P-value) | Maize | Soybean |
|-----------------|-------|---------|
|                 | Shannon | Simpson | ACE | Chao | Shannon | Simpson | ACE | Chao |
| C               | 0.815 | 0.620 | 0.021 | 0.002 | 0.000 | 0.000 | 0.356 | 0.354 |
| N               | 0.001 | 0.036 | 0.000 | 0.000 | 0.281 | 0.061 | 0.589 | 0.814 |
| D               | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.130 | 0.015 |
| C*N             | 0.020 | 0.168 | 0.714 | 0.248 | 0.974 | 0.061 | 0.171 | 0.445 |
| N*D             | 0.006 | 0.002 | 0.902 | 0.078 | 0.019 | 0.006 | 0.580 | 0.420 |
| C*D             | 0.155 | 0.733 | 0.589 | 0.126 | 0.094 | 0.000 | 0.014 | 0.056 |
| C*N*D           | 0.743 | 0.859 | 0.764 | 0.559 | 0.272 | 0.132 | 0.161 | 0.782 |

C Cropping systems; N Nitrogen application level; D Soil depths; C*N Interaction between cropping systems and the nitrogen application level; N*D Interaction between the nitrogen application level and soil depths; C*D Interaction between the cropping systems and soil depths; C*N*D Interaction among the above three factors
and 45–60 cm, respectively when compared with that in the monoculture system. Similarly, the AMF diversity in soybean soil was also affected by N application. The application of N significantly improved AMF diversity for both S (increased by 13% and 17% at 0–15 cm and 15–30 cm soil depths, respectively) and IS (increased by 25% and 8.7% for 0–15 cm and 45–60 cm soil depths, respectively) systems. However, the AMF diversity of soil decreased with increasing N application at soil depths of 30–60 cm and 15–45 cm in the S and IS systems, respectively.

Distribution of AMF composition across the soil profiles

In the M and IM systems, four genera including *Glomus_f_Glomeraceae*, *Paraglomus*, *unclassified_c_Glomeromycetes*, and others were observed at each soil depth for both N0 and N2 (Fig. 3). *Glomus_f_Glomeraceae* was the major genus of AMF in the M and IM systems, and its abundance was significantly positively correlated with the cropping system. The relative fraction of *Glomus_f_Glomeraceae* in the M system ranged from 61 to 99%, which is lower than that in the IM system, i.e., 87–99%. On the other hand, the relative fraction of *Paraglomus* in the IM system (0–6.7%) was lower than that in the M system (0–39%). However, the cropping system showed no significant influence on the relative fraction of *unclassified_c_Glomeromycetes*, while N application led to an increase in the relative fraction of *unclassified_c_Glomeromycetes*.

Compared with the AMF communities in the maize soil, *unclassified_f_Gigasporaceae* and *Gigaspora* were observed in the soybean soil at 0–15 cm depth (Fig. 3). Similarly, intercropping led to a

![Fig. 3 AMF alpha-diversity in soybean monocropping (S) and intercropping (IS) systems at different soil depths with different nitrogen application rates (N0 or N2). The three-way ANOVA results between cropping systems (C), nitrogen application level (N) and soil depth (D) are listed in Table 3](image-url)
decrease in the relative fraction of *Gigaspora* in the soybean soil at 0–15 cm depth for both N0 and N2. For example, *Gigaspora* formed 22% and 4.8% of the total AMF genera in the S and IS systems, respectively, for N0. Moreover, the relative fraction of *Glomus_f_Glomeraceae* in the soybean soil after N application was lower than that for N0, while the relative fractions of *Paraglomus* and *unclassified_c_Glomeromycetes* after N application were higher than those for N0.

Furthermore, PCoA results indicated that the AMF communities in the monoculture system were separate...
from those in the intercropping system (Fig. 5). Similarly, the N application resulted in significantly different AMF communities in the IM system from those in the IS system. In addition, the AMF communities across soil depths were different for N0 and N2. Similarly, there were differences in the AMF communities among intercropping and monoculture systems across soil depths. These results indicate that N application rates and cropping systems affect the composition of AMF communities.

Soil aggregate affected by AMF and soil nutrient

Soil nutrients, including TOC, TN, AN, AP, AK were determined in this study (Table S1 and S2). The results highlighted that the cropping patterns and N application had a significant influence on the soil nutrients. For instance, the TOC at 0–15 cm depth in the IM and IS systems for N0 was higher than that in the corresponding monoculture systems. However, the TOC at 0–15 cm depth in the IM and IS systems decreased with the N application. Moreover, the available soil nutrients (e.g., AP) in each layer of the intercropping system were higher than those in the monoculture system both for N0 and N2. Therefore, based on the changes in the soil aggregates, AMF communities, and soil nutrients, the SEM was conducted to evaluate the direct and indirect relationships between the AMF communities and soil fertility. The findings indicated that the AMF communities were positively correlated with the aggregates (> 5 mm) and soil nutrients (Fig. 6a). The SEM suggested that both the AMF communities and TOC promoted the formation of soil macro-aggregates (> 5 mm) (Fig. 6a). However, high N application was negatively correlated to the formation of macro-aggregates. Moreover, the intercropping enhanced AMF diversity and soil nutrients, thus promoting the formation of

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**Fig. 5** Principal co-ordinates analysis (PCoA) of AMF communities at different soil depths (0-15 cm, 15-30 cm, 30-45 cm, 45-60 cm) for maize monocropping and intercropping (M, IM) under two N fertilizer levels N0 and N2 (a, b), and soybean monocropping and intercropping (S, IS) under two N fertilizer levels N0 and N2 (c, d)
soil macro-aggregates (> 5 mm). As soil aggregates and TOC are directly correlated to soil stability and fertility, these results suggest that the AMF communities might indirectly influence soil stability and fertility by regulating soil aggregates and organic carbon. Moreover, N application and cropping system may also indirectly influence the soil stability and fertility via the regulation of the AMF community. Therefore, it can be inferred that the interactions between AMF communities and soil aggregates may promote soil stability and fertility.

Discussion

Effects of intercropping on the soil aggregates

The aggregates are important structural units of soil, and their composition and stability are typically used as important indicators of soil fertility (Tian et al. 2019). An analysis of different sizes of soil aggregates in this study indicated that intercropping significantly improved the formation of large soil aggregates (Table 1), which has also been reported for other intercropping systems previously. For instance, previous studies showed that the soil macro-aggregates increased by 50% in a walnut/perennial kura clover intercropping system compared to those in the respective monocultures (Kremer and Kussman 2009). Li et al. (2021) showed that when compared with monocultures, intercropping may increase soil fertility by improving the soil organic matter, TN, and macro-aggregates. In this study, the soil macro-aggregate content (> 5 mm) in the maize/soybean intercropping system at 0–15 cm soil depth was significantly higher than that in the corresponding monocultures for N0 (Table 1). Generally, the soil aggregates can be influenced by the soil microorganisms, plant roots, soil fauna, inorganic binding agents, and environmental variables (Tian et al. 2019). The results from this study implied that intercropping improved the stability of the soil structure at 0–15 cm depth by increasing the soil macro-aggregates (Fig. 1). This may be due to more developed root systems of the intercrops leading to the entanglement of soil particles and consolidation effect, which promote the formation of soil macro-aggregates. These results are also consistent with the findings based on the GMD and MWD indicators. Although the macro-aggregates increased in the IM at 15–30 cm depth and decreased in IS system compared to those in the corresponding monocultures, GMD and MWD analyses revealed that there were no significant differences between the intercropping and monoculture systems at 15–30 cm soil depth (Fig. 1). The present results indicated that the structural stability of deep soil was not affected by the intercropping system. Bai et al. (2019) found that soil macro-aggregate content at 0–10 cm soil depth was higher

Fig. 6 Structural equation model (SEM) showing the direct and indirect effects of the key factors on the soil stability and quality (a). Standard total effects (direct plus indirect effects) derived from SEM (b). Red and black solid arrows represent the pathways that are significantly positive and negative, respectively, and blue dashed arrows indicate the non-significant pathways. The path coefficients are adjacent to the arrows, * means \( P \leq 0.05 \), ** means \( P \leq 0.01 \), *** means \( P \leq 0.001 \).
than that at 10–20 cm and 20–30 cm soil depths, and was significantly correlated with root length, root surface area, and root length density, with the decrease in significant correlations with increasing soil depth. Moreover, Tian et al. (2019) showed that AMF played an important role in the formation of soil macro-aggregates by secretion of polysaccharides, as well as by fungal hyphae entanglement to form a larger fraction of macro-aggregates. In this study, the AMF diversity at 0–15 cm soil depth in the intercropping system was significantly higher than that at other soil depths (Figs. 2 and 3), which might have contributed to the increasing macro-aggregate content at 0–15 cm soil depth. These results indicated that the intercropping system can improve the stability of soil aggregates due to changes in the soil AMF diversity.

Some studies have shown that fertilizers have a significant influence on soil aggregates (Guillou et al. 2011; Guo et al. 2020). Guillou et al. (2011) found that N addition may decrease water-stable aggregates, thereby affecting the stability of the soil structure. However, the experimental results were found to be inconsistent in different regions, presumably due to the differences in the physicochemical and biological properties of various soil types (Wang et al. 2022). For instance, some studies have observed that higher fertilizer input in croplands is positively correlated with the formation of water-stable aggregates (Guo et al. 2020). In this study, the macro-aggregates (> 5 mm) in the maize/soybean intercropping system for N0 were higher than that for N2 (Tables 1 and 2), which may be due to the changes in soil structure and AMF diversity. A decrease in TOC with an increase N application was observed in the maize/soybean intercropping systems (Tables S1 and S2). Moreover, in this study, the results indicated that N application significantly reduced the AMF diversity across soil depths in the intercropping system, which may also have contributed to the reduction in soil aggregate stability. Previous studies have also highlighted that the soluble C in plant roots decreased with the increase in N, thereby negatively affecting the diversity and abundance of AMF in rhizosphere soil and roots (Schwab et al. 1991; Zhang et al. 2021). Therefore, it can be stated that higher N application levels can decrease the stability of macro-aggregates in the intercropping system.

Impact of N and intercropping on AMF communities across soil profiles

Previous studies have shown that soil fungal diversity is closely related to the soil environment and plant species (Marschner et al. 2004; Zhang et al. 2020). In this study, it was observed that the AMF diversity across soil depths in the maize/soybean intercropping system was higher than that in the monocultures for N0. These findings are consistent with those observed in the previous study involving legume/cereal intercropping systems (Wang et al. 2016). The roots of cereals in the legume/cereal intercropping system may extend to the root regions of legumes to form an interactive system. The underground root–root interactions between the intercrops can cause changes in the soil characteristics (Wang et al. 2016), which affect the diversity of soil AMF (Guo et al. 2020). Moreover, the changes in AMF alpha diversity indices in the intercropping systems may be due to the changes in soil fertility. In the maize/soybean intercropping system, maize generally shows a stronger competitive advantage for the soil nutrients, which leads to changes in the heterogeneous distribution of soil nutrients such as N (Jensen 1996; Zhang et al. 2020). In the present study, the soil nutrients may primarily lead to changes in the AMF diversity as the available nutrients content in the intercropping system was found to be higher than that in the monoculture systems (Tables S1 and S2). The changes in alpha diversity were observed in the maize/soybean intercropping system across soil depths, probably due to the differences in distribution characteristics of soil nutrients in the intercropping system. Thus, the plant diversity in the intercropping system can lead to changes in soil fertility and nutrient content, thereby affecting the diversity of soil microbes (Larkin and Honeycutt 2006; Xu et al. 2009).

This study also indicated that the diversity of AMF across soil depths was affected by N application rates. N application significantly reduced the AMF diversity in the soils at all depths in the IM system, while improving the AMF diversity at 0–15 cm and 45–60 cm soil depths in the IS system (Figs. 2 and 3). These results may be due to the different response mechanisms of different plant species to N stress (Zhang et al. 2020). Previous studies have reported that high N in the soil generally inhibits mycorrhizal colonization because the AMF growth is positively
correlated with the amount of soluble carbon in the soil system (Zhou et al. 2011). Generally, the soluble carbon content is negatively correlated with the N levels. The soluble carbon content has been reported to decrease under high N levels, resulting in the suppression of AMF communities (Zhou et al. 2011). In the intercropping system, the maize root system is usually dominant. Therefore, a large amount of N was taken up by the maize roots, resulting in a low N level in the soybean soil. This alleviates the inhibitory effect of high N on soybean mycorrhizal fungi and promotes the growth of AMF in the soybean soil (Corre et al. 2006).

This study found that the diversity and the relative fraction of AMF communities were significantly correlated with the cropping patterns. In addition, there were significant differences between different plant species, which may be because of the root distribution of different crops and competition for carbon and nutrients. Moreover, the abundance of AMF communities across soil depths was found to differ significantly between the monoculture and intercropping systems. Previous studies have indicated that an intercropping system could influence the relative abundance of AMF communities in the topsoil (Pereira et al. 2018; Yu et al. 2015; Zhang et al. 2020). However, in the present study, intercropping significantly improved the abundance of AMF communities in the deeper soil. This can be attributed to the overlap between the roots of different plants in the intercropping system leading to differences in the composition and the relative abundance of AMF communities in the soils at different depths (Zhang et al. 2020). Liu et al. (2015) demonstrated that the root overlap between the two crops in a maize/wheat intercropping system occurred at 10–80 cm depth, thereby contributing to the abundance of AMF communities in deep soils. Thus, this study further indicated that the intercropping system can influence the composition and relative fractions of AMF communities in the topsoil as well as deep soils.

Previous studies have reported that the composition of AMF communities is significantly correlated to the N application levels. Under long-term application of N at high levels, the stability and activity of the AMF communities exhibited decreasing trends (Lin et al. 2015). In this study, different AMF genera responded differently to the N application levels. For example, *Glomus_f_Glomeraceae* was the most common and abundant genus in the maize/soybean intercropping systems, indicating that *Glomus_f_Glomeraceae* can better adapt to high N environments. This may be due to their higher sporulation rates and symbiotic relationships with the plant roots (Oehl et al. 2003; Zhang et al. 2020).

Relationship between AMF communities and soil aggregates

AMF are essential for the soil ecosystem functions and play an important role in soil fertility by forming mutually beneficial mycorrhizal symbioses with plants (Bainard et al. 2014). Previous studies have indicated that the mycorrhizal symbiosis formed between AMF and plants become the bridge for nutrient fluxes. Meanwhile, various compounds (e.g., glomalin) were exuded from AMF during symbiosis, playing a pivotal role in soil carbon/nitrogen storage (Piliarova et al. 2019). Moreover, AMF was also positively correlated to the stabilization of soil aggregates in wheat/maize and faba bean/maize intercropping systems (Song et al. 2007). The SEM analysis was conducted in the present study to investigate the relationship between AMF and soil aggregates. The results highlighted a positive effect of AMF on the soil macro-aggregates (>5 mm) across soil depths. The stronger fungal hyphae of AMF entangle and enmesh primary particles or micro-aggregates to form macro-aggregates (Piotrowski et al. 2004; Wang et al. 2010). Moreover, previous studies have found that AMF facilitates the formation of soil macro-aggregates through the formation of several secondary metabolites (Caesar-TonThat and Cochran 2000; Tian et al. 2019). For example, the polysaccharides and phenolic acids secreted by AMF can combine with soil clay particles and micro-aggregates, thus promoting the formation of macro-aggregates (Piotrowski et al. 2004; Wang et al. 2010). Moreover, previous studies have found that AMF facilitates the formation of soil macro-aggregates through the formation of several secondary metabolites (Caesar-TonThat and Cochran 2000; Tian et al. 2019). Gao et al. (2017) found that AMF can increase the glomalin-related protein (GRSP) content, which can affect the formation of soil macro-aggregate fractions and contribute to the stability of soil aggregates. Lehmann and Rillig (2015) reported that saprotrophic fungi degrade organic materials (plant leaves or roots) and provide the necessary raw materials for the formation of soil aggregates. Similarly, Cao et al. (2022) indicated that AMF could indirectly contribute to the decomposition of organic materials and the abundance of overall extracellular enzymes by stimulating
the growth of saprotrophic fungi through a symbiosis between AMF and these saprotrophic fungi. This may also be a pathway by which soil AMF promotes the formation of soil aggregates. The results from this study implied that the intercropping system can positively influence the AMF alpha diversity and community composition as well as soil aggregate stability compared to the monoculture system (Figs. 1 and 2). In addition, the AMF communities in the intercropping system have a positive effect on the formation of aggregates (> 5 mm), as indicated by the SEM analysis (Fig. 6). Therefore, the intercropping system could influence the formation of soil aggregates by altering the structure and relative abundance of soil AMF communities, thus affecting the stability of the soil system.

Conclusions

The results indicated that intercropping improved the stability of aggregates at 0–15 cm soil depth but had no significant effect in deep soils. Moreover, the intercropping system led to a significant increase in the AMF diversity of maize and soybean soils, while the N application reduced soil AMF diversity across soil depths. The intercropping system could influence the formation of soil aggregates by altering the relative abundance and community composition of soil AMF, as indicated by the SEM analysis. These findings indicate that intercropping can improve the stability of soil aggregates and promote the formation of large aggregates by contributing to the soil nutrients and diversity of mycorrhizal fungi. This study focuses on understanding the changes in the AMF communities and their dynamic interactions with aggregates across soil depths in the intercropping system. Moreover, it emphasizes on the need to conduct future research on regulating the performance of the soil ecosystem by optimizing fertilization and cropping patterns.

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Data availability The sequence files were submitted to the NCBI Sequence Read Archive repository (http://www.ncbi.nlm.nih.gov/sra) and are accessible with the accession number: Bio Project PRJNA781197.

Declarations

Ethics The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

Bai L, Fan M, Wang Z, Wang T, Deng C, Li Y (2019) Relationship between root characteristics and aggregate stability in the field with maize and soybean intercropping. Res Soil Water Conserv 26(1):124–129 (In Chinese)
Bainard LD, Bainard JD, Hamel C, Gan Y (2014) Spatial and temporal structuring of arbuscular mycorrhizal communities is differentially influenced by abiotic factors and host crop in a semi-arid prairie agroecosystem. FEMS Microbiol Ecol 88:333–344
Bélanger J, Pilling D (eds) (2019) The State of the World’s Biodiversity for Food and Agriculture. FAO Commission on Genetic Resources for Food and Agriculture
Bray RH, Kurtz LT (1945) Determination of total, organic and available forms of phosphorus in soils. Soil Sci 59:39–46. https://doi.org/10.1097/00010694-194501000-00006
Caesar-TonThat TC, Cochran VL (2000) Soil aggregate stabilization by a saprophytic lignin-decomposing basidiomycete fungus. I Microbiological Aspects Biol Fertil Soils 32:374–380. https://doi.org/10.1007/s0037400000263
Cao T, Fang Y, Chen Y, Kong X, Yang J, Alharbi H, Kuzyakov Y, Tian X (2022) Synergy of saprotrophs with mycorrhiza for litter decomposition and hotspot formation depends on nutrient availability in the rhizosphere. Geoderma 410:115662. https://doi.org/10.1016/j.geoderma.2021.115662
Chao A, Lee SM (1992) Estimating the number of classes via sample coverage. J Am Stat Assoc 87:210–217. https://doi.org/10.1080/01621459.1992.10475194
Chen P, Du Q, Liu XM, Zhou L, Hussain S, Lei L, Song C, Wang XC, Liu WG, Yang F, Shu K, Liu J, Du JB, Yang WY, Yong TW (2017) Effects of reduced nitrogen inputs on crop yield and nitrogen use efficiency in a long-term maize-soybean relay strip intercropping system. PLoS
One 12(9):e0184503. https://doi.org/10.1371/journal.pone.0184503

Corre HG, Fustec J, Crozat Y (2006) Interspecific competition for soil N and its interaction with N2 fixation, leaf expansion and crop growth in pea-barley intercrops. Plant Soil 282:195–208. https://doi.org/10.1007/s11104-005-5777-4

Gao Y, Zong J, Que H, Zhou Z, Xiao M, Chen S (2017) Inoculation with arbuscular mycorrhizal fungi increases glomalin-related soil protein content and PAH removal in soils planted with Medicago sativa L. Soil Biol Biochem 115:148–151. https://doi.org/10.1016/j.soilbio.2017.08.023

Garland G, Bunemann EK, Oberson A, Frossard E, Six J (2017) Plant mediated rhizospheric interactions in maize-pigeon pea intercropping enhance soil aggregation and organic phosphorus storage. Plant Soil 415:37–55. https://doi.org/10.1007/s11104-016-3145-1

Guillou CL, Angers DA, Leterme P, Menasseri-Aubry S (2011) Differential and successive effects of residue quality and soil mineral N on water-stable aggregation during crop residue decomposition. Soil Biol Biochem 43:1955–1960. https://doi.org/10.1016/j.soilbio.2011.06.004

Guo L, Shen J, Li B, Li Q, Wang C, Guan Y, D’Acquisto LP, Luo Y, Tao Q, Xu Q, Li H, Yang J, Tang X (2020) Impact of agricultural land use change on soil aggregate stability and physical protection of organic C. Sci Tot Environ 707:136049. https://doi.org/10.1016/j.scitotenv.2019.136049

Hamel C, Strullu DG (2006) Arbuscular mycorrhizal fungi in field crop production: Potential and new direction. Can J Plant Sci 86(4):941–950. https://doi.org/10.4141/P05-099

He Y, Ding N, Shi JC, Wu M, Liao H, Xu JM (2013) Profiling of microbial PLFAs: Implications for interspecific interactions due to intercropping which increase phosphorus uptake in phosphorus limited acidic soils. Soil Biol Biochem 57:625–634. https://doi.org/10.1016/j.soilbio.2012.07.027

Hou ZN, Li PF, Li BG, Gong J, Wang YN (2007) Effects of fertigation scheme on N uptake and N use efficiency in cotton. Plant Soil 290:115–126. https://doi.org/10.1007/s11104-006-9140-1

Jensen ES (1996) Grain yield, symbiotic N2 fixation and interspecific competition for inorganic N in pea-barley intercrops. Plant Soil 182:25–38. https://doi.org/10.1007/BF00010992

Kemper WD, Rosenau RC (1986) Aggregate stability and size distribution. In: Klute, A. (Ed.), Methods of Soil Analysis. Part 1 Am Soc Ag Soil Sci Soc Am Madison WI pp:425–442

Keylock CJ (2005) Simpson diversity and the Shannon-Wiener index as special cases of a generalized entropy. Oikos 109:203–207. https://doi.org/10.1111/j.0300-1299.2005.13735.x

Kremser RJ, Kussman R (2009) Soil quality in a pecan agroforestry system is improved with kura clover. Agrofor Syst 7:377–383

Larkin RP, Honeycutt CW (2006) Effects of different 3-year cropping systems on soil microbial communities and Rhizoctonia diseases of potato. J Phytopathol 96:68–79. https://doi.org/10.1049/PHYTO-96-0068

Lehmann A, Rillig MC (2015) Understanding mechanisms of soil biota involvement in soil aggregation: a way forward with saprobic fungi? Soil Biol Biochem 88:298–302. https://doi.org/10.1016/j.soilbio.2015.06.006

Li C, Li Y, Xie J, Liu Y, Wang Y, Liu X (2019) Accumulation of organic carbon and its association with macro-aggregates during 100 years of oasis formation. CATENA 172:770–780. https://doi.org/10.1016/j.catena.2018.09.044

Li X, Wang Z, Bao X, Sun J, Yang S, Wang P, Wang C, Wu J, Liu X, Tian X, Wang Y, Li J, Wang Y, Xia H, Mei P, Wang X, Zhao J, Yu R, Zhang W, Che Z, Gui L, Callaway RM, Tilman D, Li L (2021) Long-term increased grain yield and soil fertility from intercropping. Nat Sustain 4:943–950. https://doi.org/10.1038/s41593-021-00767-7

Liang J, Shi W (2021) Cotton/halophytes intercropping decreases salt accumulation and improves soil physico-chemical properties and crop productivity in saline-alkali soils under mulched drip irrigation: A three-year field experiment. Field Crop Res 262:108027. https://doi.org/10.1016/j.fcr.2020.108027

Lin G, McCormack ML, Guo D (2015) Arbuscular mycorrhizal fungal effects on plant competition and community structure. J Ecol 103:1224–1232. https://doi.org/10.1111/1365-2745.12429

Liu X (2022) Effect of intercropping soybean on the diversity of the rhizosphere soil arbuscular mycorrhizal fungal communities in wheat fields. Clean: Soil Air Water 50:2100014. https://doi.org/10.1002/clen.202100014

Liu YX, Zhang WP, Sun JH, Li X, Christie P, Li L (2015) High morphological and physiological plasticity of wheat roots is conducive to higher competitive ability of wheat than maize in intercropping systems. Plant Soil 397:387–399. https://doi.org/10.1007/s11104-015-2654-7

Lu RK (2000) Analysis methods of soil agricultural chemistry. China Agric Sci Technol Publ House

Ma YY, Zhang HC, Wang DZ, Guo XS, Yang T, Xiang XJ, Walder F, Chu HY (2021) Differential responses of arbuscular mycorrhizal fungal communities to long-term fertilization in the wheat rhizosphere and root endosphere. Appl Environ Microbiol 87:e00349-e421. https://doi.org/10.1122/AEM.00349-21

Marschner P, Crowley D, Yang CH (2004) Development of specific rhizosphere bacterial communities in relation to plant species, nutrition and soil type. Plant Soil 261:199–208. https://doi.org/10.1023/B:PLSO.0000035569.80747.c5

Miller RM, Jastrow JD (1990) Hierarchy of root and mycorrhizal fungal interaction with soil aggregation. Soil Biol Biochem 22(5):579–584. https://doi.org/10.1016/0038-0717(90)90001-G

Moraes JMAD, Zanchi CS, Pires GC, Moretti CF, Barbosa MV, Silva AO, Pacheco LP, Carneiro MAC, Oliveira RL, Kemenmeier K, Souza ED (2019) Arbuscular mycorrhizal fungi in integrated crop livestock systems with intercropping in the pasture phase in the Cerrado. Rhizosphere 11:100165. https://doi.org/10.1016/j.rhishp.2019.100165
Oehl F, Sieverding E, Ineichen K, Mader P, Boller T, Wiemken A (2003) Impact of land use intensity on the species diversity of arbuscular mycorrhizal fungi in agroecosystems of Central Europe. Appl Environ Microbiol 69:2816–2824. https://doi.org/10.1128/AEM.69.5.2816-2824.2003

Okolo CC, Gebresamuel G, Zenebe A, Haile M, Eze PN (2020) Accumulation of organic carbon in various soil aggregate sizes under different land use systems in a semi-arid environment. Agr Ecosyst Environ 297:106924. https://doi.org/10.1016/j.agee.2020.106924

Pereira APDA, Santana MC, Bonfim JM, Mescolotti DDL, Cardoso EJB (2018) Digging deeper to study the distribution of mycorrhizal arbuscular fungi along the soil profile in pure and mixed Eucalyptus grandis and Acacia mangium plantations. Appl Soil Ecol 128:1–11. https://doi.org/10.1016/j.apsoil.2018.03.015

Piliarova M, Ondreickova K, Hudcovicova M, Mihalik D, Kraic J (2019) Arbuscular mycorrhizal fungi- their life and function in ecosystem. Agric 65(1):3–15. https://doi.org/10.2478/agri-2019-0001

Piotrowski JS, Denich T, Klironomos JN, Graham JM, Rillig MC (2004) The effects of arbuscular mycorrhizas on soil aggregation depend on the interaction between plant and fungal species. New Phytol 164:365–373. https://doi.org/10.1111/j.1469-8137.2004.01181.x

Rabot E, Wiesmeier M, Schlüter S, Vogel HJ (2018) Soil structure as an indicator of soil functions: a review. Geoderma 314:122–137. https://doi.org/10.1016/j.geoderma.2017.11.009

Rillig MC, Mummey DL (2006) Mycorrhizas and soil structure. New Phytol 171:41–53. https://doi.org/10.1111/j.1469-8137.2006.01750.x

Schwab SM, Menge JA, Tinker PB (1991) Regulation of nutrient transfer between host and fungus in vesicular-arbuscular mycorrhizas. New Phytol 117:387–398. https://doi.org/10.1111/j.1469-8137.1991.tb00002.x

Šimanský V, Juriga M, Jonczak J, Uzarowicz Ł, Stępień W (2004) The effects of arbuscular mycorrhizas on soil aggregation of mycorrhizal arbuscular fungi along the soil profile and the rhizosphere. FEMS Microbiol Ecol 41:919–925. https://doi.org/10.1111/j.1574-6941.2003.tb08137.x

Song YN, Zhang FS, Marschner P, Fan FL, Gao HM, Bao XG, Sun JH, Li L (2007) Effect of fertilization and plant species on the phosphorus availability and uptake in maize (Zea mays L.) and faba bean (Vicia faba L.). Biogeochemistry 83:51–66. https://doi.org/10.1007/s10535-006-9048-4

Stoltz E, Nadeau E (2014) Effects of intercropping on yield, weed incidence, forage quality and soil residual N in organically grown forage maize (Zea mays L.) and faba bean (Vicia faba L.). Field Crop Res 169:21–29. https://doi.org/10.1016/j.fcr.2014.09.004

Tan W, Wang G, Huang C, Gao R, Xi B, Zhu B (2017) Physicochemical protection, rather than biochemical composition, governs the responses of soil organic carbon decomposition to nitrogen addition in a temperate agroecosystem. Sci Total Environ 589:282–288. https://doi.org/10.1016/j.scitotenv.2017.04.143

Tian X, Wang C, Bao X, Wang P, Li X, Yang S, Ding G, Christie P, Li L (2019) Crop diversity facilitates soil aggregation in relation to soil microbial community composition driven by intercropping. Plant Soil 436:173–192. https://doi.org/10.1007/s11104-018-03924-8

Wang Y, Xu J, Shen JH, Luo YM, Scheu S, Ke X (2010) Tillage, residue burning and crop rotation alter soil fungal community and water-stable aggregation in arable fields. Soil Tillage Res 107:71–79. https://doi.org/10.1016/j.still.2010.02.008

Wang G, Sheng L, Zhao D, Sheng J, Wang X, Liao H (2016) Allocation of nitrogen and carbon is regulated by nodulation and mycorrhizal networks in soybean/maize intercropping system. Front Plant Sci 7:1901. https://doi.org/10.3389/fpls.2016.01901

Wang XQ, Muhmood A, Lyu T, Dong R, Liu H, Wu S (2021) Mechanisms of genuine humic acid evolution and its dynamic interaction with methane production in anaerobic digestion processes. Chem Eng J 408:127322. https://doi.org/10.1016/j.cej.2020.127322

Wang F, Liu Y, Liang B, Liu J, Zong H, Guo X, Wang X, Song N (2022) Variations in soil aggregate distribution and associated organic carbon and nitrogen fractions in long-term continuous vegetable rotation soil by nitrogen fertilization and plastic film mulching. Sci Total Environ 835:155420. https://doi.org/10.1016/j.scitotenv.2022.155420

Xiao X, Xi BD, He XS, Zhang H, Li D, Zhao XY, Zhang XH (2019) Hydrophobicity-dependent electron transfer capacities of dissolved organic matter derived from chicken manure compost. Chemosphere 222:757–765. https://doi.org/10.1016/j.chemosphere.2019.01.173

Xu Y, Wang G, Jin J, Liu J, Zhang Q, Liu X (2009) Bacterial communities in soybean rhizosphere in response to soil type, soybean genotype, and their growth stage. Soil Biol Biochem 41:919–925. https://doi.org/10.1016/j.soilbio.2008.10.027

Yang T, Liu X, Yang F, Song C, Wang X, Liu W, Su B, Zhou L, Yang W (2015) Characteristics of nitrogen uptake and transfer in a wheat-maize relay intercropping system. Plant Prod Sci 18:388–397. https://doi.org/10.1626/pps.18.388

Yu C, Hu XM, Deng W, Li Y, Xiong C, Ye CH, Han GM, Li X (2015) Changes in soil microbial community structure and functional diversity in the rhizosphere surrounding mulberry subjected to long-term fertilization. Appl Soil Ecol 86:30–40. https://doi.org/10.1016/j.apsoil.2014.09.013

Zhang Z, Mallik A, Zhang J, Huang Y, Zhou L (2019) Effects of arbuscular mycorrhizal fungi on inoculated seeding growth and rhizosphere soil aggregates. Soil till Res 194:104340. https://doi.org/10.1016/j.soltrel.2019.104340

Zhang R, Mu Y, Li X, Li S, Sang P, Wang X, Wu H, Xu N (2020) Response of the arbuscular mycorrhizal fungi diversity and community in maize and soybean rhizosphere soil and roots to intercropping systems with different nitrogen application rates. Sci Total Environ 740:139810. https://doi.org/10.1016/j.scitotenv.2020.139810
Zhang S, Luo P, Yang J, Irfan M, Dai J, An N, Li N, Han X (2021) Responses of arbuscular mycorrhizal fungi diversity and community to 41-year rotation fertilization in brown soil Region of Northeast China. Front Microbiol 12:742651. https://doi.org/10.3389/fmicb.2021.742651
Zhou X, Yu G, Wu F (2011) Effects of intercropping cucumber with onion or garlic on soil enzyme activities, microbial communities and cucumber yield. Eur J Soil Biol 47:279–287. https://doi.org/10.1016/j.ejsobi.2011.07.001
Zhou M, Liu C, Wang J, Meng Q, Yuan Y, Ma X, Liu X, Zhu Y, Ding G, Zhang J, Zeng X, Du W (2020) Soil aggregates stability and storage of soil organic carbon respond to cropping systems on Black Soils of Northeast China. Sci Rep 10:265. https://doi.org/10.1038/s41598-019-57193-1

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