

The effects of long-term residues of poly(butylene adipate-co-terephthalate) mulch film on soil microbial communities and tobacco growth

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Abstract: Long-term use of conventional polyethylene mulch film (PE) causes white pollution, whereas biodegradable poly(butylene adipate-co-terephthalate) (PBAT) mulch film can serve as an alternative to PE mulch film. However, research on the effects of long-term residual PBAT mulch film on soil microbial communities and tobacco growth remains limited. In this five-year field experiment, we compared the annual PBAT mulch treatment with a single-dose PE film treatment and a non-mulching blank control (N) to explore their effects on soil microbial communities and tobacco growth. Scanning electron microscope results indicated that the surface of the PBAT mulch film was relatively rough and exhibited varying degrees of cracking and bulging. Further chemical structural analysis revealed that the ester bonds in PBAT broke, the alkyl chains degraded, and oxygen-containing functional groups were generated. Compared with the other treatments, the long-term persistence of PBAT residues improved the soil's physicochemical properties and significantly increased the Shannon and Chao1 indices for bacteria and fungi. At the phylum level, the PBAT treatment increased the relative abundances of Proteobacteria, Actinobacteriota, and Ascomycota, whilst at the genus level, it led to the enrichment of bacteria such as *Sphingomonas*. Within the fungal community, the proportion of saprophytic fungi increased, whilst the relative abundance of the pathogenic *Fusarium* decreased. Functional prediction indicated that, in the later stages of the PBAT treatment, the relative abundance of metabolic pathways involved in the degradation of exogenous substances increased, whilst saprophytic fungal functions were enhanced and plant-pathogenic functions were suppressed. Furthermore, long-term persistence of PBAT residues had a positive effect on the plant height, stem diameter and maximum leaf area of tobacco. Overall, our results suggest that the long-term persistence of PBAT residues has positive effects on soil microbial communities and tobacco growth.

Keywords: PBAT biodegradable mulch film residues; PE mulch film residues; tobacco agronomic traits; soil microbial community; long-term effects

Since its first application in agricultural production in 1948, plastic mulch film has significantly improved agricultural productivity and generated enormous economic benefits (Zhang et al. 2022). It

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has also been widely adopted worldwide, particularly in China, Europe, and the Mediterranean Basin (Steinmetz et al. 2016). With the development of the chemical industry, the manufacturing costs of plastic mulch film have gradually decreased, resulting in increased agricultural application intensity (Zhang et al. 2025). Polyethylene (PE) is the most widely used traditional mulch material. However, PE mulch film degrades extremely slowly under natural conditions, often taking decades or even centuries to decompose fully (Steinmetz et al. 2016). Plastic mulch pollution in Chinese farmland is primarily attributed to thin commercial mulch film, which is prone to ageing and fragmentation during field use. These fragments mix with sand, crop straw, and other impurities in the 0–20 cm topsoil layer, making them difficult to recover; moreover, the economic costs of mulch film recovery are extremely high (Xie et al. 2023). Long-term, large-scale mulch application has substantially increased the amount of plastic-mulch residues in farmland (Dai et al. 2025). The total amount of plastic mulch pollution in China had reached 550 800 metric tons (Zhang et al. 2020), causing serious damage to soil structure and ecological environment. To alleviate such pollution, new alternatives to traditional plastic mulch materials are urgently required.

Poly(butylene adipate-co-terephthalate) (PBAT) mulch film is primarily manufactured from biodegradable materials, which can ultimately be decomposed by soil microbial communities into CO₂ and H₂O. Additionally, PBAT mulch film delivers performance comparable to polyethylene (PE) mulch film in soil warming, moisture conservation and weed suppression (Sun et al. 2018), making it widely regarded as a viable alternative for mitigating pollution from PE-derived mulch film residues and supporting sustainable agriculture. However, environmental safety concerns regarding the application of PBAT mulch film remain controversial (Gao et al. 2025). Numerous studies indicate that PBAT mulch film does not fully biodegrade over short-term periods, and its degradation rate is modulated by environmental factors, including soil temperature, soil moisture, pH, and microbial activity (Coutris et al. 2025). Under low-temperature or drought conditions, the degradation process may extend for months or longer, during which microplastics may persist in the soil (Wang et al. 2025), disrupt soil ecological functions and affect plant growth (Fang et al. 2025).

Current research primarily focuses on the effects of degradation of PBAT mulch film on soil nutrient

levels (Koskei et al. 2021), microbial diversity (Han et al. 2024), and crop biomass (Carneiro et al. 2025). Zhang et al. (2022) found that prolonged application of biodegradable mulch film increased the total nitrogen, available phosphorus, and potassium contents in the soil and enhanced soil microbial, urease, and catalase activities. Gao et al. (2025) reported from pot experiments that long-term application of PBAT mulch film enriches beneficial soil bacteria and suppresses soil pathogens, thereby improving soil quality and crop growth. However, other studies have reported yield reductions in multiple crops over short periods (one growing season), with crops such as corn (Yu et al. 2023) and tomatoes (Han et al. 2025) exhibiting delayed root development and reduced biomass under PBAT treatment. Most of these studies were conducted under specific pot conditions involving short durations and high application rates (Bandopadhyay et al. 2018). Although limited long-term field studies are available, systematic investigations into the responses of soil ecosystems and tobacco growth to the retention of continuous PBAT residual film remain scarce.

Tobacco is an important cash crop in China, and plastic mulching constitutes a core technique for standardised tobacco cultivation. Nevertheless, few continuous field studies have systematically elucidated the long-term effects of residual PBAT mulch film on soil ecological environments and tobacco growth in tobacco fields. Accordingly, based on a five-year continuous field experiment, this study investigated variations in tobacco agronomic traits, soil physico-chemical properties, soil microbial communities, and residual film characteristics during long-term retention of PBAT mulch film. The objectives were to clarify the comprehensive responses of soil environments and tobacco growth to aged PBAT film residues and to address the research gap regarding long-term effects of residual PBAT film in tobacco cropping systems. The findings provide a theoretical basis for the rational application, field management, and sustainable utilisation of biodegradable PBAT mulch film in tobacco production.

MATERIAL AND METHODS

Experimental sites. A five-year field experiment was conducted from March 2020 to September 2024 in Longchang Town, Fuquan City, Guizhou Province, China (26°24'N, 107°30'E). The site is located in the humid central region of a subtropical monsoon cli-

mate zone, at an elevation of 685 m a.s.l. The mean annual temperature is approximately 14 °C, with an annual rainfall of 1 033–1 220 mm and a frost-free period of 245–278 days. The soil is yellow clay loam (Yellow soil) in the Chinese Soil Classification System and classified as Acrisol in the FAO WRB. This site had no prior plastic input history, and no obvious plastic residues were detected before the experiment.

Experimental design. The experiment consisted of five consecutive years of field trials with three treatments, each replicated three times, giving a total of nine experimental plots. Buffer zones were established between plots. Tobacco was planted as the monoculture crop in the same plots for five consecutive tobacco-growing seasons. Tobacco was planted in single rows with a row spacing of 110 cm and a plant spacing of 55 cm. Field management followed the standard local practices for high-quality tobacco production. The specific treatments were as follows: (1) PBAT treatment: PBAT biodegradable mulch film was incorporated into the soil at a rate of 8 kg per 667 m² annually, simulating the consecutive application mode in production; (2) PE treatment: PE film was applied at the same rate only in 2020, with no further supplementation in subsequent years (given the recalcitrant and persistent nature of PE residues, continuous annual application would result in excessively high residual levels); (3) N treatment: blank control without film mulching or residue input throughout the experiment.

The differential application regimes – annual PBAT application *versus* single PE application – were scientifically justified based on the distinct degradation characteristics of the two film materials and represent an inherent limitation of the experimental design. Consequently, subsequent analyses focus on the long-term effects of PBAT rather than direct comparisons of film performance.

Sample collection. Film samples were collected one day after topping the tobacco plants in 2024. Three to five film pieces were randomly collected from each plot, rinsed in clean water to remove surface soil, and placed in beakers containing 75% ethanol for 30 min. The samples were then washed with deionised water, air-dried naturally, and stored in paper bags for further analysis.

To investigate the temporal succession of soil microbial communities under long-term treatments, soil samples were collected 1 day after tobacco topping at the end of one cropping season of film application during the initial (2020), middle (2022), and final

(2024) stages of the experiment. Using a five-point sampling method, soil samples were collected from the 0–20 cm plough layer of each plot. The five subsamples from each plot were thoroughly mixed to form one composite sample, from which stones, plant residues, and other debris were removed, and the sample was reduced by quartering. Each composite sample was divided into two portions: one was air-dried and sieved for determining soil physico-chemical properties, and the other was stored at –80 °C for high-throughput sequencing of soil microbial communities.

Degradation performance analysis. Surface micromorphology was examined using field emission scanning electron microscopy (SEM, Zeiss Sigma 300, Cambridge, UK) with a spatial resolution of 7.3 mm and an accelerating voltage of 5 kV. Film samples were mounted on stages, sputter-coated with gold, and observed at 1 000× and 10 000× magnifications to examine microstructural changes on the film surfaces. Fourier-transform infrared spectroscopy (FTIR, Shimadzu IRTracer-100, Tokyo, Japan) was used to record spectra in the range of 4 000–400/cm to analyse the functional group structures of the film samples.

Soil physicochemical properties and soil microbial analysis. Soil total nitrogen (TN) and alkali-hydrolysed nitrogen (AN) were determined by the Kjeldahl method and the alkaline hydrolysis diffusion method, respectively. Total phosphorus (TP) was measured using the molybdenum-antimony-scandium colourimetric method, whereas available phosphorus (AP) was analysed with a continuous flow analyser. Total potassium (TK) and available potassium (AK) were determined following sodium hydroxide fusion and ammonium acetate extraction, respectively. Soil organic carbon (SOC) was quantified by the K₂Cr₂O₇-FeSO₄ oxidation method, and soil pH was measured potentiometrically. All analyses were performed in strict accordance with the official Chinese agricultural and forestry standards: LY/T 1228-2015, NY/T 88-1988, NY/T 1121.25-2012, NY/T 87-1988, NY/T 889-2004, NY/T 1121.6-2006, and NY/T 1377-2007.

Total genomic DNA was extracted from each sample using the CTAB method. DNA concentration and integrity were assessed by 1% agarose gel electrophoresis. Based on the measured concentration, the DNA was diluted to 1 ng/μL with sterile water. The V3–V4 region of the bacterial 16S rRNA gene was amplified using primers 341F (5'-CCTAYGGGRBGCASCAG-3') and 806R

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(5'-GGACTACNNGGGTATCTAAT-3'), while the fungal ITS1–5F region was amplified with primers 1737F (5'-GGAAGTAAAAGTCGTAACAAGG-3') and 2043R (5'-GCTGCGTTCTTCATCGATGC-3'). Sequencing was performed on the Illumina NovaSeq platform. Raw reads were merged, filtered, and checked for chimaeras to obtain valid sequences, which were then clustered into operational taxonomic units (OTUs) at a 97% similarity threshold.

Measurement of tobacco growth characteristics. During the topping stage, five tobacco plants were randomly selected from each plot, and their agronomic traits were measured in accordance with the standard YC/T 142-2010. The measured traits included plant height (cm), stem diameter (mm), leaf number, and maximum leaf area (cm²). The values obtained from the five plants were averaged to represent the final data for each plot, which were then used for subsequent statistical analyses.

Data analysis. Data were collated using Microsoft Excel (Microsoft Corporation, Redmond, USA). Two-way analysis of variance (ANOVA) followed by Fisher's

least significant difference (*LSD*) test was performed using IBM SPSS Statistics 26 (IBM Corp., Armonk, USA) to determine statistically significant differences at $P < 0.05$. All figures were generated using Excel, Origin 2024 (OriginLab Corporation, Northampton, USA), GraphPad Prism 8.0 (GraphPad Software, San Diego, USA), and R software. Partial least squares path modelling (PLS-PM) was conducted using the 'plspm' package in R.

RESULTS

Changes in surface micromorphology and chemical structure of mulch films. The morphological characteristics of mulch films are illustrated in Figure 1. After prolonged use, significant changes in surface morphology have occurred, with notable heterogeneity observed across treatment groups. At 1 000× magnification, PBAT mulch film exhibits a rough surface with varying degrees of cracking, whereas PE mulch film displays a smooth surface with only minor micro-particles. At 10 000× magnification,

