Environmental Pollution 386 (2025) 127212

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol

Conventional and biodegradable agricultural microplastics: effects on soil properties and microbial functions across a European pedoclimatic gradient☆

Klára Šmídová [a], Helena Soinne [b], Shin Woong Kim [c,d,e], Jyri Tirronniemi [f], Raffaella Meffe [g], Paula E. Redondo-Hasselerharm [g], Melanie Braun [h], Matthias C. Rillig [c,d], Hannu Fritze [b], Bartosz Adamczyk [b], Johanna Nikama [b], Janne Kaseva [b], Vili Saartama [f], Wulf Amelung [h], Rachel Hurley [i], Jakub Hofman [a], Luca Nizzetto [i], Salla Selonen [f], Sannakajsa Velmala [b,*]

[a] RECETOX, Faculty of Science, Masaryk University, Kamenice 753/5, Brno, 625 00, Czech Republic
[b] Natural Resources Institute Finland (Luke), PO Box 2, FI-00791, Helsinki, Finland
[c] Institute of Biology, Freie Universität Berlin, 14195, Berlin, Germany
[d] Berlin-Brandenburg Institute of Advanced Biodiversity Research, 14195, Berlin, Germany
[e] Center for Ecotoxicology and Environmental Future Research, Korea Institute of Toxicology, 17 Jegok-gil, Jinju, 52834, Republic of Korea
[f] Finnish Environment Institute (Syke), Latokartanonkaari 11, 00790, Helsinki, Finland
[g] IMDEA Water Institute, Alcala de Henares, Madrid, 28805, Spain
[h] Institute of Crop Science and Resource Conservation (INRES) – Soil Science, Bonn University, Karlrobert-Kreiten-Strasse 13, 53115, Bonn, Germany
[i] Norwegian Institute for Water Research (NIVA), NO-0349, Oslo, Norway

A B S T R A C T

Agricultural plastics like mulching films may become a major source of microplastic (MP) soil contamination during their degradation and fragmentation. This study investigates the effects of agricultural MPs from conventional (linear low-density polyethylene, PE) and biodegradable (starch-blended polybutylene adipate co-terephthalate, PBAT-BD) mulching films on soil physicochemical properties, aggregation, microbial diversity and functions, litter decomposition, and greenhouse gases emissions (GHG). For this purpose, MPs were mixed into soils at realistic MP concentrations of 0.005 % and 0.05 % (w/w) in 2022 on experimental plots in three EU countries representing different pedoclimatic conditions (Finland, Germany and Spain), followed by monitoring of the above-mentioned variables in the subsequent growing seasons 2022 and 2023. We found several significant MP-induced effects for soil properties, aggregation, microbial diversity, litter decomposition, and GHG, but the effect endpoints were less pronounced or varied considerably. Contrarily, microbial activity, contributing to soil functions such as nitrogen cycling, was consistently reduced by both conventional and biodegradable MPs. The reductions were more pronounced after the second season and for the higher MP treatment. As the higher MP concentration (i.e., 0.05 % w/w) is environmentally relevant in Europe, our findings emphasize the potential effects of environmentally relevant MP concentrations on soil health. Furthermore, the effects increased from north to south, probably modulated by varying pedoclimatic conditions, inducing reflection of a need for regionally tailored risk assessment to protect soil from plastic pollution.

1. Introduction

Microplastics (MPs) are pervasive and persistent pollutants of all ecosystems (Schell et al., 2020; Zhu et al., 2019). They can release harmful additives and infiltrate the food chain, potentially impacting also human health (Do et al., 2022; Siddiqui et al., 2023). Soils are considered the largest environmental reservoirs of MPs globally (Bläsing and Amelung, 2018; Hurley and Nizzetto, 2018), with their presence particularly well-documented in agricultural lands (Chia et al., 2023; Corradini et al., 2019; Scheurer and Bigalke, 2018). Although research on the distribution and prevalence of MP in soils is still hindered by the lack of harmonized methodologies and comparable data, reported levels for agricultural soils vary widely (from a few hundreds to tens of thousands of particles per kg of soil mass) (Wrigley et al., 2024) and are expected to rise in the future (Meizoso-Regueira et al., 2024). A rough conversion suggests that mass ratios of plastic in soil can vary between <0.0001 % and 0.1 % which are the ratios already appeared in the literature (Corradini et al., 2019; Dierkes et al., 2019). In extreme cases,

☆ This paper has been recommended for acceptance by Klaus Kümmerer.

* Corresponding author.
E-mail address: sannakajsa.velmala@luke.fi (S. Velmala).

https://doi.org/10.1016/j.envpol.2025.127212
Received 12 May 2025; Received in revised form 11 September 2025; Accepted 3 October 2025
Available online 7 October 2025

such as the mishandling and burial of mulching films, substantially higher ratios are possible (Koutnik et al., 2021).

MPs enter soils via atmospheric deposition, sludge application, or use and mismanagement of agricultural plastics (APs) (e.g., mulching films, ground covers, solarization films, nets, irrigation pipes) (FAO, 2021). While biosolid fertilizers are considered a major source for agricultural soils (Nizzetto et al., 2016; Piehl et al., 2018; Weithmann et al., 2018), the role of APs has recently garnered increased attention (Baker and Thygesen, 2021; Briassoulis, 2023; FAO, 2021; Hofmann et al., 2023). The use of APs is expanding both in quantity (1.5–2 Mt y^{-1} of APs in the EU (Hann et al., 2021) and 11.7 Mt y^{-1} globally (Plastics Europe, 2022) and in the diversity of applications (Briassoulis, 2023; Dhevagi et al., 2024). Following ageing during use or mismanagement, APs can degrade and release MPs. Several studies, mostly conducted in laboratory conditions suggest this contamination can affect soil properties and health (Yang et al., 2024; Zhang et al., 2024a,b). However, assessment of soil responses to MP pollution originating from APs in field conditions is still limited (Jia et al., 2024; Lin et al., 2020). Unlike laboratory studies, field experiments yield more environmentally relevant insights and reflect the effects on soil physicochemical properties, soil biota and plants. Indeed, recent work has emphasised the need for long-term field experiments (Jiang et al., 2024; Johansen et al., 2024), but so far only a limited number of studies cover different pedoclimatic regions and soil types, with management practices relevant to crop production. Addressing this gap is essential for understanding the risks and long-term impacts of microplastics in agroecosystems (Hasan and Tarannum, 2025), and it forms the basis of our study design.

Plastic mulching films are widely used to help minimize water loss, increase soil temperature to support germination, and suppress weed growth, which collectively contributes to enhanced agricultural productivity (Kasirajan and Ngouajio, 2012). However, thin mulching films often experience substantial degradation due to environmental stressors like ultraviolet radiation, hydrolysis, microbial degradation, mechanical damage, and thermal gradients (Alimi et al., 2022; Karkanorachaki et al., 2022; Pfohl et al., 2022). They tend to become brittle and tear; their partial burial in the soil at the edges complicates retrieval even more, leaving fragmented plastic scattered across the field (Briassoulis, 2023). The repeated application of plastic mulch films has significantly contributed to the accumulation of MP debris in soils, especially in areas using very thin (e.g., 8 μm) films (Huang et al., 2020; Li et al., 2022; Liu et al., 2018). For example, after 32 years of continuous use, plastic mulch films accounted for 33–56 % of total MPs in soil (0–100 cm), averaging 8885 particles kg^{-1} in the topsoil (0–10 cm) (Li et al., 2022). However, as approximately 75 % of available studies were conducted in Asia, the knowledge about the MPs originating from the mulching films in European soils is limited (Büks and Kaupenjohann, 2020; Wrigley et al., 2024) with a few exceptions from, e.g., Spain, Italy and Germany (Beriot et al., 2021; Kedzierski et al., 2023; Piehl et al., 2018). For example, Piehl et al. (2018) reported that 43.7 % of MPs were potentially derived from APs.

MP contamination has been shown to hold the potential of affecting a broader range of soil characteristics and functions, including: microbial activity (De Souza Machado et al., 2018; Zhao et al., 2021; Kim et al), litter decomposition (Djukic et al., 2018), and soil properties, such as soil pH and nutrients, water holding capacity, bulk density and aggregation (De Souza Machado et al., 2018; Qiu et al., 2022; Wang et al., 2022). These effects depend on MPs' physical and chemical properties, soil contamination, and climate (Feng et al., 2022; Fu et al., 2022; Ren et al., 2020). Recent studies show that MPs significantly affect soil microbial communities and fungal biomass, fostering distinct microbial assemblages including plastic-degrading bacteria, which alter microbial diversity and richness (Aralappanavar et al., 2024; Sun et al., 2022; Zhang et al., 2023). The microbial mineralization of MP led to an increase in microbial activity and CO_2 production, and nitrogen mineralization was also shown to be altered (Reay et al., 2023; Rillig et al., 2021). Although the effects of MPs on the soil microbial community and other mentioned effects are highly dependent on the surrounding environment and anthropogenic conditions, most studies to date have been conducted in controlled environments that may differ from complex and fluctuating field conditions. Field experiments are not without their own particular set of challenges. For instance, if they are not performed with a robust statistical design, the variations in climatic conditions or spatial variation in the soil properties may mask the impacts of the factors being studied. On the other hand, a robust study based on adequate replicates, proper design and consistent management practices across tested treatments minimises the influence of uncontrolled sources of variability and strengthens the reliability of the obtained findings.

This study examines the impact of MPs on soil physicochemical properties, microbial communities, and processes under field conditions in Finland, Germany, and Spain, representing diverse pedoclimatic conditions. Here we focus on APs that originate from mulch films. Consequently, MPs from conventional and biodegradable mulching films that were selected for their widespread agricultural use and limited research were applied at environmentally relevant concentrations (0.005 % and 0.05 % w/w dry soil). Over two years, soil aggregation, bacterial and fungal community composition, microbial carbon (C) and nitrogen (N) mineralization, organic matter decomposition, and greenhouse gas emissions (in Finland) were analyzed. The hypotheses were as follows: (i) agricultural MPs affect soil properties and structure, soil microbial community composition and soil microbial processes at environmentally relevant concentrations; (ii) biodegradable and conventional agricultural MPs have different effects on these endpoints; (iii) the effects differ under contrasting pedoclimatic conditions; and (iv) the effects vary with time due to degradation. This study is among the first to investigate a wide range of realistic factors will be investigated simultaneously in a long-term experiment.

2. Materials and methods

2.1. Microplastic test materials

Two commercial mulching films, linear low-density polyethylene (LLDPE, hereafter PE) and biodegradable starch-blended polybutylene adipate co-terephthalate (PBAT-BD, hereafter Bio), were cryomilled from repelleted films to produce MPs. This cost-effective method enabled large-scale production for field experiments. Recycled film pellets retain the virgin films' chemical composition but exhibit reduced stability, resembling aged rather than new films (Hurley et al., 2024). Film thicknesses were 15.4 ± 0.6 μm (PE) and 15.1 ± 0.8 μm (Bio). Both MPs were repeatedly utilized in studies within the EU Horizon 2020 project "Plastic in Agricultural Production: Impacts, Lifecycle and Long-term Sustainability" (PAPILLONS, 2024), with internal designations M-rPE-black-A0 (PE-MP) and M-rBIO-black-A0 (PB-MP). Detailed MP characteristics are available in Hurley et al. (2024).

2.2. Field plot experiment setup

The field plot experiments were conducted with identical setups in three EU countries within the EU Horizon 2020 project PAPILLONS (PAPILLONS, 2024). The coordinates are as follows: Finland 60°51′02.4″ N 23°28′03.9″ E, Germany 50°37′00.6″ N 6°59′50.7″ E, and Spain 40°31′41.7″ N 3°17′44.4″ W. Composite soil samples for characterization of the experimental sites were taken before the establishment of the experiments, and the basic soil properties are listed in Table 1.

The experimental setup included a control (non-treated) and four MP treatments: PE low concentration at 0.005 % w/w (PEL) and high concentration 0.05 % w/w (PEH), and Bio at 0.005 % w/w (BioL) and 0.05 % w/w (BioH). These concentrations, selected based on MP occurrence in agricultural soils (Qiu et al., 2022), represent mid-to-high exposure levels. MP mass calculations for the top 10 cm soil layer assumed a bulk density (BD) of 1.5 g cm^{-3}, but site-specific variations resulted in final concentrations of 0.007 % and 0.065 % (Finland), 0.006 % and 0.055 %

Table 1
Particle size distribution, pH (in 0.01 M $CaCl_2$ and water suspension), electrical conductivity (EC), total carbon (TC), total nitrogen (TN), and organic carbon (OC) measured in the top 0–20 cm soil layer at each field experiment site.

Location	Texture[a]			pH_{CaCl2}	pH_{H2O}	EC mS cm^{-1}	TC[b]%	TN[b]%	OC[c]%
	<2 µm	2–20 µm	>20 µm						
Finland	26	9	65	5.4	6.0	0.09	2.32	0.20	2.32
Germany	19	28	53	6.9	7.3	0.17	1.21	0.14	1.21
Spain	39	21	40	7.4	7.9	0.25	1.41	0.16	1.31

[a] Determined by the pipette method.
[b] Measured with a dry-combustion method (Vario Max CN-analyzer).
[c] OC = TC-IC, IC measured with a dry-combustion method after removing OC by the loss on ignition (LOI) (Koorneef et al., 2023).

(Germany), and 0.006 % and 0.058 % (Spain). For clarity, treatment codes 0.005 % and 0.05 % are used in figures and tables.

The field experiment in each country included five replicates (n = 5) per treatment and control, totaling 25 randomized blind-to-operator plots in a Latin square design. The study area measured 51 m × 51 m, with 3.5 m × 4.5 m plots spaced 5.5 m apart (Supplementary Fig. S1a). Treatments were applied across entire plots, while sampling occurred in designated 2.5 m × 2.5 m areas, divided for sampling in 2022 and 2023 (Supplementary Fig. S1b). Litter decomposition tea bags and GHG collars were positioned outside sampling zones.

Initiated in spring 2022, MPs were distributed on the soil surface using a stainless-steel colander and incorporated into the uppermost 10 cm with a rotavator, ensuring control plots received identical disturbance. Malt barley was sown, with fertilization and plant protection treatments following site-specific regulations. Planting, fertilisation and plant protection measures were applied uniformly across all plots, including controls without MPs.

2.3. Effects endpoints, sampling, and sample storage

Soil samples for chemical properties, aggregation, and microbial activity were collected using a metallic soil auger in autumn 2022 and 2023, within seven days post-harvest. Four sub-samples were combined to form 2-kg composite samples for each depth (Supplementary Table S1). Soil aggregates were extracted as undisturbed blocks, while the remaining material was homogenized before subsampling. Composite samples designated for chemical analyses were air-dried and stored at room temperature, whereas samples for aggregate stability and microbial activity were maintained in crush-resistant containers (sterilized 50 mL-conical test tube and plastic boxes with holes in the lid >1 dm^3, respectively) at 4 ± 2 °C to ensure sample integrity.

Bulk density (BD) measurements were conducted using a 5 cm high cylinder of known volume (Supplementary Table S1), with BD calculated as dry soil mass divided by cylinder volume (g cm^{-3}). BD samples were not collected in Germany in 2022.

For microbial community and fungal biomass analyses, soil samples for DNA and ergosterol extraction were obtained from the topsoil layer using a steel corer, which was disinfected with 70 % ethanol before each use to prevent contamination. To avoid cross contamination, the corer was inserted into the soil outside the experimental area between plots before cleaning. Samples were sieved through heat-sterilized sieves in the field, maintained on ice during transport, and stored frozen at −20 °C in Eppendorf tubes for subsequent analysis.

Litter decomposition was assessed using the standardized "Tea bag" method (Didion et al., 2016) with green tea (*Camellia sinensis*) and rooibos tea (*Aspalanthus linearis*), selected for their differing cellulose and lignin content (Djukic et al., 2018). Lipton-brand tea, sponsored by CVC Capital Partners plc, was dried at 70 °C for 48 h before weighing. Due to unavailability of the original tetrahedron-shaped synthetic tea bags (Keuskamp et al., 2013) in the EU, nylon mesh bags (250 µm mesh size) were used. Each plot received three tea bags per type per incubation period (two months annually, with an additional three-month batch in 2023), buried at 8 cm depth and covered with cored soil. Bags were placed outside sampling areas to prevent disturbance. After incubation, tea bags were carefully retrieved using metal spoons, ensuring ID-tag integrity, cleaned of soil and plant debris, and dried at 70 °C for 48 h.

At the Finnish site, the greenhouse gases (GHGs) CH_4, CO_2, and N_2O were measured using LI-7810 and LI-7820 analysers (LICOR Biosciences) in May and September 2022, and twice in June, July, and August 2023. Measurement collars were installed in May, with plant removal conducted one day prior. In total, GHGs were assessed eight times during the barley growing season.

2.4. Analytical methods

Detailed descriptions for all analytical methods are available in the supplementary material under method descriptions.

2.4.1. Soil physicochemical properties

Soil pH and electric conductivity (EC) were measured from air-dried soils after water extraction (1:2.5 v:v). Nutrients (WETN, NO_3-N, NH_4-N, PO_4-P) and WEOC were analyzed via 1-h water extraction (1:10, w:v), filtration (0.7 µm glass fiber filter), and detection with a Skalar San++ autoanalyzer and TOC-V CPN Analyzer (Shimadzu, Kyoto, Japan). WEON was calculated as WETN minus mineral nitrogen (NO_3-N, NH_4-N). TC and TN were measured via dry combustion (Vario Max CN-analyser), and OC was determined by subtracting IC measured after ignition from TC. Results are expressed per soil dry weight (DW, oven-dried at 105 °C).

Soil aggregation size fractions were analyzed using a modified protocol (Rillig et al., 2019). Air-dried soil (1 week, room temperature) was sieved (4 mm), then passed through stacked sieves (4000, 2000, 1000, 250 µm) to determine aggregate size proportions.

Water-stable aggregates were assessed via a modified protocol (Kemper & Rosenau, 2018). Air-dried (1 week) soil (4–5 g) was rewetted (5 min, deionized water, 0.25 mm sieve) and wet-sieved (Eijkelkamp, Netherlands; stroke = 1.3 cm, 34 times min^{-1}, 3 min). Samples were separated into water-unstable and water-stable (>0.25 mm) fractions, dried (60 °C, 24 h) and corrected for debris and sand. Water-stable aggregate percentage was calculated as (water-stable fraction – coarse matter)/(4.0 g – coarse matter).

2.4.2. Microbial communities

Isolation of microbial genomic DNA from approximately 250 mg_{ww} fresh soil samples was done with the DNeasy PowerSoil Pro kit (Qiagen) using the QIAcube Connect (Qiagen). The bacterial V4-V5 16S rRNA region was amplified using 515F-926R primers (Parada et al., 2016; Quince et al., 2011), and the fungal ITS region with fITS7-ITS4 primers (Ihrmark et al., 2012; White et al., 1990). Bacterial and fungal libraries were prepared as described by Kim et al and sequenced on an Illumina MiSeq 2000 platform (BeGenDiv, Berlin, Germany) using 2x300-bp paired-end sequencing. ASV richness was assessed, with denoised, chimera-free, non-singleton bacterial and fungal ASVs obtained via DADA2 (Callahan et al., 2016; R Core Team, 2024), processed separately for each country and year. Taxonomy was assigned using the Silva (ver. 132) (Quast et al., 2012), and UNITE (Abarenkov et al., 2023) databases.

Alpha diversity indices were calculated using the VEGAN package from rarefied data.

Fungal biomass was quantified via its biomarker, ergosterol, using HPLC with a reverse-phase column (Adamczyk et al., 2024). Ergosterol was extracted from 0.25 g freeze-dried soil with cyclohexane and 10 % KOH in methanol, ultrasonicated for 15 min, and incubated at 70 °C for 1 h. After phase separation with water and cyclohexane, extracts were evaporated under nitrogen gas at 40 °C, re-dissolved in methanol, and filtered (0.2-μm PTFE). HPLC (Arc Waters, USA) with a C18 100A reverse-phase column (Phenomenex, USA) measured ergosterol via 10 μl injection and isocratic separation (100 % methanol, 1 ml min^{-1}), detected at 282 nm.

2.4.3. Microbial activity and decomposition

Water holding capacity (WHC) according to (ISO 11268-2, 2023) and dry mass according to (ISO 10390, 2021) were measured for each treatment, the rest of the soil samples were moistened to 40 % WHC_{max} and stored in the fridge at 4 ± 2 °C until analysis (max 8 weeks according to (ISO 18512, 2007)). Before measurements, soil samples were preincubated in darkness at 21 ± 2 °C for 14 days to stabilize the soil environment (Hund-Rinke and Simon, 2008).

Carbon mineralization was measured via basal respiration (BR) and substrate-induced respiration (SIR) (ISO 14240-1, 1997; ISO 16072, 2002), using 10 gdw samples sealed in infusion bottles, aerated, incubated at 21 ± 2 °C, and analyzed via Aquilent GC 6850, with CO_2 levels expressed as μg CO_2-C gdw^{-1} h^{-1}. SIR followed a similar procedure, with glucose addition, and CO_2 measured after 0, 3, and 6 h.

Nitrogen mineralization was assessed via potential ammonium oxidation (PAO) and potential ammonification (PAMO). PAO followed ISO 15685 (2012), using 10 gdw samples with 1.5 mmoL L^{-1} $(NH_4)_2SO_4$ as substrate, analyzed in FIAlab-2500 (1M KCl mobile phase) and expressed as μg NO_2-N gdw^{-1} h^{-1}. PAMO involved adding 1 mg L-arg mL^{-1} L-arginine to 2 gdw samples, incubated at 30 °C, analyzed via FIAlab-2500 (2M KCl mobile phase), and expressed as μg NH_4-N gdw^{-1} h^{-1}.

Litter decomposition was assessed separately for each tea type per plot by calculating the mean percentage of remaining mass in dried tea bags.

2.5. Statistical analyses

For a detailed description of study sites, see section 2.2. A total of 36 endpoints (Supplementary Table S2) were analyzed, including soil properties, microbial diversity, decomposition, and functional activity using a generalized linear mixed model (GLMM). Fixed effects included treatment, country, year (2022 and 2023), depth (0–10 cm and 10–20 cm), and their interactions, while random effects accounted for spatial variation.

Depth correlations within plots were modeled using homogeneous (CS) or heterogeneous (CSH) compound symmetry structures (R-side random effect), and year correlations via G-side random effects. Unequal variances were allowed for countries as needed, based on likelihood ratio tests. For skewed variables, gamma or log-normal distributions were applied. Residual normality was assessed graphically and it was adequate for all models. The residual pseudo-likelihood RSPL estimation was used for skewed variables, and REML for others, with degrees of freedom calculated using the Kenward-Roger method. Treatment effects were compared to controls within country-depth-year combinations using Dunnett's method ($\alpha = 0.05$). The analyses were performed using the GLIMMIX procedure of SAS Enterprise Guide 8.3 (SAS Institute Inc., Cary, NC, USA). Beta diversity was analyzed using Bray-Curtis distances, and the treatment effect was calculated using non-parametric multivariate analysis of variance test (ADONIS2) and permutational multivariate analysis of variance (PERMANOVA) in the VEGAN package of R using 9999 permutations. Non-metric multidimensional scaling (NMDS, function metaMDS) ordinations were used to visualize the microbial community. Pairwise t-tests with Bonferroni correction ($p < 0.05$) assessed significant Bray-Curtis distance differences.

2.6. Data storage and availability

Data will be available upon publication. Used data will be accessible via Zenodo (10.5281/zenodo.14825718; 10.5281/zenodo.11068075), while raw amplicon sequence data are deposited in the NCBI SRA under PRJNA1218479.

3. Results

3.1. Soil physicochemical properties

Only a few statistically significant differences were observed among measured soil properties between the control and MP treatments (Supplementary Tables S3, S4 and S5). BD varied across countries and soil layers but showed no significant differences between the treatments (Supplementary Table S3). Soil pH was lower in the BioH treatment than in the control concerning the 0–10 cm layer in Finland in the year 2023 ($p = 0.046$) and in the 10–20 cm layer in Germany in the year 2022 ($p < 0.001$). In Germany in 2022, low concentrations of PE and Bio also coincided with lower pH than control in the 10–20 cm soil layer ($p < 0.001$) (Supplementary Table S3, Fig. S2). No significant differences were detected in Spain, which inherently has the highest pH.

The MP treatment did also not affect soil OC, TN, or WEOC contents (Supplementary Tables S3, S4 and S5). In the 0–10 cm soil layer, OC was 2.5, 1.2, and 1.4 % in Finland, Germany, and Spain, respectively, and in the 10–20 cm soil layer, 2.1, 1.2, and 1.2 %. In line with the OC contents, WEOC concentrations were largest in Finland (252 and 244 mg kg^{-1} in 0–10 and 10–20 cm soil layers, respectively), followed by Spain (189 and 143 mg kg^{-1} in 0–10 and 10–20 cm soil layers, respectively) and Germany (130 and 127 mg kg^{-1} in 0–10 and 10–20 cm soil layers, respectively).

Concerning NO_3-N levels, statistically significant differences to the control were observed only in Spain in 2023. Here, lower NO_3-N contents were detected in both PEL and PEH treatments at 0–10 cm depth ($p = 0.016$ and 0.013, respectively) and in the PEH treatment at 10–20 cm depth ($p = 0.045$) (Fig. 1). These reduced NO_3-N levels contributed to lower WETDN in the treatments with the PEL and PEH at 0–10 cm ($p = 0.010$ and 0.034, respectively) (Supplementary Tables S4 and S5). Higher WEON contents relative to control plots were measured only in Germany in 0–10 and 10–20 cm soil layers in PEL ($p = 0.032$ and $p = 0.041$, respectively) and in 10–20 cm soil layer in BioL ($p = 0.001$) (Supplementary Table S3). In general, the lowest WEON contents were measured in 2022 in Germany. Lowest water extractable PO_4-P contents were measured in Spain, but MPs did not have any impact on the content of water extractable PO_4-P (Supplementary Tables S4 and S5).

The soil aggregate size fractions varied across years, and this variation was more or less similar to the variation within treatments. The amount of soil aggregates in the size range of 2–4 mm was larger in the control in 2023 than in 2022 ($p < 0.001$), which went along with a decline in the amount of the smaller soil aggregate sizes (<0.25 and 0.25–1 mm) ($p < 0.05$) (Supplementary Table S6), and overall lower portions in the fractions (%) of water-stable aggregates (Fig. 2). MP treatments, in turn, had inconsistent effects on both soil aggregate size fractions and water stable aggregates (Fig. 2, Table S6). In Finland, the amount of water-stable aggregates was larger in the PEH and BioL treatments than in the control, accompanied by a slight (from 21 % to 24 %) gain in the fractions of the small macroaggregates (0.25–1 mm; Supplementary Table S6); yet, these effects were not apparent in the other regions (Fig. 2). In Spain, we observed a loss of the 2–4 mm aggregate size fractions in the PEH trials, which went along with a gain of the 0.25–1 mm fraction (Supplementary Table S6). Soil microaggregation remained unaffected in all trials and countries (Supplementary Table S6).

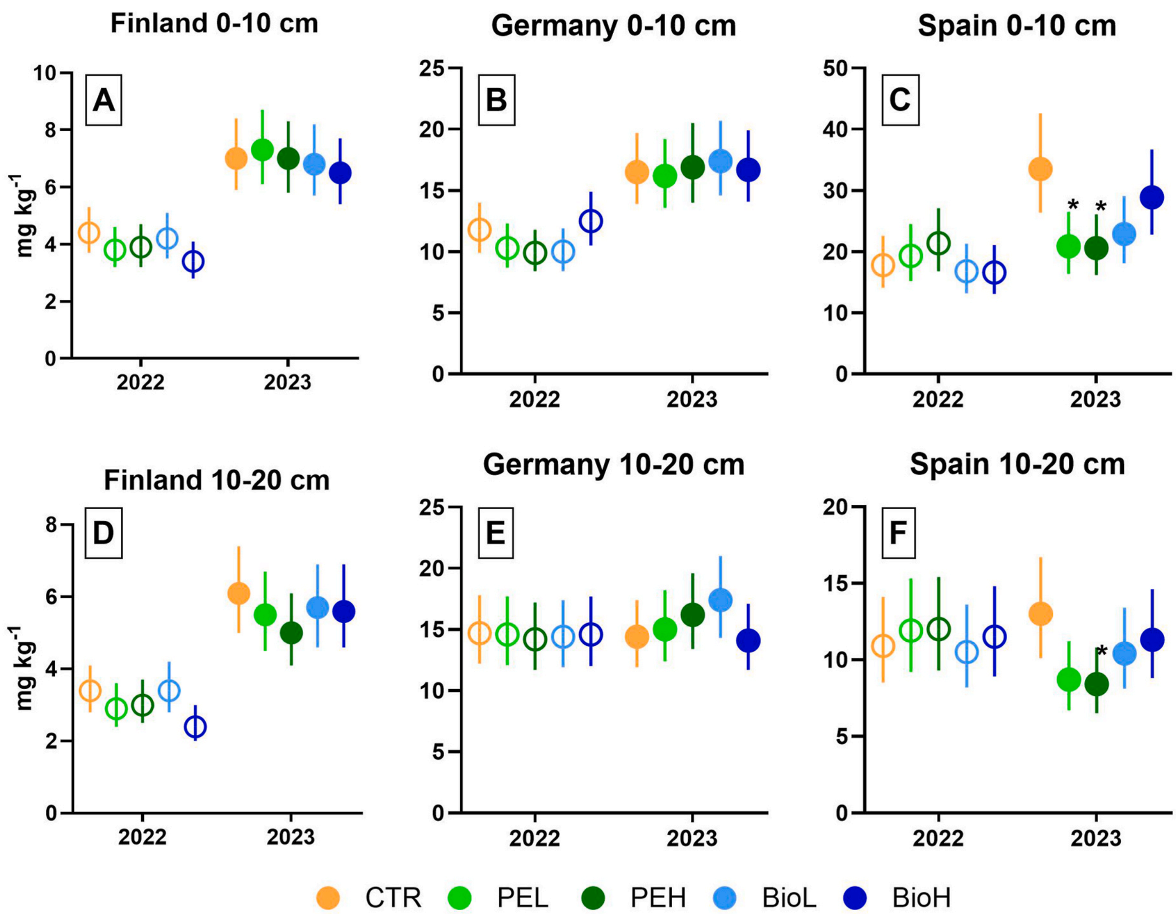

Fig. 1. Nitrate (NO_3-N) in mg kg^{-1}. The values represent estimated means (± the upper and lower limits) measured in 0–10 cm (A, B, C) and 10–20 cm soil layers (D, E, F) after the first (2022, open circles) and second (2023, closed circles) growing seasons in Finland, Germany, and Spain. Treatments from left to right include control (CTR), polyethylene MP at low (PEL) and high (PEH) concentrations, and PBAT-BD MP at low (BioL) and high (BioH) concentrations. Stars indicate statistically significant differences from the control withing the year ($p < 0.05$).

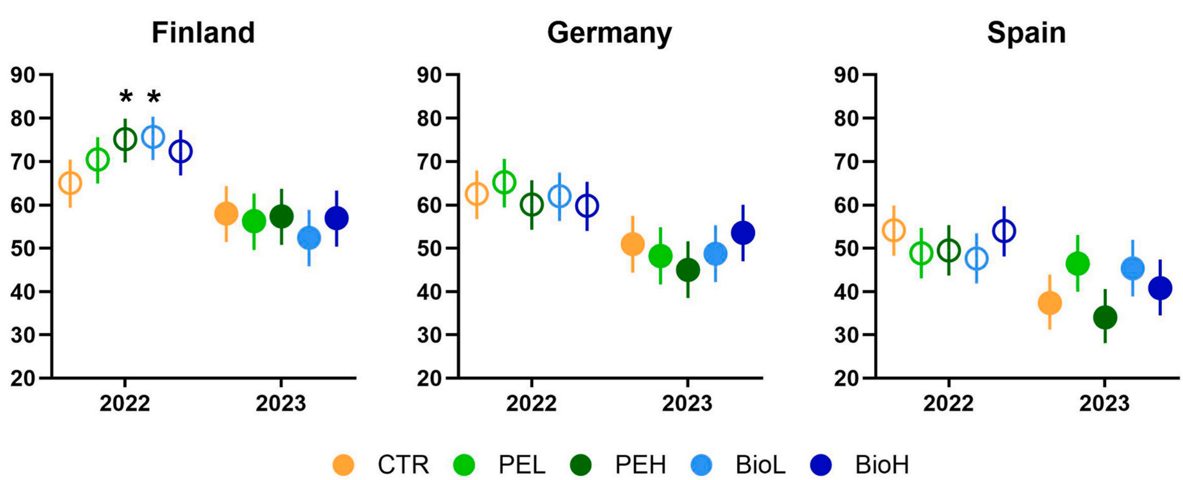

Fig. 2. Soil water stable aggregates (WSA) in %. The values represent estimated means (± the upper and lower limits) measured after the first (2022, open circles) and second (2023, closed circles) growing seasons in Finland, Germany, and Spain, respectively. Treatments from left to right include control (CTR), applications of polyethylene MP at low (PEL) and high (PEH) concentrations, and PBAT-BD MP at low (BioL) and high (BioH) concentrations, respectively. Asterisks indicate statistically significant differences from the control withing the year ($p < 0.05$).

3.2. Effects on microbial communities

Alpha diversity indices (Chao1, InvSimpson, Shannon) of the bacterial community showed no significant differences between MP treatments and the control (Supplementary Table S7). Across all field experiments, the dominant bacterial phyla were Acidobacteriota (9–21 %), Actinobacteriota (17–31 %), and Proteobacteria (21–28 %) (Supplementary Fig. S3–S4), with no MP-induced abundance changes (p

> 0.05). NMDS ordination and PERMANOVA tests indicated no significant shifts in bacterial communities at the ASV level (Supplementary Fig. S5). However, Bray-Curtis distance estimates revealed a significantly lower bacterial community similarity in Germany's PEL treatment (2023, $p = 0.041$) and a significantly higher divergence in Spain's BioH treatment (2023, $p < 0.001$) compared to controls (Supplementary Fig. S6e–S6f), suggesting a convergent effect of PEL in Germany and a divergent effect of BioH in Spain.

Fungal alpha diversity indices showed minimal changes between MP treatments and controls across all sites (Supplementary Table S7). However, in Germany, fungal ASV diversity was significantly lower in PEH (InvSimpson $p = 0.032$) and BioH (InvSimpson $p = 0.002$, Shannon $p = 0.027$) in 2022, while ASV richness was significantly higher in PEL ($p = 0.046$) in 2023. Dominant fungal phyla included Ascomycota (62–95 %), Basidiomycota (3–16 %), Mortierellomycota (1–6 %), and Chytridiomycota (<3 %) (Supplementary Fig. S7–S8). In Finland, Basidiomycota abundance increased in BioL treatment compared to the control ($p = 0.016$). NMDS ordination and PERMANOVA tests showed fungal communities remained similar across MP treatments in Finland, Germany, and Spain ($p > 0.05$, Supplementary Fig. S9). Bray-Curtis distance estimates also showed no significant MP-induced community shifts ($p > 0.05$, Supplementary Fig. S10). Variability in microbial results was high due to the relatively low number of replicates.

Fungal biomass measured via its biomarker, ergosterol, was the highest in 2022 in Finland and Spain sites (Supplementary Table S8). In Finland and Germany, there were no statistically significant differences between MP treatments in 2022 and in 2023. However, in Spain in 2022, in BioH, PEL, and PEH, fungal biomass significantly (all $p < 0.001$) decreased compared to the control.

3.3. Microbial activity and decomposition

In Finland, representing the northern Europe region, significant decreases ($p < 0.05$) were found for BR between the control and high concentrations of MPs in 2023 and for SIR between the control and BioH in 2022 ($p < 0.001$, Fig. 3, Supplementary Fig. S11). In Germany, representing the central Europe region, the trend was visible only for BR in 2023, where all treatments showed significantly lower values of microbial activity than control ($p < 0.05$, Fig. 3). Finally, in Spain, although the microbial activity related to C mineralization was mostly unaffected by the treatments in 2022 (except for SIR in BioL and BioH, $p = 0.027$ and 0.024, respectively), changes were observed in 2023

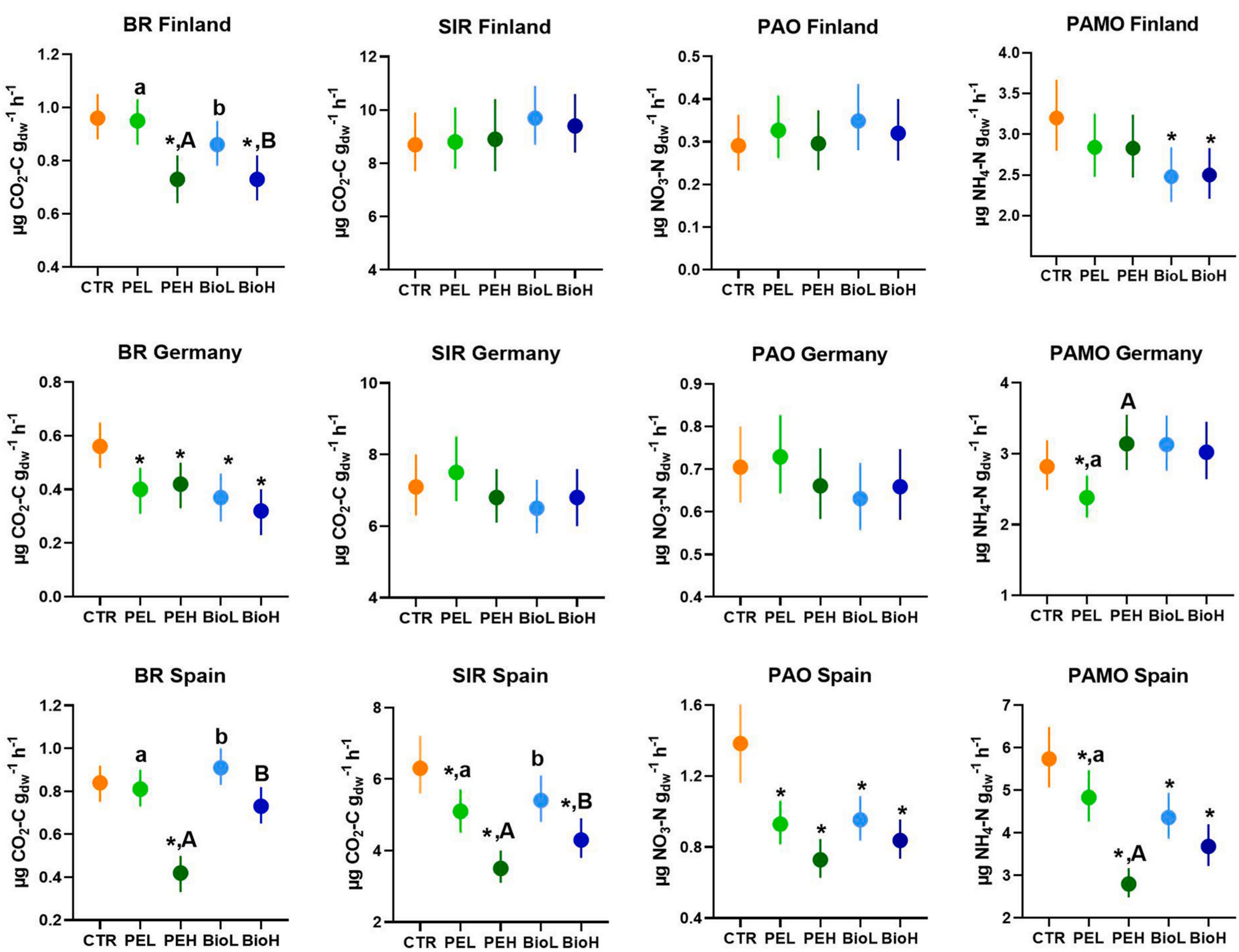

Fig. 3. Basal respiration (BR), substrate-induced respiration (SIR), potential ammonium oxidation (PAO), and potential ammonification (PAMO). The values represent estimated means (± the upper and lower limits) measured after the second (2023) growing seasons in Finland, Germany, and Spain. Treatments from left to right include control (CTR), polyethylene MP at low (PEL) and high (PEH) concentrations, and PBAT-BD MP at low (BioL) and high (BioH) concentrations. Stars indicate statistically significant differences from the control withing the year ($p < 0.05$). Letters show significant differences between low and high concentration treatments ($p < 0.05$).

(Fig. 3, Supplementary Fig. S11). Significant decreases ($p < 0.05$) in microbial activity compared to the control were shown for PEH and BioH for both BR and SIR (except for BR in BioH) and also for SIR in PEL.

Significant differences between treatments with low and high concentrations of specific MPs were observed only in 2023 except for Finland where a significant decrease of SIR in BioH compared to BioL was recorded also in 2022 ($p = 0.002$, Supplementary Fig. S11). The differences in 2023 followed a similar trend, and only significant decreases of the activity with the increased MP concentration were found: BR decreased for both studied compounds in Finland and Spain, and SIR decreased only in Spain ($p < 0.05$, Fig. 3). In Germany, low and high concentrations revealed similar results ($p > 0.05$). In none of the treatments, we observed an enhancement of microbial performance after MP application.

In Germany and Finland, N mineralization (assessed as PAO and PAMO) measured from MP treatments were primarily similar to control soils, except in Finland in 2023, where PAMO was significantly lower in BioL and BioH ($p = 0.005$ and $p = 0.003$, respectively), in Germany in 2022, where PAO in BioL and PAMO in BioH were significantly lower ($p = 0.044$ and $p = 0.008$, respectively), and in Germany in 2023, where PAMO was also lower in PEL ($p = 0.031$) than in CTR (Fig. 3, Supplementary Fig. S11). On the other hand, a consistent decrease in PAO relative to CTR was observed in the MP treatments in Spain, both in 2022 and 2023 ($p < 0.05$, Fig. 3, Supplementary Fig. S11). Moreover, PAMO was reduced by MP treatment in the same country, though only in 2023. The concentration of the MPs significantly impacted N mineralization for PE in Spain in 2023, where significantly lower PAMO was found in PEH compared to PEL ($p = 0.028$, Fig. 3). Finally, the only increase in N mineralization with the increasing MP concentration in this study was recorded in Germany in 2023 for PE ($p = 0.001$, Fig. 3).

Rooibos tea degraded more slowly than green tea in all countries (Supplementary Fig. S12), with significant yearly effects on two-month incubation for both tea types ($p < 0.001$). Green tea decomposition varied across countries ($p < 0.001$), with the fastest degradation in Germany, followed by Finland and Spain. Rooibos degraded fastest in Finland, followed by Spain and Germany ($p < 0.05$), though after three months, only Spain showed significantly greater degradation than Germany ($p = 0.025$). In Finland, BioH treatments showed slower degradation than controls for rooibos tea in 2022 ($p = 0.025$) and green tea in 2023 ($p = 0.002$), with no differences in three-month incubations. No significant MP treatment effects were observed in Germany or Spain.

All GHG fluxes varied during the season (Supplementary Fig. S13) but the MP treatment did not have effects on CH_4, CO_2 fluxes whereas the N_2O flux was higher in PEH, BioL and BioH in late May and in BioL and BioH in mid June than in the control.

4. Discussion

This study examined the impact of agricultural microplastics (MPs) from conventional and biodegradable mulching films on soil properties, microbial processes, and greenhouse gas production across diverse pedoclimatic conditions in Finland, Germany, and Spain.

4.1. Soil physicochemical properties

While lower bulk densities (BDs) have been reported in MP-contaminated soils (de Souza Machado et al., 2018), particularly with fiber-shaped MPs, no BD differences were observed across our three field experiments with PE or PBAT-BD MPs. This aligns with findings by Massaccesi et al. (2025) and Yu et al. (2023), indicating that significant BD changes occur only at MP concentrations above 0.5 %, with soil heterogeneity further masking minor variations.

The effects of environmentally relevant MP concentrations on soil pH, as studied here, were inconsistent across sites and years. Nevertheless, the few significant differences observed indicated lower pH in MP treatments compared to the controls. In general, MPs composed of different polymer types can have varying or even opposing effects on soil pH (Ma et al., 2023a,b,c; Šmídová et al., 2024; Zhao et al., 2021), and in addition to chemical composition of MPs, their impact may depend on soil properties that influence MP aging (Gong et al., 2024; Ma et al., 2023a,b,c). However, the decrease in pH was mostly observed for the treatments with the biodegradable polymer, belonging to the group of polyesters. For polyesters, a release of protons during degradation is known to cause a decrease in pH (Qi et al., 2020; Sander et al., 2023; Liu et al., 2023).

Varying effects of MPs on the solubility of OM in soils have been reported, including both increases and decreases (Jing Ma et al., 2023). These differing impacts have been attributed to factors such as MP type and concentration (Meng et al., 2022) and the size of MP particles (Ma et al., 2023a,b,c). In our study, at MP treatments of approximately 0.005 % and 0.05 %, we observed no significant effects of PE or PBAT-BD MPs on WEOC or water-extractable PO_4-P in soil samples collected post-harvest. Plant residues left in the field after harvest are a major source of organic matter and substantially contribute to an increase in extractable OM in the autumn (Schiedung et al., 2017). Consequently, the potential effects of the low MP concentrations tested in this study were likely masked by the substantial organic matter input from plant residues.

Short-term incubation studies indicate that MPs impact N cycling in soils and sediments (Huang et al., 2020; Liu et al., 2017; Yang et al., 2024). Here, soil TN showed no significant treatment differences and is likely too robust measure to reflect changes in nitrogen cycling over a two-year field experiment under typical agricultural practices. MPs can inhibit organic N mineralization (Yang et al., 2025) and reduce nitrification rates (Zhang et al., 2024a,b), limiting mineral N availability. In 2023, Spain showed the highest NO_3-N levels in control plots, while PE treatments had significantly lower levels, suggesting that active N mineralization may create a regime sensitive to subtle MP-induced changes.

Although significant changes were observed in Finland and Spain, MP effects on soil aggregate size fractions and water-stable aggregates (WSA) were inconsistent. Previous studies report both positive and negative impacts, varying with soil type, MP shape, and polymer type (Chen et al., 2025; De Souza Machado et al., 2018), often at high MP concentrations (>0.2 % dry soil mass) (De Souza Machado et al., 2018; Liang et al., 2021). However, in field experiments, environmental and management factors (e.g., field operations, drying-wetting cycles) in arable soils may overshadow small MP-induced aggregation changes. WSA increased in Finland for high PE and low PBAT-BD after the first growing season, though this change did not relate with other soil property variations and was absent by the second season, indicating a short-lived effect. No increase in microaggregates (<0.25 mm), typically associated with macroaggregate deterioration, was observed (Lobe et al., 2011; Oades & Waters, 1991). In Spain, however, macroaggregate sizes decreased, possibly linked to the decline in fungal biomass (see 4.2).

4.2. Microbial communities

MPs can form a distinct microhabitat by enriching the microbial taxa degrading plastics, and this is well known as an important mechanism shifting the structures and compositions of the microbial community by MPs (Ren et al., 2020). Most previous studies regarding these effects of MPs on microbial communities have been based on laboratory incubations, typically at very high concentration where PBAT-BD MPs can induce significant changes (positive or negative) to the bacterial community at the concentration of ≥0.2 % (Feng et al., 2022; Hu et al., 2022; Li et al., 2022; Shi et al., 2022; Wang et al., 2022). On the contrary, a field study using realistic concentrations (100 kg ha^1 equaling 0.01 % of the soil weight) showed no effects on the soil microbial community (Greenfield et al., 2022). Greenfield et al. (2022) used an MP concentration comparable to ours, but we could show in two of our field trials

MP-induced responses. We observed a decrease in fungal diversity in high exposure PE and PBAT-BD MP treatments in the first season in Germany. In addition, the fungal biomass changed significantly in Spain in 2022, showing decreased results for both levels of PE and high PBAT-BD MPs. This suggests either a potential toxic effect of MPs for fungi, or a selective promotion of bacteria strains. The latter, however, contrasts with the general decline in microbial activity parameters, suggesting that the toxic effects likely prevail. Fungal growth may influence soil aggregation, as fungi are known to retard the disintegration of large macroaggregates (Morris et al., 2019). In Spain, the PEH treatment was associated with a reduction in the proportion of large macroaggregates, corresponding to the observed decline in fungal biomass (Supplementary Table S4).

4.3. Carbon and nitrogen mineralization and decomposition

Despite the absence of statistically significant disparities in the alpha diversity indices (Chao1, InvSimpson, and Shannon) of the bacterial community between the MP treatments and the control group, microbial activity exhibited systematic decreases in MP treatments. These findings suggest that MP has a significant impact on the composition of microbial communities in soil, underscoring the importance of microbial activity assessment as a more precise metric for measuring these changes.

Our study found that soil C mineralization, measured as CO_2 production, decreased with increasing PE and PBAT-BD MP concentrations in Spain at 0.05 %, an environmentally relevant level. A similar decline in SIR was observed in mesocosm experiments at 0.05 % PBAT-BD (Kim et al), while at 0.8 % w/w, BR and SIR increased significantly (Kim et al). Rauscher et al. (2023) reported no effect of PBAT-BD MPs on CO_2 emissions at 0.1 %, but a significant increase at 1 %. These findings suggest that low MP levels may suppress microbial activity due to altered soil properties or energy sources, while higher concentrations may stimulate activity as PBAT-BD MPs serve as a C source. MP-induced microbial effects may also result from additives or small molecular compounds (Hahladakis et al., 2018; Li et al., 2021). PBAT-BD MP consumption is associated with bacterial species (Rauscher et al., 2023), though we found no MP-related shifts in bacterial community composition. Using 13C-labeled polymers, Zumstein et al. (2018) demonstrated that PBAT-BD is converted to CO_2 during microbial respiration and contributes to biomass formation. The threshold for these effects remains uncertain; adverse impacts were noted at 0.05 % (this study) and 0.4 % (Zhao et al., 2021), while positive effects emerged at 0.8–1 % (Kim et al; Rauscher et al., 2023). These findings apply specifically to PBAT-BD MPs, as Rauscher et al. (2023) detected no effect at low or high PE-MP concentrations, likely due to PBAT-BD's higher degradability. This study's novel finding is that MP contamination may impair microbial activity even at environmentally relevant levels. Furthermore, prior studies were limited to laboratory or mesocosm settings, underscoring the need for insights from field experiments.

Whereas in Spain, the effect of MP concentration was observed for both monitored microbial activities related to the C mineralization in 2023, different behaviour was found in Germany and Finland. In these countries, significant differences were found for BR in 2023, but SIR was mostly comparable between the controls and MP treatments. This may indicate that the effect of MPs on C mineralization is site/region-specific and may be connected with, e.g., the degree of MP degradation, the amount of released additives, and degrading products. In 2022, the PBAT-BD MP-induced effect was observed only for SIR in Finland and Spain. More differences were reported in 2023, which supports the potential for cumulative effects on the soil environment (Brandes et al., 2021).

In our study, N mineralization revealed similar trends as C-mineralization. Compared to the controls, a significant activity reduction was observed in soil contaminated by MPs in 2022 and 2023 in Spain, and a few inconsistent differences were also noted in Germany and Finland. Therefore, we assume that studied MPs originating from the mulching films at environmentally relevant concentrations have similar modes of action on N mineralization as described previously for C mineralization. PAO has been previously shown to be sensitive to heavy metals and organic contaminants independent of their biodegradability (Hund-Rinke and Simon, 2008). Our study confirms this phenomenon and shows that microbial activity is a sensitive tool also for testing the impacts of agricultural MPs on soil processes. On one hand, Reay et al. (2023) observed the retention of NH_4^+ and NO_3^- pools in the soil contaminated by PLA-PBAT, and they hypothesized that both nitrification and denitrification processes might have been suppressed by contamination of these MPs. On the other hand, in the same study in soils with added PE-MPs, the N assimilation was found to be comparable to the control, indicating no negative effects on microbial activities (Reay et al., 2023). Although the potential ammonification and potential ammonium oxidation were measured in our study, which should not be directly dependent on the availability of actual N sources in the soils, systematic decreases in microbial activities were observed for both PBAT-BD and PE MPs in Spain.

There were very mild MP effects on decomposition. We observed lower degradation in Finland in 2022 in the high PBAT-BD MP treatment compared to the control which is in line with the decrease of C mineralization. However, C mineralization decreased in MP treatments also in 2023, and this phenomenon was not observed for litter degradation. As expected, green tea degraded faster than rooibos, due to its higher fraction of water-soluble compounds that leach out early in the degradation process (Didion et al., 2016). Green tea degraded the fastest in Germany, followed by Finland and Spain. This trend aligns with previous observations, where green tea degrades more slowly in warmer and drier Mediterranean climates compared to boreal or temperate climates (Djukic et al., 2018). Interestingly, rooibos degraded slower in Germany than in Finland or Spain. Climatic conditions and management practices used in agriculture affect degradation (Gmach et al., 2024), which may explain why the degradation differed among countries.

5. Implications

A significant proportion of MP impact studies have been conducted in controlled laboratory environments, often using higher MP concentrations and excluding plants (Jiang et al., 2024; Johansen et al., 2024). In contrast, this study examined PE and PBAT-BD MP effects at environmentally relevant concentrations in three European field experiments with barley cultivation. In natural settings, soil heterogeneity and plant growth can obscure MP-induced changes or buffer harmful effects, explaining why many measured soil properties showed no alterations. These effects varied across countries and years and did not encompass the full range of natural variability, yet findings indicate that even low MP concentrations can influence N cycling and microbial functions.

At low MP concentrations, N cycling-related changes were observed in Germany (WEON) and Spain (NO_3-N). However, MP impacts on soil properties were not consistently mirrored in microbial activity under controlled lab conditions. For instance, OC and WEOC, key substrates for soil biota, showed comparable levels across MP treatments and controls, yet microbial activity measured in the lab displayed MP-related effects. Of several N cycling-related properties, only a few MP-induced changes aligned with laboratory-assessed N mineralization. In Germany, higher WEON was detected in the low PE soil treatment after the first growing season, but microbial activity related to N mineralization remained unchanged. By 2023, following the second growing season, Spanish plots with low and high PE soil treatment exhibited lower NO_3-N and WETN, with significant declines in PAO and PAMO. These findings suggest microbial activity is a sensitive indicator of MP effects, demonstrating that MP can alter N cycling processes. While generalizations on GHG emissions remain challenging due to limited data, increasing evidence supports MP-induced N_2O emissions (Su et al., 2023; Zhang et al., 2024a,b), corroborated by our field data.

Our results align with recent literature (Chen et al., 2025; Thompson et al., 2024; Zeb et al., 2024), emphasizing MP effects as soil type- and environment-dependent. Spain showed the most significant MP-induced effects, consistent with studies indicating stronger MP impacts on the nitrogen cycle at elevated temperatures (Shi et al., 2022; Xiang et al., 2023). Although lab tests for PAO and PAMO were conducted under uniform temperatures, MP-treated soils displayed long-term changes, likely influenced by prolonged high-temperature exposure. Shi et al. (2022) suggested that lower mineral N levels in MP-treated soils result from reduced mineralization rates, particularly under higher temperatures, with MP additive release and transformation potentially accelerating at elevated temperatures (Lv et al., 2024).

Aggregate stability in Spain was lower than in Finland and Germany, despite higher clay content usually correlating with increased WSA (Schweizer et al., 2019). Spain's high clay content and low organic matter resulted in an SOC/clay ratio of 1:30, indicating structural vulnerability (Dexter et al., 2008; Johannes et al., 2017) and limited organic N availability for mineralization (Ros et al., 2011; Soinne et al., 2021). High temperatures, weak structural stability, and reduced mineralizable OM availability stress soil microbiota. The pronounced MP effects observed in Spain align with Rillig et al. (2024), who suggest multiple stressors heighten the risk of impaired soil ecosystem functioning. While MP impacts may vary by ecosystem health, our results indicate that under multiple stressors, even low MP concentrations can contribute to soil degradation, highlighting the need for region-specific risk assessments, particularly in Southern Europe.

6. Conclusion

This study investigated the effects of polyethylene (PE) and PBAT-BD microplastics (MPs) at environmentally relevant concentrations across three European field sites, revealing that even low MP levels can alter nitrogen (N) cycling and microbial activity, particularly under site-specific conditions. While many soil properties showed no significant MP-induced changes due to environmental buffering, microbial responses, especially related to N cycling, proved more sensitive, most notably in Spain and Germany. These effects were influenced by factors like temperature, soil structure, and organic matter availability, with Spain showing the strongest MP impacts due to its high clay content, low organic matter, and elevated temperatures. The findings underscore the environment- and soil-dependent nature of MP effects and support growing evidence that MPs contribute to N_2O emissions and potential soil degradation, particularly under multiple stressors, emphasizing the need for tailored regional risk assessments.

CRediT authorship contribution statement

Klára Šmídová: Writing – original draft, Visualization, Validation, Methodology, Formal analysis, Data curation. **Helena Soinne:** Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Data curation. **Shin Woong Kim:** Writing – original draft, Visualization, Methodology, Formal analysis, Data curation. **Jyri Tirronniemi:** Writing – original draft, Methodology, Investigation, Formal analysis. **Raffaella Meffe:** Writing – review & editing, Investigation, Funding acquisition. **Paula E. Redondo-Hasselerharm:** Writing – original draft, Project administration, Investigation, Funding acquisition. **Melanie Braun:** Writing – original draft, Investigation, Funding acquisition, Conceptualization. **Matthias C. Rillig:** Writing – review & editing, Project administration, Conceptualization. **Hannu Fritze:** Writing – original draft, Funding acquisition, Formal analysis, Conceptualization. **Bartosz Adamczyk:** Writing – original draft, Formal analysis, Data curation. **Johanna Nikama:** Writing – review & editing, Validation, Methodology, Investigation. **Janne Kaseva:** Writing – original draft, Validation, Data curation. **Vili Saartama:** Writing – review & editing, Investigation. **Wulf Amelung:** Writing – review & editing, Funding acquisition, Conceptualization. **Rachel Hurley:** Writing – review & editing, Conceptualization. **Jakub Hofman:** Writing – original draft, Supervision, Funding acquisition, Conceptualization. **Luca Nizzetto:** Writing – review & editing, Resources, Project administration, Funding acquisition, Conceptualization. **Salla Selonen:** Writing – review & editing, Supervision, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization. **Sannakajsa Velmala:** Writing – original draft, Visualization, Validation, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

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

Acknowledgments and funding instruments

This study received funding under grant agreements No 101000210 (project PAPILLONS) and was supported under grant agreement No 857560 (CETOCOEN) from the European Union's Horizon 2020 research and innovation program. This publication reflects only the author's view, and the European Commission is not responsible for any use that may be made of the information it contains. The authors thank the RECETOX Research Infrastructure (No LM2023069), financed by the Ministry of Education, Youth, and Sports, for the supportive background, and Korea Institute of Toxicology, Republic of Korea (2710008768). The publication reflects the authors' views; the European Commission is not responsible for its use.

We thank Sanna Maula, Susanna Lehtonen, Jani Anttila, Riikka Keskinen, Juha-Matti Pitkänen (Luke); Marek Šudoma, Zuzana Hochmanová (RECETOX); Venla Forsell, Anna Kangas, Minna Sepponen, Ilse Heiskanen (Syke); Kirsten Unger, Rene Heyse (BONN); and Lesly Ayala Cabana, Lucia Barbero, Melina Crettaz, Daniel Franco, Ana García Arcos, Sara Martínez, Virtudes Martínez Hernández, Pedro Vicente Mauri Ablanque, David Mostaza Colado, Andreu Rico, Cynthia Rieckhof Rivas, Teresa Schell, Gloría Teijón Ávila (IMDEA Water Institute) for their support in field sampling and laboratory work.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2025.127212.

Data availability

shared trough Zenodo.

References

Abarenkov, K., Zirk, A., Piirmann, T., Pöhönen, R., Ivanov, F., Nilsson, R.H., Kõljalg, U., 2023. UNITE general FASTA release for fungi. https://doi.org/10.15156/BIO/2938067.

Adamczyk, S., Lehtonen, A., Mäkipää, R., Adamczyk, B., 2024. A step forward in fungal biomass estimation – a new protocol for more precise measurements of soil ergosterol with liquid chromatography-mass spectrometry and comparison of extraction methods. New Phytol. 241, 2333–2336. https://doi.org/10.1111/nph.19450.

Alimi, O.S., Claveau-Mallet, D., Kurusu, R.S., Lapointe, M., Bayen, S., Tufenkji, N., 2022. Weathering pathways and protocols for environmentally relevant microplastics and nanoplastics: what are we missing? J. Hazard. Mater. 423, 126955. https://doi.org/10.1016/j.jhazmat.2021.126955.

Aralappanavar, V.K., Mukhopadhyay, R., Yu, Y., Liu, J., Bhatnagar, A., Praveena, S.M., Li, Y., Paller, M., Adyel, T.M., Rinklebe, J., Bolan, N.S., Sarkar, B., 2024. Effects of microplastics on soil microorganisms and microbial functions in nutrients and carbon cycling – a review. Sci. Total Environ. 924, 171435. https://doi.org/10.1016/j.scitotenv.2024.171435.

Baker, E., Thygesen, K., 2021. Working Paper: Plastics in Agriculture: Sources and Impacts, No. 762.

Beriot, N., Peek, J., Zornoza, R., Geissen, V., Huerta Lwanga, E., 2021. Low density-microplastics detected in sheep faeces and soil: a case study from the intensive vegetable farming in Southeast Spain. Sci. Total Environ. 755, 142653. https://doi.org/10.1016/j.scitotenv.2020.142653.

Bläsing, M., Amelung, W., 2018. Plastics in soil: analytical methods and possible sources. Sci. Total Environ. 612, 422–435. https://doi.org/10.1016/j.scitotenv.2017.08.086.

Brandes, E., Henseler, M., Kreins, P., 2021. Identifying hot-spots for microplastic contamination in agricultural soils—a spatial modelling approach for Germany. Environ. Res. Lett. 16, 104041. https://doi.org/10.1088/1748-9326/ac21e6.

Briassoulis, D., 2023. Agricultural plastics as a potential threat to food security, health, and environment through soil pollution by microplastics: problem definition. Sci. Total Environ. 892, 164533. https://doi.org/10.1016/j.scitotenv.2023.164533.

Büks, F., Kaupenjohann, M., 2020. Global concentrations of microplastics in soils – a review. SOIL 6, 649–662. https://doi.org/10.5194/soil-6-649-2020.

Callahan, B.J., McMurdie, P.J., Rosen, M.J., Han, A.W., Johnson, A.J.A., Holmes, S.P., 2016. DADA2: high-resolution sample inference from illumina amplicon data. Nat. Methods 13, 581–583. https://doi.org/10.1038/nmeth.3869.

Chen, H., Ingraffia, R., Schloter, M., Brüggemann, N., Rillig, M.C., 2025. Effects of multiple microplastic types on growth of winter wheat and soil properties vary in different agricultural soils. PLANTS PEOPLE PLANET 7, 194–203. https://doi.org/10.1002/ppp3.10573.

Chia, R.W., Lee, J.-Y., Cha, J., Çelen-Erdem, İ., 2023. A comparative study of soil microplastic pollution sources: a review. Environ. Pollut. Bioavailab. 35, 2280526. https://doi.org/10.1080/26395940.2023.2280526.

Corradini, F., Meza, P., Eguiluz, R., Casado, F., Huerta-Lwanga, E., Geissen, V., 2019. Evidence of microplastic accumulation in agricultural soils from sewage sludge disposal. Sci. Total Environ. 671, 411–420. https://doi.org/10.1016/j.scitotenv.2019.03.368.

De Souza Machado, A.A., Lau, C.W., Till, J., Kloas, W., Lehmann, A., Becker, R., Rillig, M.C., 2018. Impacts of microplastics on the soil biophysical environment. Environ. Sci. Technol. 52, 9656–9665. https://doi.org/10.1021/acs.est.8b02212.

Dexter, A.R., Richard, G., Arrouays, D., Czyż, E.A., Jolivet, C., Duval, O., 2008. Complexed organic matter controls soil physical properties. Geoderma 144, 620–627. https://doi.org/10.1016/j.geoderma.2008.01.022.

Dhevagi, P., Poornima, R., Keerthi Sahasa, R.G., Ramya, A., Karthika, S., Sivasubramanian, K., 2024. The crux of microplastics in soil - a review. Int. J. Environ. Anal. Chem. 104, 6546–6578. https://doi.org/10.1080/03067319.2022.2148528.

Didion, M., Repo, A., Liski, J., Forsius, M., Bierbaumer, M., Djukic, I., 2016. Towards harmonizing leaf litter decomposition studies using standard tea bags—A field study and model application. Forests 7, 167. https://doi.org/10.3390/f7080167.

Dierkes, G., Lauschke, T., Becher, S., Schumacher, H., Földi, C., Ternes, T., 2019. Quantification of microplastics in environmental samples via pressurized liquid extraction and pyrolysis-gas chromatography. Anal. Bioanal. Chem. 411, 6959–6968. https://doi.org/10.1007/s00216-019-02066-9.

Djukic, I., Kepfer-Rojas, S., Schmidt, I.K., Larsen, K.S., Beier, C., Berg, B., Verheyen, K., Caliman, A., Paquette, A., Gutiérrez-Girón, A., Humber, A., Valdecantos, A., Petraglia, A., Alexander, H., Augustaitis, A., Saillard, A., Fernández, A.C.R., Sousa, A.I., Lillebø, A.I., Da Rocha Gripp, A., Francez, A.-J., Fischer, A., Bohner, A., Malyshev, A., Andrić, A., Smith, A., Stanisci, A., Seres, A., Schmidt, A., Avila, A., Probst, A., Ouin, A., Khuroo, A.A., Verstraeten, A., Palabral-Aguilera, A.N., Stefanski, A., Gaxiola, A., Muys, B., Bosman, B., Ahrends, B., Parker, B., Sattler, B., Yang, B., Juráni, B., Erschbamer, B., Ortiz, C.E.R., Christiansen, C.T., Carol Adair, E., Meredieu, C., Mony, C., Nock, C.A., Chen, C.-L., Wang, C.-P., Baum, C., Rixen, C., Delire, C., Piscart, C., Andrews, C., Rebmann, C., Branquinho, C., Polyanskaya, D., Delgado, D.F., Wundram, D., Radeideh, D., Ordóñez-Regil, E., Crawford, E., Preda, E., Tropina, E., Groner, E., Lucot, E., Hornung, E., Gacia, E., Lévesque, E., Benedito, E., Davydov, E.A., Ampoorter, E., Bolzan, F.P., Varela, F., Kristöfel, F., Maestre, F.T., Maunoury-Danger, F., Hofhansl, F., Kitz, F., Sutter, F., Cuesta, F., De Almeida Lobo, F., De Souza, F.L., Berninger, F., Zehetner, F., Wohlfahrt, G., Vourlitis, G., Carreño-Rocabado, G., Arena, G., Pinha, G.D., González, G., Canut, G., Lee, H., Verbeeck, H., Auge, H., Pauli, H., Nacro, H.B., Bahamonde, H.A., Feldhaar, H., Jäger, H., Serrano, H.C., Verheyen, H., Bruelheide, H., Meesenburg, H., Jungkunst, H., Jactel, H., Shibata, H., Kurokawa, H., Rosas, H.L., Rojas Villalobos, H.L., Yesilonis, I., Melece, I., Van Halder, I., Quirós, I.G., Makelele, I., Senou, I., Fekete, I., Mihal, I., Ostonen, I., Borovská, J., Roales, J., Shoqeir, J., Lata, J.-C., Theurillat, J.-P., Probst, J.-L., Zimmerman, J., Vijayanathan, J., Tang, J., Thompson, J., Doležal, J., Sanchez-Cabeza, J.-A., Merlet, J., Henschel, J., Neirynck, J., Knops, J., Loehr, J., Von Oppen, J., Þorláksdóttir, J.S., Löffler, J., Cardoso-Mohedano, J.-G., Benito-Alonso, J.-L., Torezan, J.M., Morina, J.C., Jiménez, J.J., Quinde, J.D., Alatalo, J., Seeber, J., Stadler, J., Kriiska, K., Coulibaly, K., Fukuzawa, K., Szlavecz, K., Gerhátová, K., Lajtha, K., Käppeler, K., Jennings, K.A., Tielbörger, K., Hoshizaki, K., Green, K., Yé, L., Pazianoto, L.H.R., Dienstbach, L., Williams, L., Yahdjian, L., Brigham, L.M., Van Den Brink, L., Rustad, L., Zhang, L., Morillas, L., Xiankai, L., Carneiro, L.S., Di Martino, L., Villar, L., Bader, M.Y., Morley, M., Lebouvier, M., Tomaselli, M., Sternberg, M., Schaub, M., Santos-Reis, M., Glushkova, M., Torres, M.G.A., Giroux, M.-A., De Graaff, M.-A., Pons, M.-N., Bauters, M., Mazón, M., Frenzel, M., Didion, M., Wagner, M., Hamid, M., Lopes, M.L., Apple, M., Schädler, M., Weih, M., Gualmini, M., Vadeboncoeur, M.A., Bierbaumer, M., Danger, M., Liddell, M., Mirtl, M., Scherer-Lorenzen, M., Růžek, M., Carbognani, M., Di Musciano, M., Matsushita, M., Zhiyanski, M., Puşcaş, M., Barna, M., Ataka, M., Jiangming, M., Alsafran, M., Carnol, M., Barsoum, N., Tokuchi, N., Eisenhauer, N., Lecomte, N., Filippova, N., Hölzel, N., Ferlian, O., Romero, O., Pinto, O.B., Peri, P., Weber, P., Vittoz, P., Turtureanu, P.D., Fleischer, P., Macreadie, P., Haase, P., Reich, P., Petřík, P., Choler, P., Marmonier, P., Muriel, P., Ponette, Q., Guariento, R.D., Canessa, R., Kiese, R., Hewitt, R., Rønn, R., Adrian, R., Kanka, R., Weigel, R., Gatti, R.C., Martins, R.L., Georges, R., Meneses, R.I., Gavilán, R.G., Dasgupta, S., Wittlinger, S., Puijalon, S., Freda, S., Suzuki, S., Charles, S., Gogo, S., Drollinger, S., Mereu, S., Wipf, S., Trevathan-Tackett, S., Löfgren, S., Stoll, S., Trogisch, S., Hoeber, S., Seitz, S., Glatzel, S., Milton, S.J., Dousset, S., Mori, T., Sato, T., Ise, T., Hishi, T., Kenta, T., Nakaji, T., Michelan, T.S., Camboulive, T., Mozdzer, T.J., Scholten, T., Spiegelberger, T., Zechmeister, T., Kleinebecker, T., Hiura, T., Enoki, T., Ursu, T.-M., Di Cella, U.M., Hamer, U., Klaus, V.H., Rêgo, V.M., Di Cecco, V., Busch, V., Fontana, V., Piscová, V., Carbonell, V., Ochoa, V., Bretagnolle, V., Maire, V., Farjalla, V., Zhou, W., Luo, W., McDowell, W.H., Hu, Y., Utsumi, Y., Kominami, Y., Zaika, Y., Rozhkov, Y., Kotroczó, Z., Tóth, Z., 2018. Early stage litter decomposition across biomes. Sci. Total Environ. 628–629, 1369–1394. https://doi.org/10.1016/j.scitotenv.2018.01.012.

Do, A.T.N., Ha, Y., Kwon, J.-H., 2022. Leaching of microplastic-associated additives in aquatic environments: a critical review. Environ. Pollut. 305, 119258. https://doi.org/10.1016/j.envpol.2022.119258.

FAO, 2021. Assessment of Agricultural Plastics and Their Sustainability: a Call for Action. FAO. https://doi.org/10.4060/cb7856en.

Feng, X., Wang, Q., Sun, Y., Zhang, S., Wang, F., 2022. Microplastics change soil properties, heavy metal availability and bacterial community in a Pb-Zn-contaminated soil. J. Hazard. Mater. 424, 127364. https://doi.org/10.1016/j.jhazmat.2021.127364.

Fu, Q., Lai, J., Ji, X., Luo, Z., Wu, G., Luo, X., 2022. Alterations of the rhizosphere soil microbial community composition and metabolite profiles of Zea mays by polyethylene-particles of different molecular weights. J. Hazard. Mater. 423, 127062. https://doi.org/10.1016/j.jhazmat.2021.127062.

Gmach, M.R., Bolinder, M.A., Menichetti, L., Kätterer, T., Spiegel, H., Åkesson, O., Friedel, J.K., Surböck, A., Schweinzer, A., Sandén, T., 2024. Evaluating the Tea bag index approach for different management practices in agroecosystems using long-term field experiments in Austria and Sweden. SOIL 10, 407–423. https://doi.org/10.5194/soil-10-407-2024.

Gong, K., Peng, C., Hu, S., Xie, W., Chen, A., Liu, T., Zhang, W., 2024. Aging of biodegradable microplastics and their effect on soil properties: control from soil water. J. Hazard. Mater. 480, 136053. https://doi.org/10.1016/j.jhazmat.2024.136053.

Greenfield, L.M., Graf, M., Rengaraj, S., Bargiela, R., Williams, G., Golyshin, P.N., Chadwick, D.R., Jones, D.L., 2022. Field response of N2O emissions, microbial communities, soil biochemical processes and winter barley growth to the addition of conventional and biodegradable microplastics. Agric. Ecosyst. Environ. 336, 108023. https://doi.org/10.1016/j.agee.2022.108023.

Hahladakis, J.N., Velis, C.A., Weber, R., Iacovidou, E., Purnell, P., 2018. An overview of chemical additives present in plastics: migration, release, fate and environmental impact during their use, disposal and recycling. J. Hazard. Mater. 344, 179–199. https://doi.org/10.1016/j.jhazmat.2017.10.014.

Hann, S., Fletcher, E., Molteno, S., Sherrington, C., Elliot, L., Kong, M., Koite, A., Sastre, S., Martinez, V., 2021. Conventional and Biodegradable Plastics in Agriculture.

Hasan, M.M., Tarannum, M.N., 2025. Adverse impacts of microplastics on soil physicochemical properties and crop health in agricultural systems. J. Hazard. Mater. Adv. 17, 100528. https://doi.org/10.1016/j.hazadv.2024.100528.

Hofmann, T., Ghoshal, S., Tufenkji, N., Adamowski, J.F., Bayen, S., Chen, Q., Demokritou, P., Flury, M., Hüffer, T., Ivleva, N.P., Ji, R., Leask, R.L., Maric, M., Mitrano, D.M., Sander, M., Pahl, S., Rillig, M.C., Walker, T.R., White, J.C., Wilkinson, K.J., 2023. Plastics can be used more sustainably in agriculture. Commun. Earth Environ. 4, 332. https://doi.org/10.1038/s43247-023-00982-4.

Hu, X., Gu, H., Wang, Y., Liu, J., Yu, Z., Li, Y., Jin, J., Liu, X., Dai, Q., Wang, G., 2022. Succession of soil bacterial communities and network patterns in response to conventional and biodegradable microplastics: a microcosmic study in Mollisol. J. Hazard. Mater. 436, 129218. https://doi.org/10.1016/j.jhazmat.2022.129218.

Huang, Y., Liu, Q., Jia, W., Yan, C., Wang, J., 2020. Agricultural plastic mulching as a source of microplastics in the terrestrial environment. Environ. Pollut. 260, 114096. https://doi.org/10.1016/j.envpol.2020.114096.

Hund-Rinke, K., Simon, M., 2008. Bioavailability assessment of contaminants in soils via respiration and nitrification tests. Environ. Pollut. 153, 468–475. https://doi.org/10.1016/j.envpol.2007.08.003.

Hurley, R., Binda, G., Briassoulis, D., Carroccio, S.C., Cerruti, P., Convertino, F., Dvořáková, D., Kernchen, S., Laforsch, C., Löder, M.G.L., Pulkrabova, J., Schettini, E., Spanu, D., Tsagkaris, A.S., Vox, G., Nizzetto, L., 2024. Production and characterisation of environmentally relevant microplastic test materials derived from agricultural plastics. Sci. Total Environ. 946, 174325. https://doi.org/10.1016/j.scitotenv.2024.174325.

Hurley, R.R., Nizzetto, L., 2018. Fate and occurrence of micro(nano)plastics in soils: knowledge gaps and possible risks. Curr. Opin. Environ. Sci. Health 1, 6–11. https://doi.org/10.1016/j.coesh.2017.10.006.

Ihrmark, K., Bödeker, I.T.M., Cruz-Martinez, K., Friberg, H., Kubartova, A., Schenck, J., Strid, Y., Stenlid, J., Brandström-Durling, M., Clemmensen, K.E., Lindahl, B.D., 2012. New primers to amplify the fungal ITS2 region–evaluation by 454-sequencing of artificial and natural communities. FEMS Microbiol. Ecol. 82, 666–677. https://doi.org/10.1111/j.1574-6941.2012.01437.x.

ISO 10390, 2021. Soil, Treated Biowaste and Sludge - Determination of pH, 2021. ISO - International Organization for Standardization, Genève, p. 10.

ISO 11268-2, 2023. Soil Quality — Effects of Pollutants on Earthworms Part 2: Determination of Effects on Reproduction of Eisenia fetida/Eisenia Andrei and Other Earthworm Species.

ISO 14240-1, 1997. Soil Quality — Determination of Soil Microbial Biomass Part 1: Substrate-Induced Respiration Method.

ISO 15685, 2012. Soil Quality — Determination of Potential Nitrification and Inhibition of Nitrification — Rapid Test by Ammonium Oxidation.

ISO 16072, 2002. Soil Quality — Laboratory Methods for Determination of Microbial Soil Respiration.

ISO 18512, 2007. Soil Quality — Guidance on Long and Short Term Storage of Soil Samples, vol. 3. ISO - International Organization for Standardization, Genève.

Jia, X., Yao, Y., Tan, G., Xue, S., Liu, M., Tang, D.W.S., Geissen, V., Yang, X., 2024. Effects of LDPE and PBAT plastics on soil organic carbon and carbon-enzymes: a mesocosm experiment under field conditions. Environ. Pollut. 362, 124965. https://doi.org/10.1016/j.envpol.2024.124965.

Jiang, M., Zhao, W., Liang, Q., Cai, M., Fan, X., Jiang, Y., Li, T., Wang, Y., Peng, C., Liu, J., 2024. Advances in physiological and ecological effects of microplastic on crop. J. Soil Sci. Plant Nutr. 24, 1741–1760. https://doi.org/10.1007/s42729-024-01752-7.

Johannes, A., Matter, A., Schulin, R., Weisskopf, P., Baveye, P.C., Boivin, P., 2017. Optimal organic carbon values for soil structure quality of arable soils. Does clay content matter? Geoderma 302, 14–21. https://doi.org/10.1016/j.geoderma.2017.04.021.

Johansen, J.L., Magid, J., Vestergård, M., Palmqvist, A., 2024. Extent and effects of microplastic pollution in soil with focus on recycling of sewage sludge and composted household waste and experiences from the long-term field experiment CRUCIAL. TrAC, Trends Anal. Chem. 171, 117474. https://doi.org/10.1016/j.trac.2023.117474.

Karkanorachaki, K., Tsiota, P., Dasenakis, G., Syranidou, E., Kalogerakis, N., 2022. Nanoplastic generation from secondary PE microplastics: microorganism-induced fragmentation. Microplastics 1, 85–101. https://doi.org/10.3390/microplastics1010006.

Kasirajan, S., Ngouajio, M., 2012. Polyethylene and biodegradable mulches for agricultural applications: a review. Agron. Sustain. Dev. 32, 501–529. https://doi.org/10.1007/s13593-011-0068-3.

Kedzierski, M., Cirederf-Boulant, D., Palazot, M., Yvin, M., Bruzaud, S., 2023. Continents of plastics: an estimate of the stock of microplastics in agricultural soils. Sci. Total Environ. 880, 163294. https://doi.org/10.1016/j.scitotenv.2023.163294.

Kemper, W.D., Rosenau, R.C., 2018. Aggregate stability and size distribution. In: Klute, A. (Ed.), SSSA Book Series. Soil Science Society of America, American Society of Agronomy, Madison, WI, USA, pp. 425–442. https://doi.org/10.2136/sssabookser5.1.2ed.c17.

Keuskamp, J.A., Dingemans, B.J.J., Lehtinen, T., Sarneel, J.M., Hefting, M.M., 2013. Tea bag index: a novel approach to collect uniform decomposition data across ecosystems. Methods Ecol. Evol. 4, 1070–1075. https://doi.org/10.1111/2041-210X.12097.

Kim, S.W., Šmídová, K., van Loon, S., van Gestel, C.A.M., Rillig, M.C., Fritze, H., Velmala, S. Effects of biodegradable microplastics on soil microbial communities and activities: insight from an ecological mesocosm experiment. STOTEN 975, 179288. https://doi.org/10.1016/j.scitotenv.2025.179288.

Koorneef, G.J., De Goede, R.G., Pulleman, M.M., Van Leeuwen, A.G., Barré, P., Baudin, F., Comans, R.N., 2023. Quantifying organic carbon in particulate and mineral-associated fractions of calcareous soils – a method comparison. Geoderma 436, 116558. https://doi.org/10.1016/j.geoderma.2023.116558.

Koutnik, V.S., Leonard, J., Alkidim, S., DePrima, F.J., Ravi, S., Hoek, E.M.V., Mohanty, S. K., 2021. Distribution of microplastics in soil and freshwater environments: global analysis and framework for transport modeling. Environ. Pollut. 274, 116552. https://doi.org/10.1016/j.envpol.2021.116552.

Li, C., Gan, Y., Zhang, C., He, H., Fang, J., Wang, L., Wang, Y., Liu, J., 2021. "Microplastic communities" in different environments: differences, links, and role of diversity index in source analysis. Water Res. 188, 116574. https://doi.org/10.1016/j.watres.2020.116574.

Li, Shitong, Ding, F., Flury, M., Wang, Z., Xu, L., Li, Shuangyi, Jones, D.L., Wang, J., 2022. Macro- and microplastic accumulation in soil after 32 years of plastic film mulching. Environ. Pollut. 300, 118945. https://doi.org/10.1016/j.envpol.2022.118945.

Liang, Y., Lehmann, A., Yang, G., Leifheit, E.F., Rillig, M.C., 2021. Effects of microplastic fibers on soil aggregation and enzyme activities are organic matter dependent. Front. Environ. Sci. 9, 650155. https://doi.org/10.3389/fenvs.2021.650155.

Lin, D., Yang, G., Dou, P., Qian, S., Zhao, L., Yang, Y., Fanin, N., 2020. Microplastics negatively affect soil fauna but stimulate microbial activity: insights from a field-based microplastic addition experiment. Proc. R. Soc. B Biol. Sci. 287, 20201268. https://doi.org/10.1098/rspb.2020.1268.

Liu, H., Yang, X., Liu, G., Liang, C., Xue, S., Chen, H., Ritsema, C.J., Geissen, V., 2017. Response of soil dissolved organic matter to microplastic addition in Chinese loess soil. Chemosphere 185, 907–917. https://doi.org/10.1016/j.chemosphere.2017.07.064.

Liu, R., Liang, J., Yang, Y., Jiang, H., Tian, X., 2023. Effect of polylactic acid microplastics on soil properties, soil microbials and plant growth. Chemosphere 329, 138504. https://doi.org/10.1016/j.chemosphere.2023.138504.

Lobe, I., Sandhage-Hofmann, A., Brodowski, S., Du Preez, C.C., Amelung, W., 2011. Aggregate dynamics and associated soil organic matter contents as influenced by prolonged arable cropping in the South African Highveld. Geoderma 162, 251–259. https://doi.org/10.1016/j.geoderma.2011.02.001.

Lv, M., Chen, H., