

Manure-based slurry film promotes reduced fertilizer input through microbially-mediated nutrient activation mechanism and its life cycle assessment

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KEYWORDS

Cow manure film, life cycle assessment, reduced fertilizer input, silage maize, soil microorganism

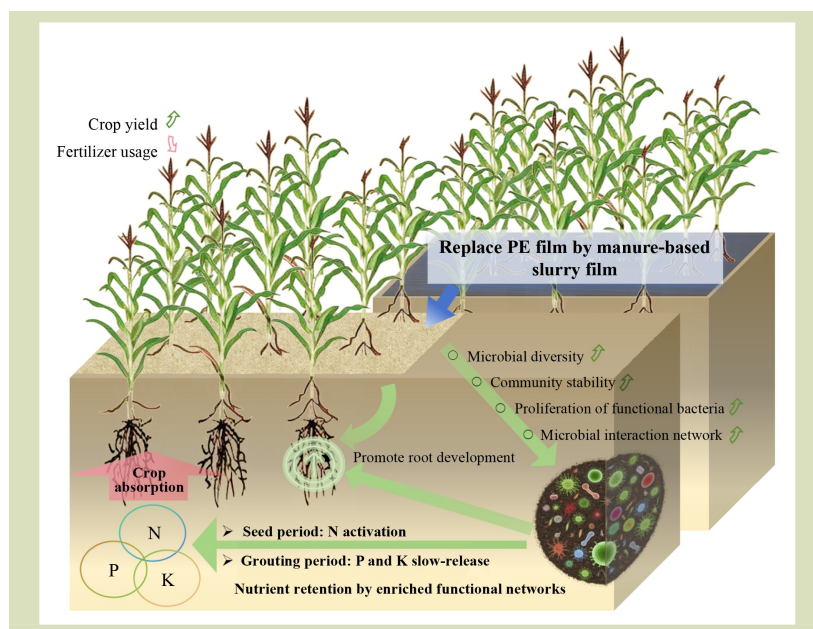
HIGHLIGHTS

- Manure-based slurry film (MSF) reduces fertilizer use while maintaining corn yield compared with PE film.
- MSF degrades naturally, reducing micro-plastic risks.
- MSF alters microbes to boost nutrient efficiency.

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GRAPHICAL ABSTRACT



ABSTRACT

This study investigated the effectiveness of manure-based slurry film (MSF) technology and validated its dual benefits in reducing mineral fertilizer input and enhancing fertilizer use efficiency through field experiments. The results showed that MSF maintained silage maize yield while reducing mineral

fertilizer input by 30%, and achieved a significant yield increase under 15% reduction in fertilizer input compared to standard polyethylene film. An investigation of soil microorganisms demonstrated that MSF changed the microbial community structure, promoting the activation and use efficiency of nutrients such as nitrogen, phosphorus and potassium, while increasing soil organic matter content. Life cycle assessment was performed with SimaPro 9.5 revealing that the environmental impact of MSF is significantly greater than that of polyethylene film across various environmental assessment factors. The high environmental impact of MSF production stems from its energy and water consumption, necessitating a focus on process simplification while maintaining high yield. This study provides new insights into the development of cleaner technologies, examines diversified uses of cow manure, emphasizes the role of MSF in reducing mineral fertilizer application, and highlights potential environmental risks.

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1 Introduction

In recent years, with the continuous rise in food demand, in pursuit of higher yields, excessive amounts of fertilizers are often applied, causing a range of damage to the cropping soil^[1]. The damage to the soil is mainly reflected in the retention of hydrogen ions in the soil, causing soil acidification and a reduction in soil colloids, leading to soil compaction, gradually reducing fertilizer use efficiency, resulting in crop yield reduction^[2,3]. Subsequently, farmers increase the amount of fertilizer application to again pursue high yields, forming a vicious cycle.

It has been proven that excessive application of mineral fertilizers has a lasting and significant impact on the physical and chemical properties of soil. This impact also involves various aspects, such as microbial communities, soil aggregate formation, and soil water retention. Therefore, agronomists have proposed several methods to enhance fertilizer use efficiency and reduce the amount of mineral fertilizers used^[4]. Currently, methods to increase fertilizer use efficiency mainly fall into three categories: application of fertilizer enhancers^[5], deep application of mineral fertilizers^[6], and combined use of organic and mineral fertilizers^[7]. Of these, the research and application of fertilizer enhancers are the most common. Typical enhancers (nitrification inhibitors and urease inhibitors) have been applied in various contexts and have shown promising results^[8]. For example, adding urease inhibitors^[9] or polyaspartic acid^[10] as enhancers to fertilizers

can reduce nutrient loss, extend the effectiveness period of fertilizers, and improve nutrient utilization rates. However, enhancers are not completely harmless; for example, urease inhibitors may affect microbial communities in the soil, leading to a reduction in microbial diversity^[11]. This may impact soil health and plant growth. Also, trace chelating agents and complexing agents in enhancers can also affect the surrounding environment after decomposition. Long-term use of enhancers may lead to increased soil dependency, degradation of overall effectiveness and rising cost pressures. Therefore, reports on their use in large-scale grain crop cultivation are rare. In addition, agronomists have suggested that deep application of mineral or organic fertilizers can improve fertilizer use efficiency^[12]. Research indicates that appropriate deep application of nitrogen fertilizers can prevent nitrogen volatilization and leaching, reducing nitrogen loss caused by nitrification and denitrification processes^[13]. However, deep application of nitrogen fertilizers requires more energy, equipment, and labor, which raise relative input costs and makes it difficult to promote and implement in actual production.

Currently, the combined application of organic and mineral fertilizers to improve fertilizer use efficiency is widely applied and has been extensively studied^[14]. For example, the study by Yang et al.^[15] found that the combined application of organic fertilizer and urea can increase organic colloids in the soil, form more soil aggregate structures, and significantly improve soil physical properties. Hu et al.^[16] found that the combined

application of organic and mineral fertilizers is conducive to the accumulation of soil organic carbon and active organic carbon, improving soil aeration and water and nutrient retention capacity. The research by Zhang et al.^[17] discovered that the combined application of organic and mineral fertilizers can effectively optimize the soil microbial community structure. Therefore, the combined application of organic and mineral fertilizers can improve soil physical, chemical microbiological properties, increasing soil organic matter (OM) and optimizing microbial colonies, and is one of the effective means to improve fertilizer use efficiency. However, the high cost of organic fertilizers made it challenging for individual farmers to combine organic and mineral fertilizers in a practical way^[18]. Medium and large-scale farms, due to the low profitability of organic fertilizers and the environmental pressure from the government, tend to treat manure in situ with the goal of harmless treatment rather than focusing on its fertilizer use efficiency^[19]. Therefore, in practical application, the combined application of organic and mineral fertilizers lacks strategic approach a systematic mechanism driven by profit incentives.

In our research, we introduced a method of preparing agricultural film with cow manure, creating a manure-based slurry film (MSF). The use of MSF also falls under the category of organic-mineral combined application and serves as an organic fertilizer application. The MSF could reduce planting costs by discontinuing the use of standard agricultural plastic film and generate income by increasing crop yield. The MSF technology uses an organic-mineral mixed fertilization approach, reducing mineral fertilizer use while eliminating microplastic pollution. Its biodegradable carrier design balances environmental friendliness with long-term cost advantages. In the long-term, this technology is expected to decrease soil microplastic content by reducing plastic mulch use, thereby mitigating the physical and biological impacts of microplastic on soil and lowering the risk of ecological degradation^[20,21].

Thus, it is evident that the promotion and application of MSF have significant motivational support from agronomists. Based on this, we propose a new concept that leverages the benefits of MSF, reduces the amount of mineral fertilizer applied and breaks the vicious cycle of soil degradation caused by excessive application of mineral fertilizers. In this study, we conducted an experiment to reduce the amount of mineral fertilizer input by applying MSF to silage maize planting. Throughout the

experiment, we monitored both the dry matter content of the crop and soil nutrient content across various growth stages. We also conducted a comparative analysis of the differences in microbial communities between MSF and standard plastic film. The objectives of this study were to: (1) validate the productivity outcomes for silage maize with reduced mineral fertilizer input using MSF, (2) analyze the mechanisms by which MSF influences crop yield, and (3) comparative analyze environmental effect between MSF and polyethylene (PE) film. This study aimed at contribute to this growing area research on limiting mineral fertilizer input by exploring the practical application value of MSF.

2 Materials and methods

2.1 Preparation of manure-based slurry film

MSF was developed by our group and prepared by the methods detailed in the Supplementary materials. Soil sampling method is shown in Fig.S1.

2.2 Silage maize planting experiment

Natural conditions: The experiment was conducted at Huarui Agricultural Co., Ltd. in Minle County, Zhangye City, Gansu Province, China (100°40'20" E, 38°43'33" N). Minle County features a temperate continental desert grassland climate, characterized by long sunshine hours and abundant heat resources. The region experiences significant temperature variations, low precipitation, strong evaporation, an arid climate and frequent dust storms. The altitude is 1629 m, with an annual solar radiation of approximately 586 kJ·cm⁻². The area receives between 2670 h and 3250 h of sunshine annually, with an average annual temperature ranging from 3.4 °C to 5.6 °C, a frost-free period of 78–188 d, and annual precipitation ranging from 246 to 531 mm. The average annual wind speed ranges from 2.0 to 2.8 m·s⁻¹.

The treatments were: standard fertilizer rate with MSF (MSF100%F), 85% fertilizer rate with MSF (MSF85%F) and 70% fertilizer rate with MSF (MSF70%F); and standard fertilizer rate with PE film (PE100%F), 85% fertilizer rate with PE film (PE85%F) and 70% fertilizer rate with PE film (PE70%F).

Production and sampling methods were as follows: The

planting period spanned from May 10 to August 22, 2024 (105 d). The growth period of maize was divided into two parts, vegetative growth (VE, V1, V2, V4, V6, V10, V12, V14 and VT) and reproductive growth (R1, R2, R3, R4, R5 and R6). The plant samples and soil samples of each sampling time were taken before each topdressing time (6 times totally at V1, V6, V12, VT, R2 and R6, Fig. 1(a)). A wide-narrow row planting pattern was employed (as illustrated in Fig. 1(b)), and the maize cultivar was Jinlin 57, which is locally adapted. The soil underwent plowing, compaction, fertilization, and plastic mulching prior to sowing. Seeds were planted in double-seed per hill, with empty hills reserved for future seedling preservation to maintain a sowing rate above 98%. A pre-emergence herbicide mixture of alachlor and oxyfluorfen was applied for weed control. All fields utilized drip irrigation for water management.

2.3 Distribution of soil nutrients

Before conducting the field planting experiment, we cleared

debris such as stones, weed roots, and residual film from the sandy soil, ensuring that there were no soil clods with a diameter > 5 cm on the surface, and kept the soil loose. Then, we used the five-point cross method for sampling (a total of 20 samples were taken to 20 cm deep), and divided all samples into three subsamples for testing (samples were biological replicates mixed across plots). The soil bulk density was 1.66 g·m⁻³, porosity 33%, water content 11% and aggregate (> 0.25 mm) content 1%. The soil nitrogen, phosphorus, potassium and OM distributions during the whole growth process of the silage maize were also measured (details are given in the Supplementary materials).

2.4 DNA extraction and high-throughput sequencing of soil

To examine the variation of microbial species in soil with PE film and MSF, soil samples were collected from PE100%F and MSF100%F treatments at the vegetative tasseling (VT) and harvest (R6) stages (respectively) along with an unmulched

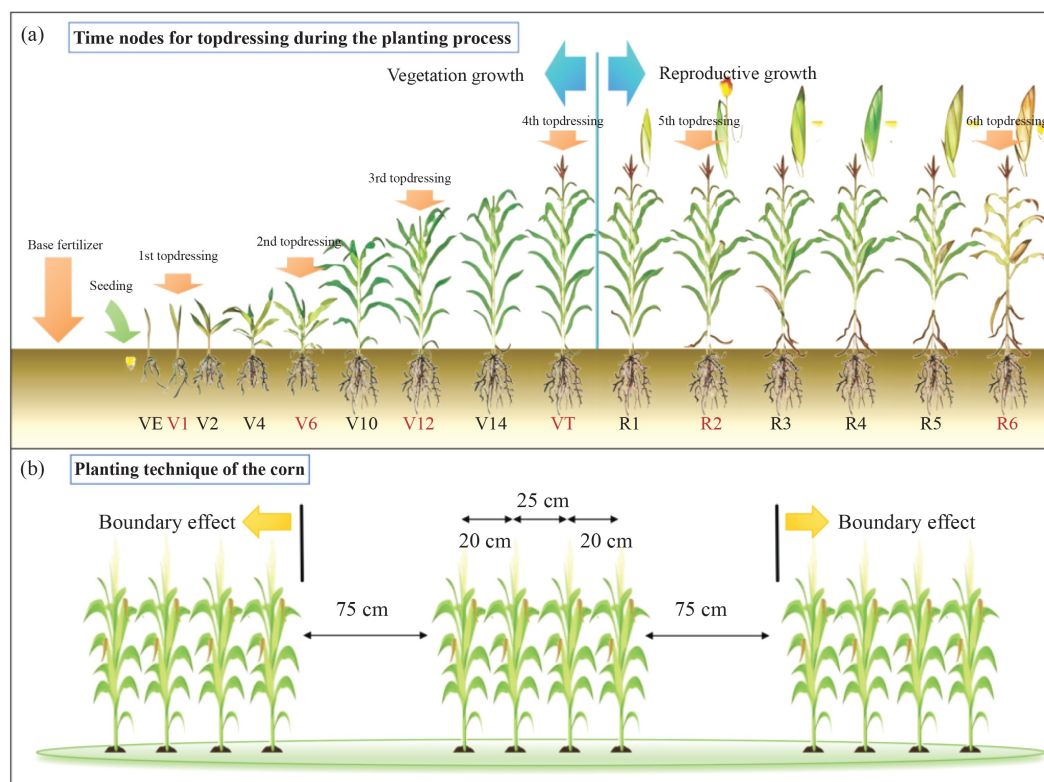


Fig. 1 (a) Time of topdressing during maize production, (b) maize planting technique.

control group (CK). The soil samples obtained were mixed and three subsamples of each were measured, samples are labeled as PE(VT)1–3, MSF(VT)1–3, CK(VT)1–3, PE(R6)1–3, MSF(R6)1–3 and CK(R6)1–3. Subsequently, DNA from the soil samples was extracted using a Power Soil DNA Isolation Kit (MOBIO Laboratories, Inc., New York, NY, USA) following the manufacturer's instructions. The DNA quality, including integrity, purity and concentration, was evaluated via a 1% agarose gel and a Nanodrop spectrophotometer (Nanodrop Technologies Inc., Wilmington, DE, USA). The 16S *rDNA* V3–V4 bacterial regions of the extracted genomic DNA were amplified using the forward primer 5'-CCTAYGGGRBGCASCAG-3' and the reverse primer 5'-GGACTACNNGGTATCTAAT-3'. PCR reactions were performed in triplicate, and the triplicate PCR products were combined and purified using the ENZA Gel Extraction Kit (Omega Bio-Tek Inc., Norcross, GA, USA). Subsequently, the amplification products were subjected to Illumina MiSeq (San Diego, California, USA) high-throughput sequencing and analysis. Sequencing and bioinformatic services were provided by Shanghai Majorbio Bio-pharm Technology Co., Ltd., (Shanghai, China). The microbial community composition at the genus level was analyzed using the amplicon sequencing data processed through the Wekemo Bioincloud platform^[22]. Bacterial α -diversity (Chao1 and Shannon indices) and β -diversity were analyzed for the samples using QIIME2^[23].

A correlation heat map was used to show the Spearman's correlations between microorganisms and soil nutrient content (N, P, K and OM).

Redundancy analysis (RDA) was performed using Wekemo Bioincloud^[22] to examine the relationships between microbial community features and environmental factors. Permutation tests (999 permutations) were applied to assess the statistical significance of these relationships. PERMANOVA dispersion analysis was used to account for potential confounding between experimental stages and treatment groups.

2.5 Life cycle assessment

Life cycle analysis was performed to evaluate the ecological effects of MSF, with the application context of manure-derived films integrated into a dairy feed and maize silage cultivation model. The assessment adhered to ISO 14040/14044 guidelines, establishing silage production of 1 t as the functional unit. System boundaries encompassed all input materials (including

irrigation water, agrochemicals and energy inputs). Environmental impacts were quantified using ReCiPe 2016 methodology across six categories: soil acidification, climate change effects on land ecosystems, tropospheric ozone creation, aquatic nutrient pollution, non-renewable resource depletion and land contamination potential. All modeling was conducted using SimaPro version 9.5 software.

2.5.1 System description

The system boundary is presented in Fig. 2, starting from cow manure treatment, then silage maize sowing through to harvest. The FU was defined as 1 t of silage maize from the different films.

This boundary covers the entire life cycle of silage maize, including, manure treatment, plastic film laying, sowing, irrigation and fertilization. The preparation processes of MSF, as well as the silage maize planting process described above and in the Supplementary materials.

The research scope (Fig. 2) encompassed the complete agricultural cycle from manure processing to silage maize cultivation and final harvest. This framework incorporated all key agricultural phases: mulch film application, seeding, water supply, fertilizer application and waste management. Additionally, it accounts for the manufacturing procedures of MSF alongside previously described crop growth methodologies.

Field-collected empirical data were supplemented with Ecoinvent 3.8 database references, with all inputs processed through SimaPro 9.5. The evaluation adopted a midpoint-focused analytical strategy, measuring six critical environmental parameters: soil acidification potential, carbon footprint, non-renewable energy consumption, aquatic nutrient enrichment, ground-level ozone generation and soil contamination risks. This methodology aligned with established agricultural life cycle analysis practices^[24] for quantifying ecological consequences across multiple dimensions. The specific list of each treatment has been listed in the supporting materials.

2.5.2 Life cycle inventory

The data on silage maize production required both investigation on the farm and support from background inventory databases. Primary data on material, electricity, and

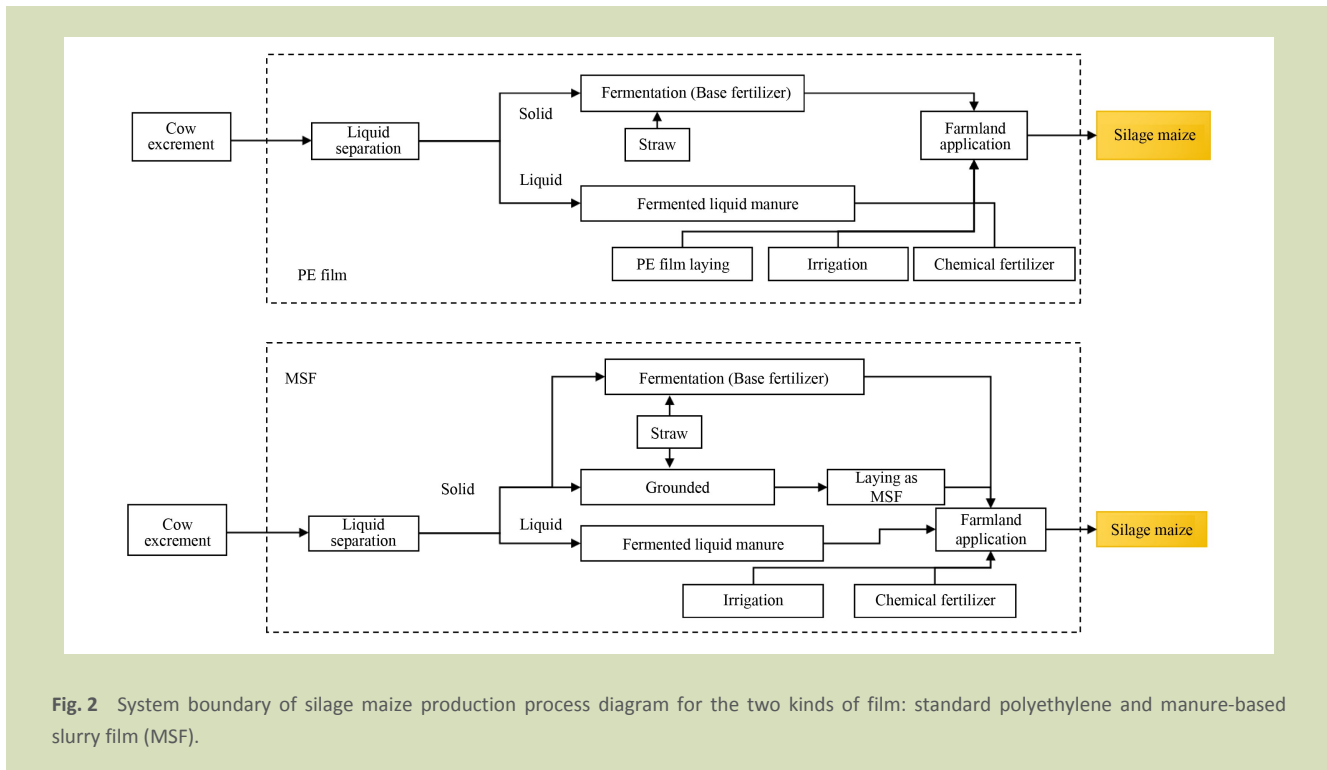


Fig. 2 System boundary of silage maize production process diagram for the two kinds of film: standard polyethylene and manure-based slurry film (MSF).

fuel consumption, fertilizer application, material straw use and transportation distance were derived from interviews with Huarui Agriculture Co., Ltd. In addition, on-site emission factors for CH₄, N₂O, NH₃ and P were mainly collected from the existing literature. Ecoinvent 3.8 was used for the upstream production processes of materials and energy, including electricity, diesel oil, straw and water. Priority was given to the background of the inventory that was suitable for the current production situation in China.

The production stage of this study was divided into manure treatment and farmland application, and the impact factors were calculated as:

$$I_i = I_{fai} \times S_i \tag{1}$$

Where, I_i is impact factors of film i (PE100%, PE85%, PE70%, MSF100%, MSF85%, and MSF70%) and I_{fai} is the impact factors of film i farm land application per hectare, and is calculated as:

$$I_{fai} = I_{film} + I_{irrigationwater} + I_{fertilizer} + I_{basefertilizer} \tag{2}$$

S_i is the area required to produce 1 t of silage maize with film i , calculated as:

$$S_i = \frac{1}{Q_i} \tag{3}$$

I_{film} is the impact factor of film per hectare, calculated as:

$$I_{film} = l_{film} \times Q_{film} \tag{4}$$

$I_{irrigationwater}$ is impact factor of water per hectare, obtained from field investigation, $I_{fertilizer}$ is the impact factor of fertilizer per hectare, obtained from field investigation and $I_{basefertilizer}$ is the impact factor of base fertilizer per hectare, and these are calculated as (using fertilizer as an example):

$$I_{basefertilizer} = l_{basefertilizer} \times Q_{bf} \tag{5}$$

Where, Q_i is the yield with film i per hectare. Likewise, l_{film} is the impact factor of film i per ton, obtained from field investigation, Q_{film} is the consumption of film i per hectare, obtained from field investigation, $l_{basefertilizer}$ is the impact factor of base fertilizer per ton, obtained from field investigation and Q_{bf} is consumption of base fertilizer per hectare, obtained from field investigation. The result was expressing in proportion P_i , calculated as:

$$P_i = \frac{I_i}{I_{max}} \tag{6}$$

Where, I_i is impact factors of film i (PE100%, PE85%, PE70%, MSF100%, MSF85% and MSF70%), I_{max} is the maximum impact factor of the films (PE100%, PE85%, PE70%, MSF100%, MSF85% and MSF70%).

Manure treatment and film preparation.

The cow manure in this study was treated using two methods. The first is the preparation of base fertilizer by fermentation: the untreated cow manure was placed into the fermentation tank after liquid separation by a solid-liquid separator, then aerobically fermented in the tank, which was operated by a flip throwing machine. The second was MSF preparation, and the amount of the matter was calculated using the production parameters of silage maize for different films. The impact factors of PE film and the manure treatment method are listed in support material.

Farmland application.

The inventory for planting 1 ha of farmland is presented in Supplementary materials. The planting processes for PE film and MSF were identical; thus, the basic data for 1 ha were the same. The date for producing 1 t of silage maize considered the yield differences of the films; therefore, the inventory of farmland applications for each film was multiplied by the yield coefficient, being the farmland area for producing 1 t of silage maize.

3 Results

3.1 Individual growth of silage maize under measures to reduce fertilizer input

The individual weights of silage maize produce in response to reduced fertilizer input were significantly different between PE film and MSF treatment groups (Fig. 3(a)). In the PE film treatment group, the dry matter weight was highest under standard fertilizer application (214 g at R6), while reducing fertilizer by 15% and 30% resulted in decreases of 2.0% and 8.7%, respectively, compared to the standard fertilizer treatment. Particularly from VT to grain filling stage, the PE film group showed higher sensitivity to reduced fertilizer, with the dry matter accumulation rate under 30% reduction being about 18% lower than the standard fertilizer treatment, possibly because to the limited water and fertilizer retention capacity of PE film led to early nutrient stress. Conversely, the MSF treatment group showed greater tolerance under reduced fertilizer application. When fertilizer was reduced by 15%, the dry matter weight increased by 9.6% compared to the standard fertilizer treatment (239 g at harvest); even with a 30% reduction, the weight did not differ significantly from the standard fertilizer treatment. Further analysis of the growth

period data revealed that the MSF had a clear advantage in dry matter accumulation from the V12 to harvest, with the 30% reduction treatment reaching 230 g at R6, significantly higher than the PE film group under the same treatment (at 195 g). Prior to the VT stage, the dry matter content of MSF maize was generally lower than that of PE film group.

The analysis of root dry matter weight at VT and R6 revealed that the MSF100%F treatment group had the highest average weight of 32.4 g at VT, significantly surpassing the average weight of 25.3 g for the PE film treatment groups (PE100%F, PE85%F and PE70%F) ($P < 0.05$). By R6, while the root dry matter weight of the PE70%F group decreased, the MSF treatment groups (MSF100%F, MSF85%F, MSF70%F) maintained stable weights that were consistently higher than those of the PE film treatment groups. In particular, the MSF100%F and MSF85%F groups had root dry matter weights of 34.49 g and 32.86 g, respectively, at R6. These results demonstrate that the use of MSF in conjunction with proper fertilizer application provides a notable benefit in increasing the accumulation of maize root dry matter (Fig. 3(b)).

3.2 Soil properties with manure-based slurry and polyethylene films

Experimental data indicate that MSF treatment is generally superior to standard PE film treatment in enhancing soil P, K and N content. For P content, soil treated with MSF had higher levels throughout the entire growth period (Fig. 4). At the V1, the average P content was 15.5 mg·kg⁻¹ (13.0–17.3 mg·kg⁻¹), significantly higher than that of the PE film treatment at 9.2 mg·kg⁻¹ (7.5–9.6 mg·kg⁻¹). By R6, P was 15.0 mg·kg⁻¹ (14.6–15.8 mg·kg⁻¹) for MSF treatment, compared to 12.3 mg·kg⁻¹ (9.8–14.0 mg·kg⁻¹) for PE film treatment. The difference in K content was even more pronounced. At V6, MSF treatment peaked at 190 mg·kg⁻¹ (164–209 mg·kg⁻¹), significantly higher than the 145 mg·kg⁻¹ (137–163 mg·kg⁻¹) of the PE film treatment. At VT, MSF treatment had a high level of K at 153 mg·kg⁻¹ (109–166 mg·kg⁻¹), while PE film treatment only had 116 mg·kg⁻¹ (91–142 mg·kg⁻¹). Total N content was also enhanced MSF treatment. At V1, the average N content was 473 mg·kg⁻¹ (412–522 mg·kg⁻¹), which was 21.3% higher than the PE film treatment at 390 mg·kg⁻¹ (243–398 mg·kg⁻¹). At V6, MSF treatment averaged 569 mg·kg⁻¹ (560–582 mg·kg⁻¹), slightly higher than the 548 mg·kg⁻¹ (523–562 mg·kg⁻¹) for the PE film treatment. The OM content had stage-specific differences. At V1, PE film

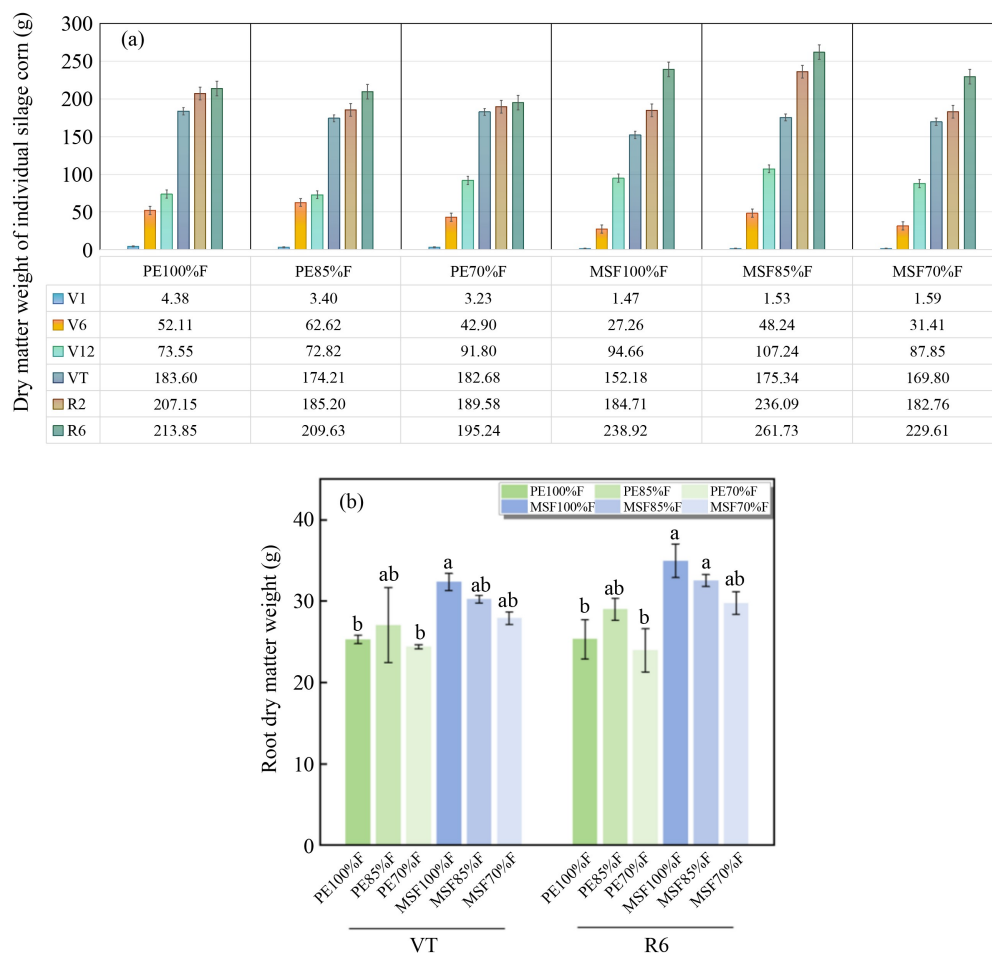


Fig. 3 (a) Average individual dry matter weight with different treatments and (b) root dry matter weight of the maize in the VT and R6 stages.

treatment was 1.03% (1.00%–1.05%), higher than MSF at 0.86% (0.75%–0.94%). However, from V6 onwards, the OM content of the MSF treatment was higher than that of the PE film treatment (2.91% vs 2.73%) and remained higher. It reached 0.83% (0.73%–0.93%) at R6, which was significantly higher than the PE film treatment at 0.47% (0.44%–0.55%).

Under the same type of film treatment, reducing the fertilizer application rate from 100% to 85% and 70% fertilizer application resulted in a general decline in overall soil nutrient content. However, MSF treatment provided greater tolerance to reduced fertilizer application. For example, in the PE film treatment, with 100% fertilizer application, P content at V1 was 9.6 mg·kg⁻¹, dropping to 7.5 mg·kg⁻¹ (22.4% decrease) with

70% fertilizer application. In contrast, MSF treatment decreased from 17.3 mg·kg⁻¹ with 100% fertilizer application to 13.0 mg·kg⁻¹ with 70% fertilizer application (24.5% decrease). Although the reductions were similar, the absolute value of MSF treatment remained significantly higher. In terms of K content, MSF treatment reached 209 mg·kg⁻¹ at the V6 stage with 100% fertilizer application, dropping to 164 mg·kg⁻¹ with 70% fertilizer application (21.5% decrease). The PE film treatment fell from 163 to 137 mg·kg⁻¹ (15.7% decrease) during the same period, indicating that MSF was more efficient in K release under high fertilizer application. Total N content remained stable in MSF treatment. At V1, the N content was 521.81 mg·kg⁻¹ with 100% fertilizer application, dropping to 412 mg·kg⁻¹ with 70% fertilizer application (21.0% decrease).

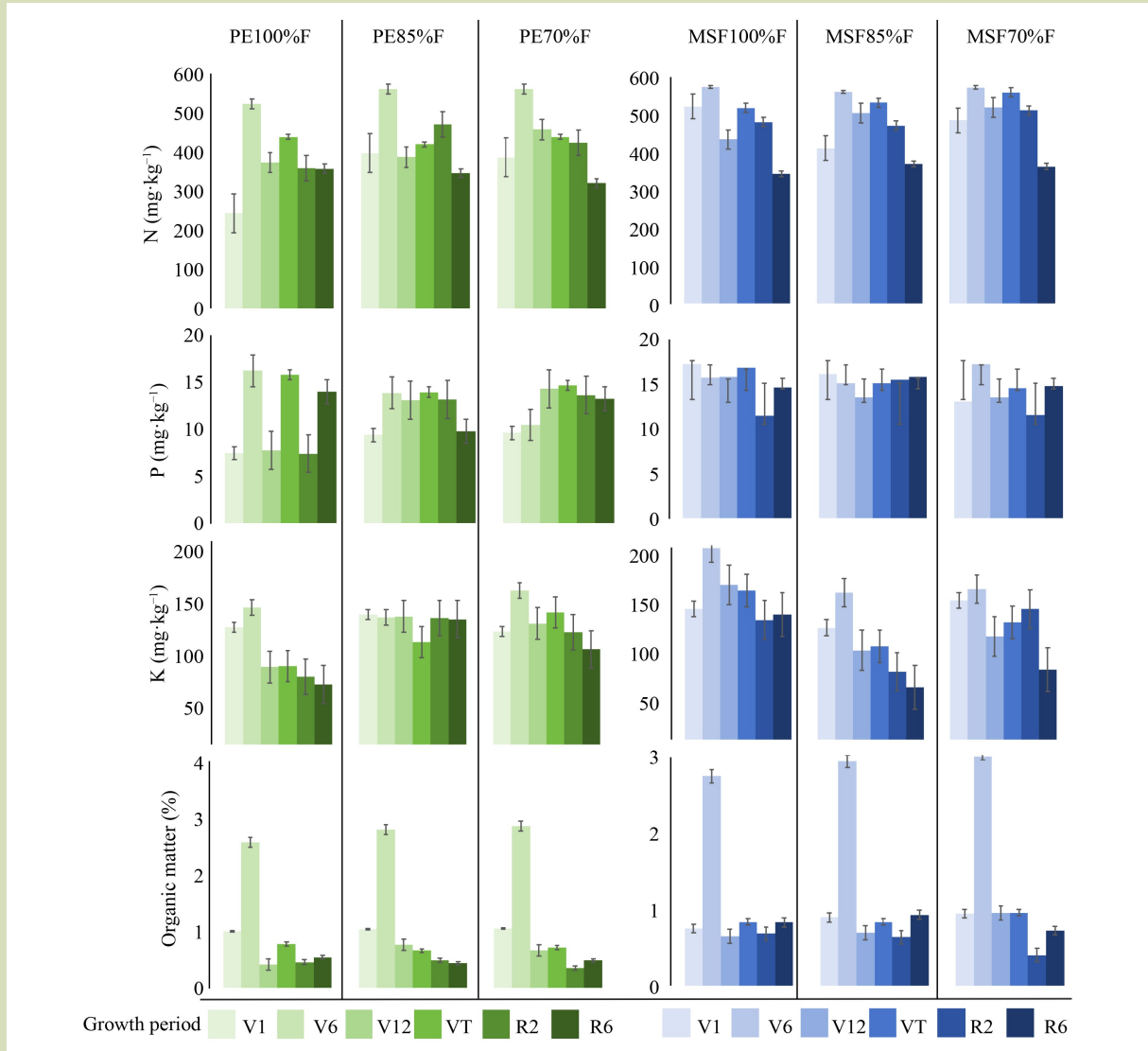


Fig. 4 Soil nutrient content in each growth period with two film (PE film, polyethylene film; and MSF, manure-based slurry film) and three fertilizer treatments (100%, 85% and 70% standard rate) in silage maize production system.

However, it was still higher than the 398 mg·kg⁻¹ in PE film treatment with 100% fertilizer application. The OM content was less affected by the fertilizer application rate. MSF treatment resulted in 0.73% with 70% fertilizer application at R6 (21.5% decrease from 0.93% with 100% fertilizer application), while the PE film treatment was only 0.44% (20.0% decrease from 0.55% with 100% fertilizer application). Notably, MSF treatment gave relatively high nutrient levels even with reduced fertilizer application. For example, at VT,

the OM content with 70% fertilizer application was 0.84%, higher than the 0.78% of PE film treatment with 100% fertilizer application.

The MSF treatment significantly had enhanced the soil P, K and total N content, especially as measured at the critical growth stages from V1 to VT, and this demonstrates stronger tolerance to reduced fertilizer application. PE film only excelled in OM content at V1 but was surpassed later.

3.3 Comparison of soil microorganisms between manure-based slurry and polyethylene films

The evaluation of the microbial community showed that the original soil was dominated by resident genera (e.g., *Pedospaera* and *Faecalibacterium*) and functional bacteria (e.g., nitrifying bacteria *Nitrospira* F 437423), with their abundance proportions being 12.3% and 1.8%, respectively. This reflected a relatively balanced carbon and N cycle function in undisturbed soil. However, after different mulch treatments, the microbial community significantly deviated from the original state: at VT with PE film treatment, the proportion of rapidly decomposing bacteria (*Romboutsia* B) increased from 4.2% in the original soil to 9.6%, while the resident bacteria *Faecalibacterium* dropped from 5.1% to 1.3%. At R6, the proportion of pathogenic bacteria *Escherichia* 710834 (not detected in the original soil) reached 2.5%. Under MSF treatment, the abundance of nitrifying bacteria significantly increased from 0.6% initially to 1.2% at maturity, which was higher than the original proportion of nitrifying bacteria in the

soil at 0.5%. Additionally, the saprophytic bacteria *Faecalibacterium*, which was at 0.2% in the original soil, increased to 0.8% by R6. In the CK treatment, the dominant bacteria in the original soil, *Pedospaera* (12.3%), decreased to 3.1%, while the proportion of competitive bacteria such as *Romboutsia* B increased to 7.9% (Fig. 5).

By analyzing the soil samples at VT and R6 stages, we determined the α -diversity distribution patterns of different treatment groups and growth stages (Fig. 6(a)). This showed that in the PE film treatment group, the diversity index of the VT stage samples was overall high, indicating that the richness and evenness of microbial communities were higher at that stage. However, the diversity had significantly decreased by R6, with the Chao1 index of PE(R6)3 for PE at R6 decreasing by 93% compared to VT. The Shannon index also had the lowest values, indicating that the PE film may inhibit the stability of microbial communities leading up the harvest period.

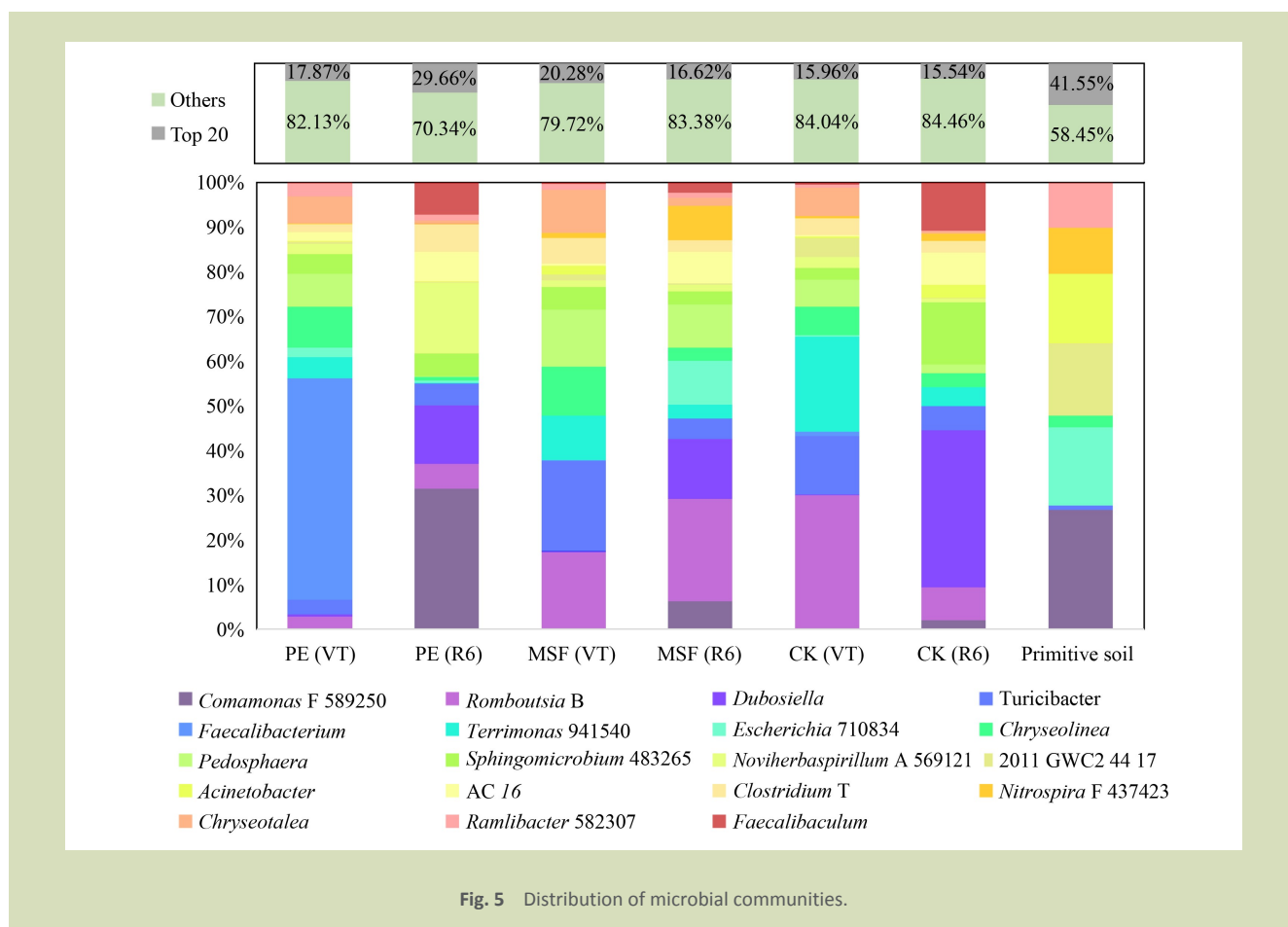


Fig. 5 Distribution of microbial communities.

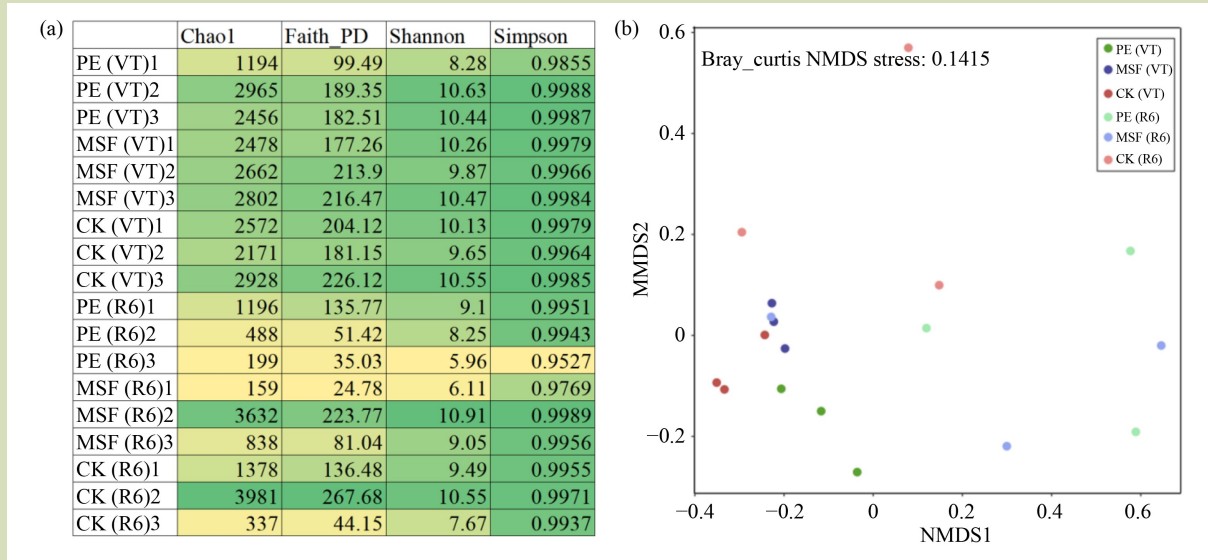


Fig. 6 (a) α -Diversity and (b) β -diversity analyses.

In the MSF treatment group, the VT stage samples showed stability characteristics, with Chao1 index ranging from 2478 to 2802, and high evenness indicated by Shannon and Simpson indices. However, extreme differentiation in diversity was evident by R6: the Chao1 index for MSF at R6 was the highest in the experiment, with Shannon index reaching a peak, while the Chao1 and Shannon indices for MSF(R6)1 of MSF at R6 group dropped to the lowest levels in the experiment, and the Chao1 index for MSF(R6)3 had decreased by about 70% from VT, indicating significant variability in the long term impact of MSF on microbial communities.

In CK, the Chao1 and Faith PD indices at the VT stage showed moderately high richness. However, the diversity had fluctuated drastically by R6. The Chao1 index of CK(R6)2 for CK at R6 was the highest in the experiment, with Shannon and Simpson indices reaching the highest values recorded in the experiment. On the other hand, the Chao1 and Faith PD indices of CK(R6)3 for CK at R6 decreased by 88% from VT. This indicates that microbial communities under natural conditions may have significant imbalance due to environmental fluctuations. Additionally, the positive correlation between Shannon and Simpson indices further verifies the robustness of the evenness indicator.

Regulation of diversity by mulch type. In the PE film group from VT to R6, the diversity indices of all samples consistently

decreased. The Chao1 index of PE(R6)3 for PE at R6 was the lowest in the experiment, indicating that PE film may inhibit the later recovery of microbial communities through physical barrier effects or chemical leaching. With MSF, extreme polarization had occurred by R6, indicating that OM input may promote the proliferation of some microorganisms but is limited by uneven resource distribution or local environmental differences. In CK, the Chao1 index of CK(R6)2 for CK at R6 was significantly higher than the peak value of other treatment groups, with Shannon and Simpson indices reaching global highs. This indicates that some undisturbed microbial communities under natural conditions may have stronger competitive advantages in ecological niches.

From the perspective of the impact of growth period on diversity, it was observed that the average diversity at R6 of all treatment groups was lower than at VT. This difference may be linked to reduced root exudates, intensified nutrient competition, or changes in the soil microenvironment at the crop maturity stage. The variability among replicates within a treatment may be due to the spatial heterogeneity of the field microenvironment or local variations in organic mulch degradation rates. Notably, the Chao1 index gap in CK reached 11.8 times, indicating a significant imbalance in microbial community distribution under natural conditions. Additionally, the trend differences between Chao1 and Faith PD indices may reflect the low sensitivity of α -diversity to rare

species. Overall, mulch type and growth period influenced soil microbial α -diversity, but specific environmental driving mechanisms require multidimensional verification combining soil physicochemical properties, microbial functional activity, and metagenomic data.

Also, the analysis of β -diversity indicated that VT and R6 samples from the PE film group had significant spatial separation, indicating a notable restructuring of the community structure in the later stages of crop growth (Fig. 6(b)). In contrast, the samples from the MSF treatment group were relatively concentrated across the two stages, indicating that it could maintain community stability through continuous nutrient release. CK had the least variation in community structure from VT to R6, reflecting the continuity of microbial succession under natural conditions.

3.4 Relevance analysis

By constructing a soil microbial interaction network (Fig. 7(a), which shows Spearman analysis data can be found in the supporting materials), the core role of key groups in maintaining ecosystem stability was revealed. Network analysis showed that microorganisms such as *Terrimonas* 941540, *Chryseolinea*, AC 16, *Gynuricola*, *Comamonas* F 589250 and OLB9 had high node degree, indicating that they have direct or indirect connections with many other microorganisms in the network. These microorganisms may, therefore, be important in the network, with extensive interactions. Low-degree microorganisms such as *Romboutsia* B, *Faecalibacterium*, *Sphingomicrobium* 483265 and *Kordiimonas* 482971 have lower node degree, indicating fewer connections in the network, possibly being at the network periphery with limited interactions with other microorganisms. Central microorganisms like *Terrimonas* 941540, *Chryseolinea* and OLB9 had high closeness centrality values. These microorganisms may, therefore, be important in information transmission within the network. Peripheral microorganisms, such as *Romboutsia* B and *Sphingomicrobium* 483265, have both low degree and low closeness centrality values, indicating they occupy peripheral positions in the network with lower interaction and information transmission efficiency. Bridge microorganisms such as *Herbaspirillum*, *Chryseotalea*, *Solirubrobacter* and UBA5216 had high betweenness centrality values, indicating they act as bridges connecting different microbial groups, which is crucial for network connectivity and information transmission. The suppression of these

microorganisms may lead to significant changes in the network structure. Non-bridge microorganisms, such as *Romboutsia* B, *Faecalibacterium* and *Terrimonas* 941540 (despite their high degree and closeness centrality, have low betweenness centrality), do not serve as major bridge microorganisms in the network. Instead, they form small clusters or local networks with surrounding microorganisms.

When conducting a detailed analysis of the correlation between soil microorganisms and soil N, P, K and OM, we used the Spearman rank correlation coefficient (r) and corresponding p -values as evaluation indicators (Fig. 7(b)). By interpreting these data using a heat map, we could presumptively deduce the interaction patterns between microbial species and soil nutrient factors.

First, for most microbial species, such as RSA9, *Streptomyces* G 395345 and AC 16, they had significant negative correlations with P, K, N and OM ($r = -0.87$ and -0.12), and these negative correlations were highly significant statistically. This indicates that these microorganisms may be reducing P, K, N, and OM levels in the soil. This reduction could be attributed to their metabolic activities, which may involve the absorption, utilization, transformation or loss of these nutrients.

However, there are also some microbial species, such as GCA 900066495, GCA 2753745 and J137, that had positive correlations with P, K, N and OM ($r = 0.48$ to 0.78 , $p = 0.01$ – 0.05). These microorganisms may help in maintaining or increasing soil nutrients, possibly by promoting nutrient cycling, reducing loss or directly contributing OM.

Particularly noteworthy are microorganisms such as *Comamonas* F 589250, *Nonomuraea* 394555 and *Solirubrobacter*, which had extremely strong correlations with K, N and OM (r near -1 , $p < 0.001$). This strong negative correlation may indicate specific roles of these microorganisms in soil nutrient cycling, such as efficient nutrient absorbers or transformers. Conversely, microorganisms such as B1, *Pseudomethylobacillus* and *Pedobacter* 881387 showed significant positive correlations with K, N and OM (r between 0.75 and 0.85 , $p < 0.05$), indicating that they may be important for maintaining or enhancing soil nutrient levels.

In summary, the correlation patterns between soil microorganisms and soil N, P, K and OM was found to be diverse and complex. Different microbial species have varying

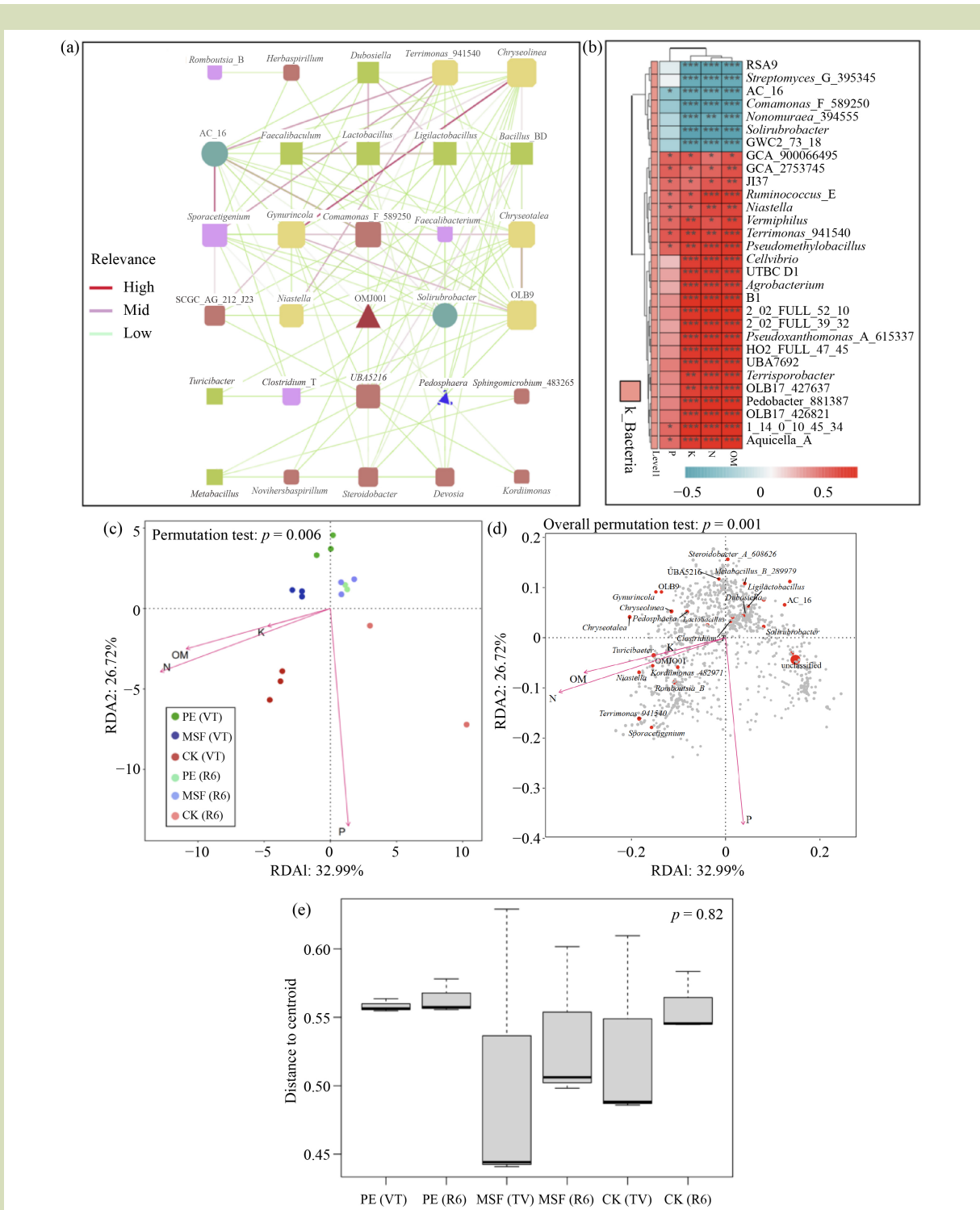


Fig. 7 (a) Microbial interaction network, (b) correlation analysis, (c) redundancy analysis (RDA) between films and nutrients, (d) RDA between soil microorganisms and nutrients, and (e) PERMANOVA dispersion analysis.

impacts on soil nutrients, with some promoting nutrient retention and increase (positive correlation microorganisms) and others leading to nutrient reduction (negative correlation microorganisms). These findings are significant for understanding the role of soil microorganisms in nutrient cycling, optimizing soil management practices and improving soil fertility. Future research should further examine the specific mechanisms of these microorganisms and their potential applications in agricultural ecosystems.

RDA was performed to investigate the relationship between soil samples and soil nutrients (N, P, K and OM) during the growth of silage maize under different film treatments (Fig. 7(c)). This analysis indicated significant impacts of both PE film and MSF on soil nutrients at different growth stages (VT and R6). Specifically, soil samples from under PE (VT) and PE (R6) film were clearly separated on RDA1, indicating significant differences in soil nutrient composition. Similarly, MSF (VT) and MSF (R6) treatments also separated on RDA1, further confirming the impact of different growth stages on soil nutrients. Notably, the separation between CK (VT) and CK (R6) treatments was relatively small, indicating that without film, soil nutrient changes might be more influenced by the growth stage than the planting method. The distribution on RDA2 revealed more details, such as the positive values of PE (VT) on RDA2, which might be related to higher P and K content, while the negative values of PE (R6) might be associated with nutrient consumption at R6. Overall, in the nutrient RDA distribution, P had the highest positive loading on the RDA1, indicating it as a key driver distinguishing PE and MSF treatments. The CK, with a higher association with nutrient factors, was distributed at the negative end of RDA2, indicating significant nutrient loss in soils without film treatment.

We further used RDA to examine the relationship between soil microbial communities and soil nutrients (Fig. 7(d)). The results showed that soil microbial communities had significant distribution differences on RDA1, which were closely related to soil nutrient content. The RDA revealed significant relationships between microbial features and environmental factors. The overall permutation test had a *p* of 0.001, indicating a strong association. The first two axes (RDA1 and RDA2) explained 33% and 27% of the variance, respectively. Notable features such as *Steroidobacter* A 608626, *Faecalibacterium*, and *Chryseolinea* F 598240 were significantly correlated with OM and N. For example, certain microbial species (e.g., *W Chloroflexi* 9 and *Desulfotruncus*) were highly enriched in the positive value regions of RDA1, which typically correspond to higher P and K content, indicating that these microbes may benefit from P- and K-rich soil environments. In contrast, other microbes (e.g., *Sporocytophaga* and *Adhaeribacter*) were distributed in the negative value regions of RDA1, which often had lower OM content, indicating that these microbes may be more adapted to barren soil conditions.

The PERMDISP analysis gave a *p*-value of 0.82, indicating no significant difference in intergroup dispersion. Therefore the separation displayed by RDA axes can be interpreted as the effect of treatment on the central position of community composition (Fig. 7(e)).

3.5 Life cycle assessment

The life cycle assessment (Fig. 8) indicated that under the planting conditions with PE film, as the amount of fertilizer decreases from 100% to 70%, the environmental impact of silage maize production changed. Specifically, the impact on global warming (terrestrial ecosystems) slightly increased,

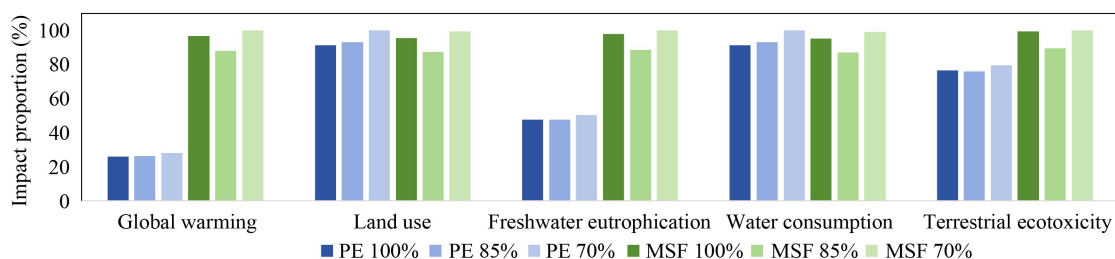


Fig. 8 Life cycle assessment.

while other categories such as freshwater eutrophication remained relatively stable. This phenomenon may suggest that merely reducing the amount of fertilizer may not significantly lower the overall environmental burden under the current agricultural technology system, and may even slightly increase the environmental impact per unit product due to potential changes in yield or efficiency. In contrast, life cycle assessment of the MSF group had a similar trend in the environmental impact change with reduced fertilizer input as the PE film group, but all indicators are significantly higher in absolute terms.

Comparing the types of films, MSF had higher environmental impact values at all fertilizer application rates, with particularly significant differences, indicating that the environmental performance of MSF may be limited by its own characteristics (e.g., emissions during the degradation process or resource consumption). For example, the freshwater eutrophication potential of MSF was twofold that of PE film, and the global warming indicator was even more than threefold higher. This disparity may stem from factors such as implicit carbon emissions during the production or use stages of MSF, energy consumption in raw material processing, or changes in land use. It is noteworthy that the reduction in fertilizer input (70% fertilizer application group) in MSF did not effectively offset its high environmental costs, indicating that the type of mulch may be a more critical factor than the amount of fertilizer applied.

4 Discussion

4.1 Stage-specific regulation by soil nutrients on crop growth under the influence of MSF and PE films

4.1.1 Stage-specific nutrient driving characteristics of dry matter accumulation in silage maize under MSF and PE films

This study integrates the spatiotemporal dynamic data of dry matter weight of silage maize throughout the entire growth period with soil P, K, OM and total N, revealing the growth demand-nutrient supply response mechanism between crop growth and soil nutrients. From the seedling stage to the jointing stage (30–60 d after sowing), the dry matter weight of individual plants in all treatment groups was in a slow accumulation phase, while the soil total N content and OM content had a significant positive correlation. This phenomenon is consistent with the results of Hu et al.^[25] where

organic fertilizer prepared using biochar and pig manure promoted crop growth and dry matter accumulation. That is, soil OM enhances the N mineralization process driven by microorganisms (e.g., increased activity of ammonifying bacteria)^[26], improving N availability, thereby accelerating the synthesis and transport of early photosynthates in crops^[27].

Notably, when fertilizer application rate was reduced by 30%, there was no significant decrease in dry matter accumulation in the MSF treatment group, whereas it decreased by an average of 18.6 g per plant in the PE film group. This may be mainly attributed to the promotion of crop root development and the improvement of the soil microenvironment by the MSF. The MSF may enhance long-term and stable root growth by improving the water retention and nutrient-holding capacity of soil and providing a sustained nutrient release, whereas the insufficient nutrient-holding capacity of PE film restricts root development when less fertilizer is applied^[28]. Existing studies have shown that organic mulch can regulate soil moisture and temperature conditions and promote the formation of humus-mineral complexes, thus slowing down the rate of N leaching^[29,30]. Therefore, the use of MSF to reduce fertilizer use and increase efficiency in this study further supports the feasibility of organic alternatives and green development strategies in agriculture.

4.1.2 Mechanism of enhanced dry matter accumulation during grain-fill driven by P and K

After entering the grain filling stage (90–120 d after sowing), the dry matter accumulation rate of silage maize significantly increases. At this time, the driving effects of soil P and K gradually replace N as the dominant factors. The available P content in the soil of the MSF group was 11.2% higher than that of the PE film group during the grain filling stage and is significantly positively correlated with the dry matter accumulation rate. This result is consistent with classical crop physiology theory: P, as a core element in ATP and nucleic acid synthesis, directly affects the starch synthesis efficiency in grains during the grain filling period^[31,32]. In addition, the K content in the MSF group was higher at R6 compared to the PE group. This may enhance turgor pressure and the transportation of photosynthetic products (e.g., sucrose-K co-transport), thereby promoting the dry matter accumulation rate to the grain and increasing crop yield^[33].

It is particularly noteworthy that with 30% less fertilizer, the

dry matter weight of the MSF group remained at a relatively high level, while that of the PE film group significantly decreased. This could be due to two possible reasons. (1) After the application of MSF, an OM layer is formed on the soil surface, which may form organic-mineral complexes with phosphate in the fertilizer^[34,35], reducing the solidification and leaching loss of P. Even though the total amount of P decreases, the increase in the proportion of Olsen-P does not cause a significant P deficiency in crop nutrient supply. (2) Some K-solubilizing bacteria (e.g., *Paenibacillus mucilaginosus*) are naturally present in cow dung^[36], and they are not killed during the preparation of MSF. After rehydration during field application, they become part of the soil microbial community. Some organic acids secreted by these microorganisms can activate mineral K in soil and fertilizer, thereby enhancing the efficiency of K fertilizer^[37].

4.1.3 Sustainability driven by MSF nutrients and its potential for ecological regulation

This study found that the application of MSF not only enhanced soil nutrient availability but also increased the complexity of microbial networks (e.g., increasing the soil bacterial Shannon index by 0.42–0.92, VT and R6 stages respectively). This effect can prolong the nutrient cycling of nutrients within microbial communities, thereby achieving slow-release nutrient supply through microbial metabolic processes^[38]. The soil microbial abundance under MSF was higher than that of the PE groups, and exhibited a strong positive correlation with the dry matter accumulation of silage maize. Previous studies have indicated that the increase in soil microbial abundance implies a higher potential for nutrient turnover^[39,40]. For example, arbuscular mycorrhizal fungi can expand the range of P absorption for crops through mycelial networks. Also, this may also lead to enhanced soil enzyme activity under MSF coverage, and achieving the regulation of soil nutrients.

Based on the regulatory capacity of MSF toward soil microorganisms, we propose a demand-responsive nutrient management framework from an ecological regulation perspective: (1) during the seedling stage, prioritize the use of organic improvement measures (MSF in this study) to enhance N availability, to maximizing root biomass (thus building crop absorption potential); and (2) in the middle and late stages, combine microbial inoculation techniques (e.g., phosphate-solubilizing bacteria) to selectively activate soil P and K pools, achieving the goal of reducing mineral fertilizer input and

improving fertilizer use efficiency. This strategy aligns with the concept of “Soil management based on constant” proposed by Lehmann et al.^[41], which aims to achieve efficient utilization of nutrient resources through artificial regulation of soil biological processes.

4.2 Differential regulation of soil microbial communities by PE films and MSF

4.2.1 Regulatory mechanisms for soil microbial communities of PE film and MSF

PE film and MSF fundamentally differ in their regulatory pathways for soil microbial communities, a phenomenon that may stem from their distinct methods of supplying soil resources. MSF continuously release soluble OM, N, P and other nutrients, providing a rich substrate basis for the proliferation of heterotrophic microorganisms (e.g., *Proteobacteria* and *Actinobacteria*)^[42]. These microorganisms typically have high metabolic activity and can rapidly decompose complex OM (e.g., cellulose and lignin), thereby forming a positive feedback loop involving nutrient input-functional bacteria activation and enhancing fertilizer use efficiency, as confirmed in the study by Sokol et al.^[43]. This mechanism may be related to the OM-driven microbial chemotaxis: polysaccharides released during the degradation of MSF may act as chemical signals, preferentially attracting functional bacteria with degradation capabilities (e.g., *Chryseolinea*), while inhibiting the competition of oligotrophic groups (e.g., *Acidobacteria*)^[44]. Additionally, MSF does not block oxygen supply to the soil surface, favoring the metabolic activities of aerobic microorganisms (e.g., certain fungi and aerobic bacteria) and the nutrient transformation processes they dominate.

In contrast, PE film relatively lacks bioavailable carbon sources, and its covering effect mainly alters the soil microenvironment through physical barriers (e.g., temperature increase and humidity stabilization), indirectly selecting for drought-tolerant or oligotrophic microorganisms (e.g., *Acinetobacter*)^[45,46]. These microorganisms typically have low metabolic rates and efficient resource capture abilities, possibly adapting to resource-limited environments by secreting extracellular polymers or forming biofilms. Notably, the significant enrichment of nitrification-related microorganisms (e.g., *Nitrospira*) in both PE film and MSF groups suggests that film coverage may accelerate the ammonia oxidation process^[47]. However, the scarcity of carbon sources may

disrupt the metabolic balance of denitrifying microorganisms, leading to simplification N cycling pathways^[48], this phenomenon was less pronounced in MSF.

In summary, PE film promotes beneficial bacterial activity in the short-term, but long-term oxygen deficiency and residual pollution inhibit microbial diversity; conversely, MSF restores permeability through degradation and releases organic carbon sources, supporting bacterial proliferation and ecological balance, thereby making it more soil-friendly.

4.2.2 Differential regulation on the differentiation and ecological risks of microbial functional networks

The regulatory effect of MSF on microorganisms can optimize soil microbial functional networks. This effect stems from the reconstruction of microbial interactions as a result of OM input. For example, the synergistic proliferation of nitrogen-fixing and cellulose-degrading bacteria may form stable functional modules through metabolic complementation. The latter decomposes plant residues to release N sources, while the former could fix them into bioavailable forms, thereby reducing dependence on exogenous N fertilizers^[49,50].

This mutualistic relationship is further reflected in the microbial co-occurrence network, where the connectivity of key nodes (e.g., *Terrimonas*) in the MSF group is significantly higher than that in the PE film groups, indicating that its community has higher functional redundancy and resistance to disturbance^[51]. However, this positive regulation may also be accompanied by potential risks: incompletely degraded lignin fragments in MSF may selectively enrich specific taxa (e.g., *Actinobacteria*), leading to functional convergence of microbial communities, potentially weakening the degradation potential for novel pollutants in the long-term^[52]. The negative impact of PE film on microbial communities is more likely to be reflected in the niche compression effect caused by soil degradation. Microplastic residues in soil not only damage microbial cell structures through physical injury (e.g., decreased membrane integrity of Gram-negative bacteria)^[53], but the hydrophobic pollutants (e.g., polycyclic aromatic hydrocarbons) adsorbed on their surface may also interfere with microbial electron transfer chains, inhibiting the activity of key enzymes^[54]. More concerningly, microplastic may act as artificial carriers to promote the spread of pathogenic bacteria (e.g., *Escherichia*)^[55] and antibiotic resistance genes^[56]. Such effects may lead to functional degradation of soil microbial communities, resulting in a shift from multidimensional

nutrient cycling to defensive metabolism dominated by survival pressure responses.

MSF has been shown to optimize functional networks and improve resistance to disturbance through the reconstruction of microbial interactions, such as metabolic complementation between nitrogen-fixing bacteria and cellulose-degrading bacteria. However, there is a potential risk of functional convergence. However, PE film induces niche compression through microplastic residues, which cause physical damage. This can lead to dominance of defensive metabolism and functional degradation of microbial communities.

4.2.3 Connection between mulch and soil ecosystem sustainability

From the perspective of ecological succession, MSF regulates soil microbial communities in a manner closer to native ecological succession patterns and OM application accelerates succession dynamics. For example, the easily decomposable carbon sources released by MSF during the early growth stages of silage maize stimulate the rapid proliferation of r-strategists (e.g., Proteobacteria). These microorganisms quickly consume easily degradable substrates, such as glucose and amino acids, through rapid growth and short life cycles, thereby swiftly establishing population dominance. In the later stages, the higher soil oxygen content and improved soil physicochemical structure compared to PE promote the development of soil microorganisms that more closely resemble those in the original ecosystem. This dynamic succession may maintain the diversity balance of the community^[57].

However, the continuous application of manure in agricultural practices may lead to excessive proliferation of certain functional bacteria (e.g., enzyme-producing *Bacillus*)^[58], disrupting the original interspecies competitive relationships and reducing community stability. We observed this phenomenon, such as the rapid proliferation of *Acinetobacter* and *Escherichia* in MSF treatment. This phenomenon has been reported in long-term fixed-point experiments and may be related to the threshold effect of soil resource use efficiency^[59,60]. The application of MSF, by reducing the number and activity of functional bacteria in cow manure, may alleviate this phenomenon. Long-term application of PE film, may induce a community reconstruction dominated by abiotic stress. Its coverage blocks the natural material exchange between plants and soil, forcing microorganisms to rely on

native soil OM for metabolism. This unsustainable consumption may lead to the decline of key functional bacterial groups^[61]. Especially in semiarid regions, the intensified water and heat stress caused by PE film may synergize with microplastic toxicity to induce a dormancy strategy in microbial communities^[62], numerous microorganisms enter metabolic stagnation.

4.3 Environmental impact of manure-based slurry and polyethylene films

The high environmental impact of MSF determined by the life cycle assessment mainly stems from its raw material characteristics and degradation process. First, as an organic raw material, treatment processing of cow manure can result in significant CH₄ and N₂O emissions, both of which have a global warming potential far greater than CO₂, directly elevating the global warming-related indicators^[63]. Secondly, the degradation of MSF may be accompanied by the release of nutrients such as N and P, exacerbating freshwater eutrophication and terrestrial acidification problems, whereas the inert nature of PE film results in lesser impacts in these indicators^[64]. In contrast, although the industrial production efficiency and high stability of PE film actually offer advantages in certain environmental indicators. Notably, industrial production has environmental benefits through high efficiency and stability, primarily achieved via yield-based numerator reductions.

It is evident that to reduce the environmental impact of MSF, optimization is needed in three areas: raw material treatment, production process and field management. In terms of raw material treatment, pre-fermentation or the addition of inhibitors (e.g., nitrification inhibitors) can reduce CH₄ and N₂O emissions, thereby reducing environmental impact. In the production process, reducing energy consumption in extrusion and grinding processes and improving molding processes (e.g., adding natural adhesives) to enhance the mechanical strength of the MSF can effectively reduce environmental impact by

decreasing the amount of film used per unit area. From the perspective of field management, precision fertilization techniques should be combined to avoid nutrient overload caused by the simultaneous application of MSF, and organic or mineral fertilizers. These improvements can specifically address the shortcomings of MSF in terms of greenhouse gas emissions and eutrophication but the actual effects still need to be investigated.

In the long-term, if MSF can be integrated into the circular agriculture system, it will achieve a closed loop from waste to resource, ultimately balancing environmental needs and agricultural productivity. However, its environmental impact cannot be ignored, and both its preparation and field application techniques require further improvement to match the effectiveness of PE film while reducing the amount used in the field.

5 Conclusions

This study demonstrated that manure-based soil film (MSF) can effectively enhance fertilizer use efficiency through microbially-mediated nutrient activation mechanisms, showing potential for reducing mineral fertilizer input. There were three specific research findings. (1) When producing the same quantity of silage maize, MSF could reduce mineral fertilizer by 30%. (2) Different films (PE film and MSF) significantly alter soil microbial community structure and diversity at various crop growth stages. Generally, PE film inhibits microbial stability, while MSF promotes the proliferation of specific functional bacteria, affecting the metabolic processes of soil nutrients. (3) MSF has a higher environmental impact than PE film.

Application of MSF directly supports China's policy to reduce fertilizer input by enabling significant nitrogen substitution while maintaining crop yields. Its integration into current farming systems offers a practical way to achieve sustainable agricultural goals.

Supplementary materials

The online version of this article at <http://doi.org/10.15302/J-FASE-2026671> contains supplementary material (Fig. S1).

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Compliance with ethics guidelines

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