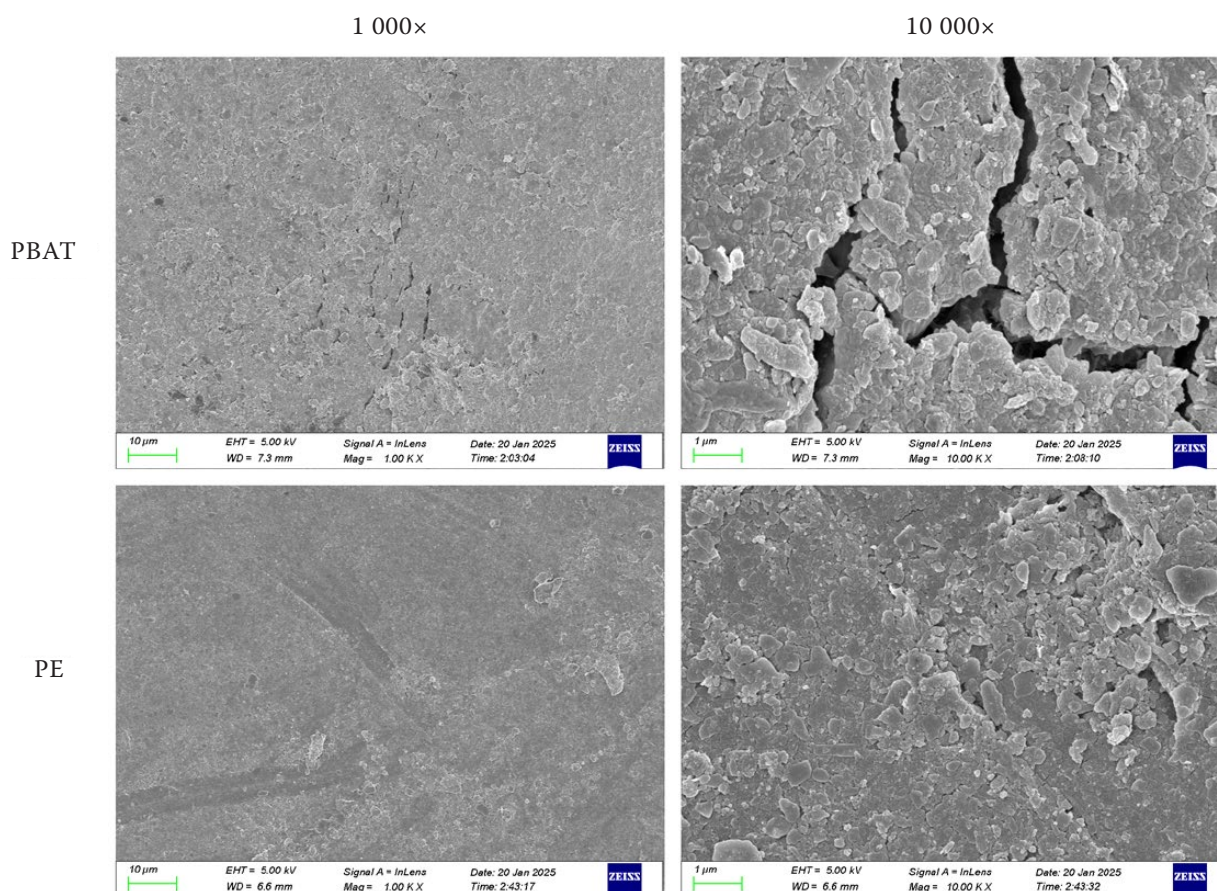


Figure 1. Scanning electron microscopy (SEM) images of poly(butylene adipate-co-terephthalate) (PBAT) and polyethylene mulch film (PE) mulch films

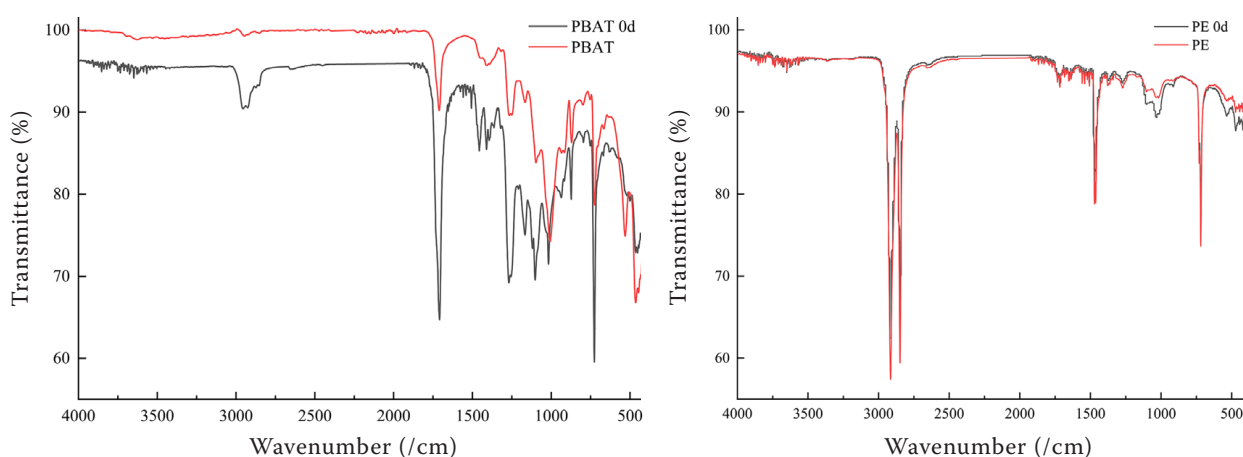


Figure 2. Fourier-transform infrared spectroscopy (FTIR) spectra of poly(butylene adipate-co-terephthalate) (PBAT) and polyethylene (PE) mulch films. PBAT 0d and PE 0d represent the FTIR spectra of untreated PBAT mulch film and PE mulch film, respectively

PBAT film shows visible cracks and protrusions, while PE film exhibits abundant white powdery particles, indicating distinct degradation patterns: PBAT with pronounced degradation, whereas PE with gradual decomposition at a slower rate.

The Fourier transform infrared spectra of PBAT and PE mulch films in the range of 4 000–400/cm are shown in Figure 2. All PBAT mulch film samples exhibited common strong absorption peaks around 2 960/cm, 1 716/cm, 1 245/cm, 1 008/cm, and 723/cm. The peak at 2 960/cm corresponds to the stretching vibrations of $-\text{CH}_3$ and $-\text{CH}_2$; the peak at 1 716/cm represents the symmetric stretching vibration of $\text{C}=\text{O}$ in aromatic esters; the peaks at 1 245/cm and 1 008/cm correspond to the stretching vibrations of the $\text{C}-\text{O}-\text{C}$ group; and the peak near 723/cm indicates the bending vibration of $\text{C}-\text{H}$ bonds in benzene rings. After prolonged use, the $\text{C}=\text{O}$ and $\text{C}-\text{O}-\text{C}$ peaks weakened, the $\text{C}-\text{H}$ peak diminished, and an $\text{O}-\text{H}$ peak emerged. For PE mulch film, the characteristic peaks at 2 917/cm and 2 849/cm correspond to the asymmetric and symmetric stretching vibrations of $\text{C}-\text{H}$ bonds, respectively, with enhanced intensity compared to the pre-treatment level; the peaks at 1 468/cm and 720/cm represent the stretching vibration of $\text{C}=\text{C}$ bonds and the bending vibration of $\text{C}-\text{H}$ bonds, respectively. The complexity of the absorption peaks in PBAT suggests that it may contain more small organic compounds.

Physical and chemical properties of soil. During the experimental period, no significant differences in pH, soil organic carbon (SOC), or total phosphorus (TP) were observed either among treatments or

between years. Total nitrogen (TN), total potassium (TK), and available potassium (AK) increased over time across all treatments, with the most pronounced interannual increase in AK. Alkali-hydrolysed nitrogen (AN) showed a dynamic pattern of initial decrease followed by an increase across all treatments, generally remaining within the normal range of soil nutrient levels. Overall, long-term retention of PBAT residues steadily increased the contents of available potassium, phosphorus, and other available nutrients in the tobacco-planting soil.

Microbial community structure. The alpha diversity indices (Shannon and Chao1) were used to characterise the diversity and richness of bacterial and fungal communities in the samples (Figure 3). Intergroup comparisons revealed significant differences in soil microbial alpha diversity among treatments during the early and middle stages of the experiment. From 2020 to 2022, bacterial and fungal richness and diversity in the PBAT treatment were significantly higher than those in the PE treatment and the blank control, except that the PE treatment showed a temporary advantage in bacterial evenness in 2022. By the final stage in 2024, differences in microbial community diversity among treatments were generally non-significant, although the PBAT treatment still maintained the highest levels. With increasing experimental duration, bacterial richness and evenness increased annually across all treatments. Fungal richness in the PBAT treatment increased steadily year by year, whereas the PE and blank treatments exhibited an initial increase followed by a decrease. Collectively, these results indicate that

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Table 1. Soil physicochemical properties

Index	Year	PBAT	PE	N
Total nitrogen (%)	2020	0.187 ± 0.012 ^{Aa}	0.184 ± 0.013 ^{Ba}	0.177 ± 0.011 ^{Ba}
	2022	0.197 ± 0.013 ^{Aa}	0.198 ± 0.013 ^{Ba}	0.192 ± 0.005 ^{ABa}
	2024	0.221 ± 0.027 ^{Aa}	0.235 ± 0.024 ^{Aa}	0.208 ± 0.019 ^{Aa}
Alkali-hydrolysed nitrogen (mg/kg)	2020	157.82 ± 2.87 ^{Ab}	153.04 ± 3.86 ^{Ab}	172.18 ± 16.97 ^{Aa}
	2022	134.17 ± 3.51 ^{Bb}	154.64 ± 11.33 ^{Aa}	137.63 ± 10.96 ^{Bb}
	2024	151.57 ± 17.45 ^{ABa}	176.97 ± 60.57 ^{Aa}	168.03 ± 14.49 ^{Aa}
Total phosphorus (%)	2020	0.096 ± 0.017 ^{Aa}	0.097 ± 0.011 ^{Aa}	0.095 ± 0.021 ^{Aa}
	2022	0.123 ± 0.027 ^{Aa}	0.148 ± 0.032 ^{Aa}	0.093 ± 0.024 ^{Aa}
	2024	0.102 ± 0.051 ^{Aa}	0.121 ± 0.029 ^{Aa}	0.124 ± 0.009 ^{Aa}
Available phosphorus (mg/kg)	2020	24.45 ± 9.12 ^{Ba}	27.93 ± 3.81 ^{Aa}	28.16 ± 10.32 ^{Ba}
	2022	76.77 ± 18.19 ^{Aa}	74.30 ± 38.83 ^{Aa}	55.97 ± 15.93 ^{Aa}
	2024	63.80 ± 5.21 ^{Aa}	61.23 ± 5.73 ^{Aa}	67.80 ± 9.33 ^{Aa}
Total potassium (%)	2020	1.153 ± 0.134 ^{Ba}	1.177 ± 0.032 ^{Ba}	1.107 ± 0.154 ^{Aa}
	2022	1.380 ± 0.223 ^{ABa}	1.513 ± 0.105 ^{Aa}	1.337 ± 0.352 ^{Aa}
	2024	1.477 ± 0.065 ^{Aa}	1.590 ± 0.225 ^{Aa}	1.407 ± 0.015 ^{Aa}
Available potassium (mg/kg)	2020	400.63 ± 130.15 ^{Ba}	395.26 ± 65.08 ^{Bab}	306.29 ± 140.72 ^{Bb}
	2022	597.83 ± 46.64 ^{ABa}	630.52 ± 128.78 ^{ABa}	393.96 ± 86.59 ^{Bb}
	2024	879.33 ± 359.94 ^{Aa}	781.33 ± 234.59 ^{Aa}	616.00 ± 32.19 ^{Aa}
Soil organic matter (g/kg)	2020	34.07 ± 0.35 ^{Aa}	33.49 ± 1.54 ^{ABa}	33.15 ± 1.34 ^{Aa}
	2022	29.43 ± 1.94 ^{Aa}	27.88 ± 3.15 ^{Ba}	30.20 ± 3.49 ^{Aa}
	2024	35.10 ± 4.52 ^{Aa}	36.70 ± 3.72 ^{Aa}	35.37 ± 2.32 ^{Aa}
pH	2020	5.29 ± 0.23 ^{Aa}	5.51 ± 0.73 ^{Aa}	5.16 ± 0.24 ^{Aa}
	2022	5.12 ± 0.42 ^{Aa}	5.33 ± 0.41 ^{Aa}	5.11 ± 0.20 ^{Aa}
	2024	5.40 ± 0.10 ^{Aa}	5.47 ± 0.47 ^{Aa}	5.37 ± 0.21 ^{Aa}

Different lowercase letters in the table indicate significant differences among treatments within the same year ($P < 0.05$), with means sorted in descending order ($a > b > c$); different uppercase letters indicate significant differences between years under the same treatment ($P < 0.05$), with means sorted in descending order ($A > B > C$). The same notation applies to the following tables. PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

long-term residual PBAT film sustainably maintained higher soil microbial richness and evenness without inhibiting microbial communities.

The mean relative abundance of the top 10 dominant phyla across all treatments is shown in Figure 4. The dominant bacterial phyla mainly included Proteobacteria, Actinobacteriota, and Chloroflexi. The total relative abundance of dominant bacterial phyla ranged from 88.31% to 89.88% in 2020 and increased to 94.94–96.29% by 2024. Compared with 2020, the relative proportions of Proteobacteria and Actinobacteriota in the PBAT treatment were markedly higher in 2024; the relative abundance of Chloroflexi showed a year-over-year increase. Long-term PBAT residue significantly altered the propor-

tional structure of the dominant bacterial phyla. The dominant fungal phyla were mainly Basidiomycota and Ascomycota. In 2020, the cumulative relative abundance of dominant fungal phyla across treatments was only 57.32–62.14%, but by 2024 it had reached 98.63–99.57%. The relative abundance of Ascomycota increased with experimental duration across all treatments, reaching approximately 75% in all treatments in 2024, with the PBAT treatment showing a lower relative abundance of Ascomycota than the PE treatment during the same period.

The top 10 genera by relative abundance across all treatments were considered the dominant genera (Figure 5). The relative proportions of dominant bacterial and fungal genera showed clear divergence

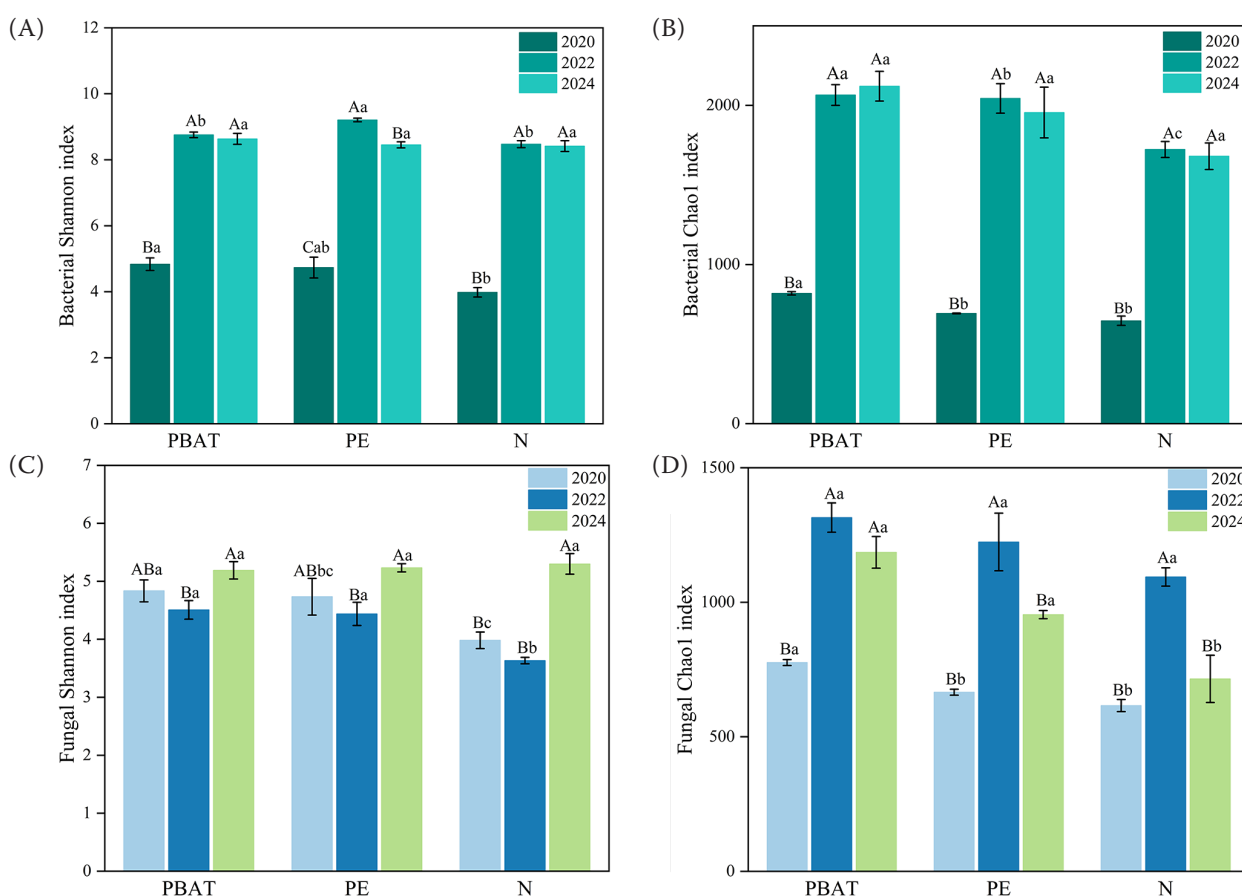


Figure 3. Shannon index and Chao1 index for bacteria (A, B) and fungi (C, D). Different lowercase letters indicate significant differences between treatments within the same year ($P < 0.05$), with means sorted in descending order ($a > b > c$); different uppercase letters indicate significant differences between years under the same treatment ($P < 0.05$), with means sorted in descending order ($A > B > C$). PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

with experimental duration across treatments. At the bacterial level, *Sphingomonas* maintained a relatively high proportion in the PBAT treatment across all years. In the fungal community, the relative proportion of *Fusarium* gradually decreased with experimental duration across all treatments, and it was consistently lowest in the PBAT treatment. Long-term PBAT residue increased the relative proportions of the saprophytic fungi *Humicola* and *Chaetomium*. In addition, the presence of *Penicillium*, *Saitozyma*, and several unclassified genera indicated further community complexity.

The results of microbial functional predictions.

Predictions of soil microbial functions under different treatments in 2020 and 2024 are shown in Figure 6. At level 2 functional classification of bacteria, the PBAT treatment showed relatively higher proportions of translation, amino acid metabolism, and transport and catabolic metabolism functions in

2020. By 2024, the proportion of exogenous substance degradation and metabolism functions in the PBAT treatment had increased, whereas the blank control and PE treatment enriched energy metabolism and carbohydrate-lipid metabolism pathways, respectively, indicating significant succession of bacterial functional profiles with increasing film residue duration. Fungal functional predictions revealed that the PBAT treatment had a higher proportion of saprotrophic fungal functional groups and a lower proportion of plant-pathogenic functional groups in 2020; this difference was further amplified by 2024, with the PE treatment and blank control showing relatively higher proportions of plant-pathogenic fungal functional groups.

Tobacco growth characteristics. Within the same year, the maximum leaf area under film treatments in 2020 was significantly higher than that under the no-film treatment. The PBAT treatment showed

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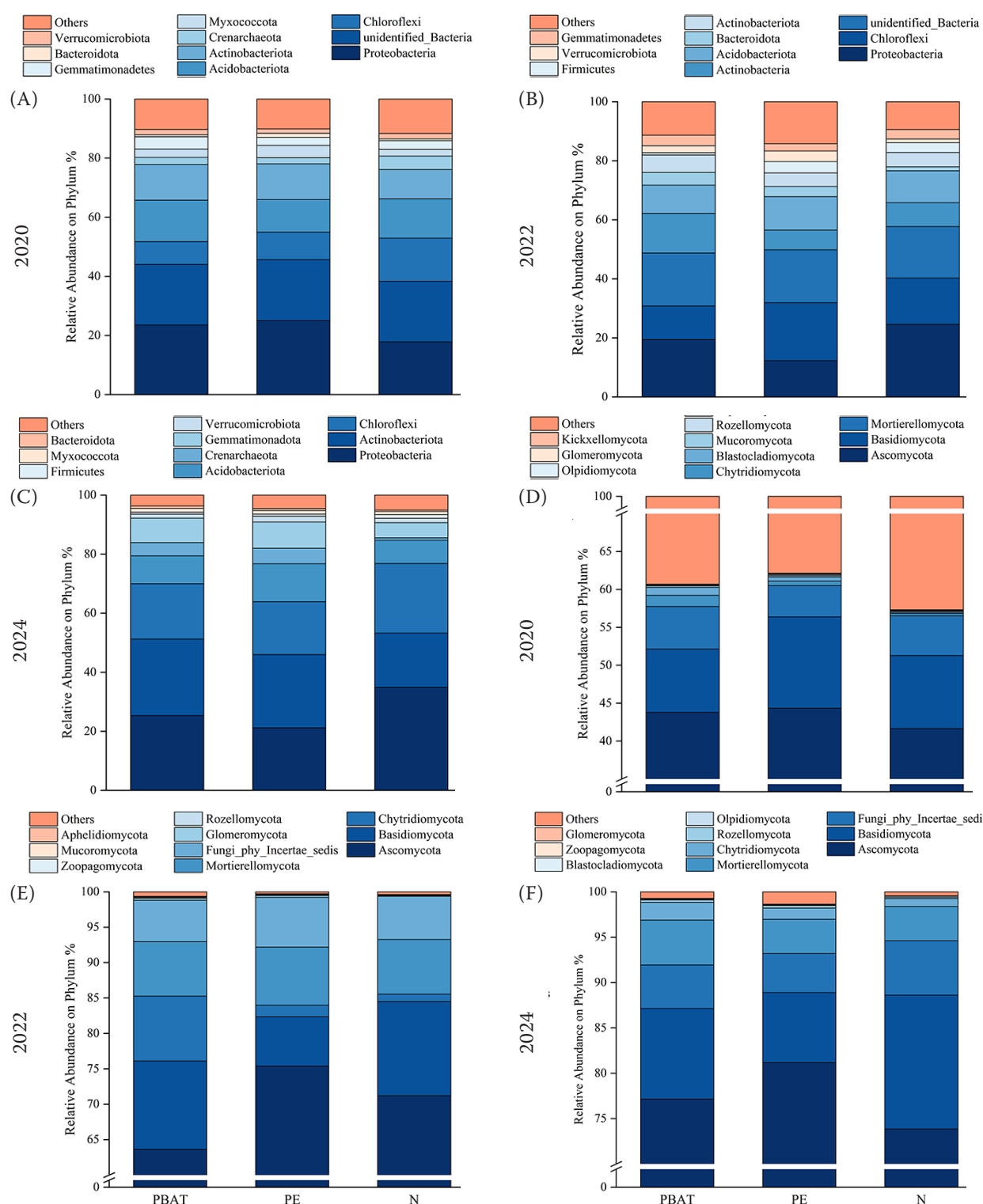


Figure 4. Relative abundances of the top 10 phyla for bacteria (A–C) and fungi (D–F). PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

significantly greater stem diameter in 2022 and plant height in 2024 than the other treatments, whereas leaf number showed no significant differences among treatments in any year. Within the same treatment,

plant height in the PE and N treatments and stem diameter in the N treatment decreased initially, then increased (lowest in 2022), whereas maximum leaf area in the N treatment increased significantly. In

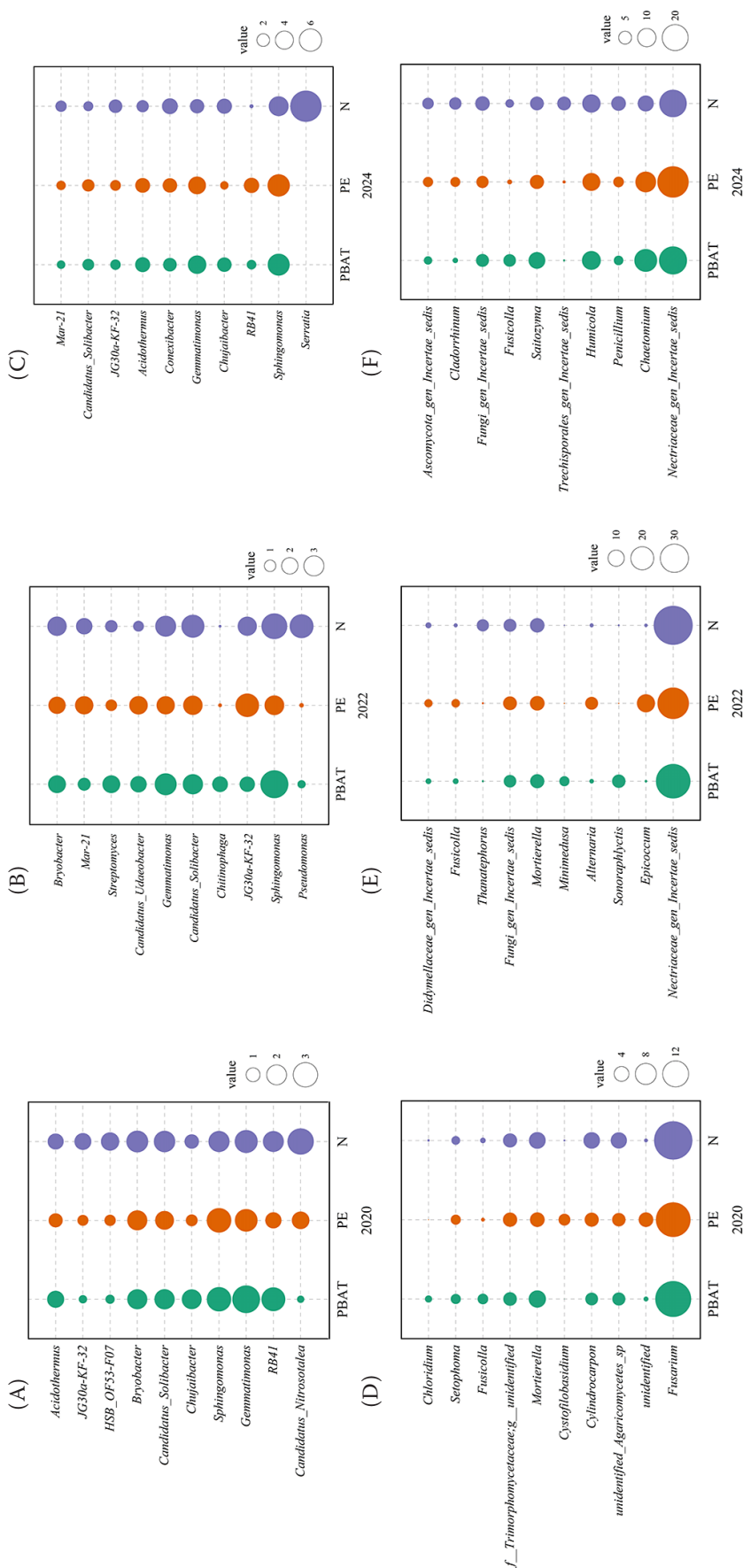


Figure 5. Bubble plots of the top 10 genera of bacteria (A–C) and fungi (D–F). PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

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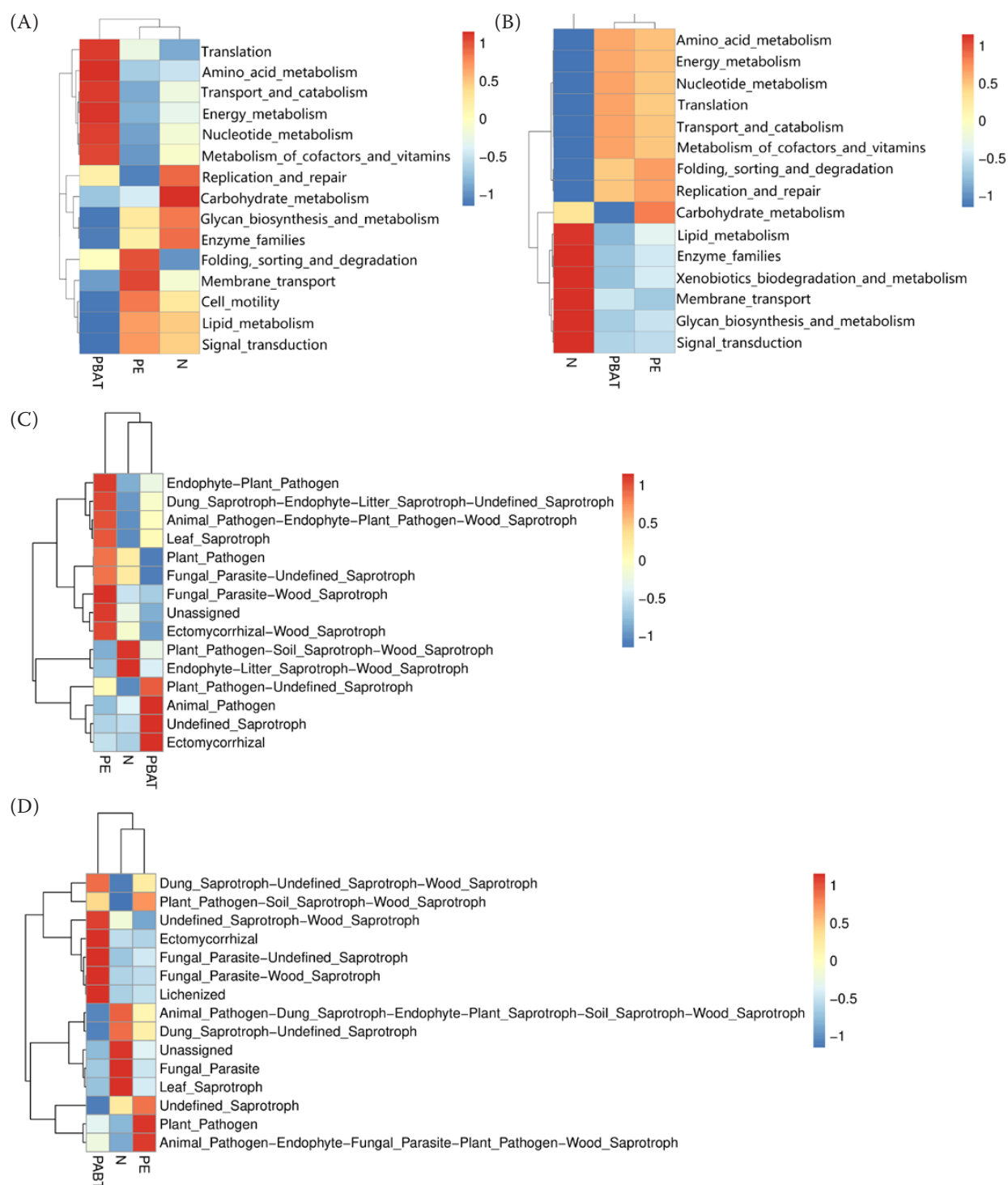


Figure 6. Level 2 functional classification of bacteria in (A) 2020 and (B) 2024 by Tax4Fun; guild functional classification of fungi in (C) 2020 and (D) 2024 by FUNGuild. PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

contrast, the PBAT treatment showed no significant interannual changes in any growth parameters. Overall, the PBAT treatment showed no significant interannual variation and contributed positively to

improvements in plant height, stem diameter, and maximum leaf area.

Analysis of the PLS-PM path model. Partial least squares path modelling (PLS-PM) was used to ana-

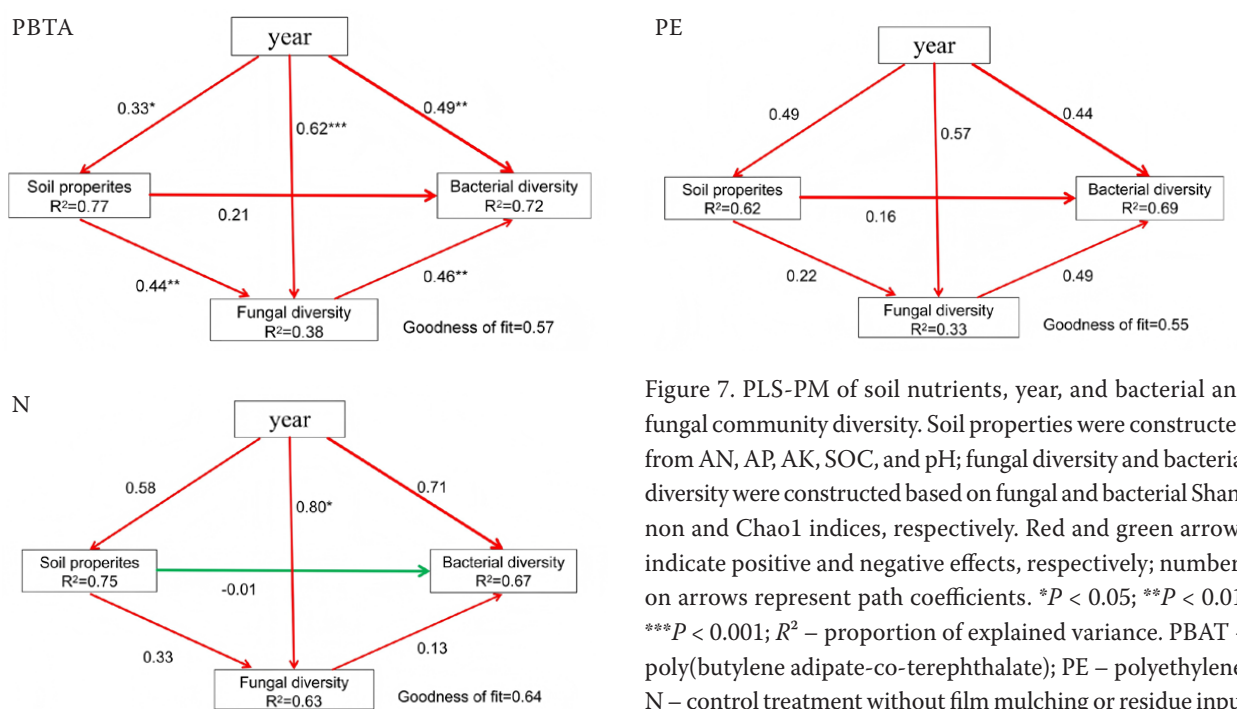


Figure 7. PLS-PM of soil nutrients, year, and bacterial and fungal community diversity. Soil properties were constructed from AN, AP, AK, SOC, and pH; fungal diversity and bacterial diversity were constructed based on fungal and bacterial Shannon and Chao1 indices, respectively. Red and green arrows indicate positive and negative effects, respectively; numbers on arrows represent path coefficients. $^*P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$; R^2 – proportion of explained variance. PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

lyse the relationships among cultivation year, soil physicochemical properties, and microbial alpha diversity, as shown in Figure 7. The models for all three treatments showed good fit. In the PBAT treatment, year had significant positive effects on soil physicochemical properties and bacterial and fungal diversity. In the PE treatment, the path coefficients between soil properties and microbial communities were relatively low, and year primarily regulated soil physicochemical and microbial indicators through direct effects. Under the blank control conditions, year had the strongest direct effect on microbial diversity, but soil physicochemical properties showed no significant effect on bacterial diversity.

DISCUSSION

The long-term impacts of mulch film residue on the soil ecological environment were evaluated in terms of film degradation behaviour, soil physicochemical properties, and bacterial and fungal community dynamics. Combined SEM and FTIR analyses revealed that, after years of field retention, the PE film surface exhibited only a few powdery particles, and its characteristic functional groups showed no significant changes, indicating that PE film is highly recalcitrant to decomposition and mineralisation in soil over the long term. In contrast, the surface structure of

the PBAT film underwent substantial degradation, with increasingly distinct layered features, and its FTIR spectra displayed decreased C=O and C-O-C peaks, a reduced C-H peak, and the emergence of an O-H band, collectively confirming the favourable in situ degradability of PBAT mulch film under field conditions (Liu et al. 2022).

The results of this study indicate that long-term PBAT residue did not induce soil acidification, nutrient depletion, or a decline in organic matter. Instead, it steadily increased the contents of available phosphorus, available potassium, and total potassium, thereby optimising the soil nutrient status (Zhang et al. 2022). Although previous studies have reported that the application of biodegradable films may adversely affect crop growth and development (Serrano-Ruiz et al. 2023), our findings demonstrated positive effects of long-term PBAT residue on tobacco growth (Table 2), possibly attributable to improved soil aeration. PE film tends to accumulate in soil, forming physical barriers that may impede root growth. In contrast, PBAT film may form pores during degradation, thereby enhancing soil aeration and promoting root respiration and nutrient uptake (Liu et al. 2025). In parallel, the sustained increase in soil-available potassium may provide adequate nutrition for tobacco, thereby contributing to improved agronomic traits (Yu et al. 2026).

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Table 2. Tobacco growth characteristics

Index	Year	PBAT	PE	N
Plant height (cm)	2020	99.33 ± 2.52 ^{Aa}	99.67 ± 5.13 ^{Aa}	95.83 ± 9.52 ^{Aa}
	2022	92.45 ± 9.56 ^{ABa}	86.82 ± 8.10 ^{Ba}	85.97 ± 6.95 ^{Ba}
	2024	104.8 ± 1.80 ^{Aa}	100 ± 1.66 ^{Aab}	96.53 ± 2.12 ^{Ab}
Stem diameter (mm)	2020	27.71 ± 0.40 ^{Aa}	27.93 ± 0.25 ^{Aa}	27.93 ± 0.31 ^{Aa}
	2022	27.72 ± 0.88 ^{Aa}	25.71 ± 1.01 ^{Ab}	24.94 ± 1.67 ^{Bb}
	2024	28.13 ± 0.82 ^{Aa}	26.77 ± 0.41 ^{Aa}	27.36 ± 0.81 ^{Aa}
Number of leaves	2020	17.67 ± 1.15 ^{Aa}	17.00 ± 1.00 ^{Aa}	16.67 ± 1.53 ^{Aa}
	2022	17.83 ± 1.60 ^{Aa}	17.00 ± 1.26 ^{Aa}	17.83 ± 3.31 ^{Aa}
	2024	18.47 ± 0.38 ^{Aa}	18.13 ± 0.29 ^{Aa}	18.67 ± 0.43 ^{Aa}
Maximum leaf area (cm ²)	2020	1 250.18 ± 96.15 ^{Aa}	1 165.10 ± 3.11 ^{Aa}	1 037.93 ± 3.11 ^{Bb}
	2022	1 129.35 ± 139.89 ^{Aa}	1 103.59 ± 240.21 ^{Aa}	1 045.16 ± 76.27 ^{ABa}
	2024	1 199.78 ± 88.71 ^{Aa}	1 038.88 ± 17.65 ^{ABa}	1 120.01 ± 81.69 ^{Aa}

PBAT – poly(butylene adipate-co-terephthalate); PE – polyethylene; N – control treatment without film mulching or residue input

Soil microbial communities are an important component of soil and a key indicator for evaluating soil quality and fertility, playing a vital role in maintaining soil ecology (Giovannetti et al. 2023). PBAT treatment significantly increased the species richness and evenness of the soil microbial community, indicating that it enhanced the stability of the microbial community structure. Among the bacterial communities, Proteobacteria and Actinobacteriota were the dominant phyla, both of which have been reported to play important roles in PBAT degradation. Upon entering the soil, PBAT film is progressively decomposed by microbial hydrolases, releasing small-molecule soluble organic carbon compounds such as terephthalic acid and adipic acid. These compounds provide exogenous available carbon sources for soil microorganisms and alter the dynamics of microbial resource competition (Li et al. 2023). At the genus level, the PBAT treatment enriched bacterial taxa such as *Sphingomonas*, a genus known for its strong capacity to degrade organic pollutants and to utilise the small-molecule organic compounds generated from PBAT breakdown (Tanunchai et al. 2025). In the fungal community, the PBAT treatment induced a sustained decrease in the relative abundance of the pathogenic genus *Fusarium* (Todorović et al. 2023), while increasing the proportions of saprophytic fungi *Humicola* and *Chaetomium* (Lu et al. 2023), which can efficiently utilise the organic matter released during PBAT degradation (Wang, Liu, et al. 2025). Functional predictions using Tax4Fun and FUNGuild further revealed that, in the later stage

of the PBAT treatment, the relative proportion of exogenous substance degradation metabolic pathways increased, corroborating the enrichment of degradation-associated microorganisms. In terms of fungal trophic modes, saprotrophic functions were enhanced while plant-pathogenic functions were suppressed. By contrast, the PE treatment and the blank control mainly enriched carbohydrate metabolism and energy metabolism pathways, with increased proportions of pathogen-related functions. The PLS-PM path model further revealed that the cultivation year was a core driver of soil physicochemical properties and microbial diversity. In the PBAT treatment, film degradation influenced the soil environment, preferentially driving fungal community succession, which in turn affected bacterial communities through interspecific interactions. In contrast, the PE treatment group showed weaker mediation pathways, with year predominantly driving changes in all indicators through direct effects; the physical stress imposed by residual film weakened the synergistic response between the soil environment and microbial communities. Soil physicochemical properties and bacterial diversity exhibited high explanatory power across all treatments, whereas the R^2 for fungal diversity under film residue treatments was markedly lower than that of the blank control, indicating that film residue introduced additional environmental disturbance that altered the driving mechanisms of fungal community succession (Li et al. 2023). However, we note an inherent limitation of our experimental design: PBAT was applied annu-

ally while PE was added only once at trial initiation. Differences between these two treatments may come from both material properties and residual dose/exposure duration. Therefore, the primary focus of this study is to reveal the long-term soil ecological effects induced by continuous annual input of PBAT film, rather than to directly compare the performance of the two film materials.

In conclusion, this five-year field study evaluated the effects of long-term PBAT film residue on soil microorganisms and tobacco growth. The results showed that PBAT film exhibited superior degradability, and its residual presence over time increased available phosphorus and potassium contents. Regarding tobacco growth, the PBAT treatment positively influenced plant height, stem diameter, and maximum leaf area, while overall growth parameters remained stable throughout the study period. Microbial community analysis revealed enhanced species richness and evenness, greater community stability, and enrichment of degradation-associated bacteria (e.g., *Sphingomonas*) and saprophytic fungi, alongside a consistent decline in *Fusarium* relative abundance. Functional predictions further indicated that, in the later stage, PBAT treatment upregulated exogenous substance degradation pathways and saprotrophic functions while suppressing plant-pathogenic functions. Collectively, these findings suggest that PBAT film holds promise for improving soil microecology and promoting tobacco growth, providing a scientific theoretical basis for the sustainable development of tobacco production.

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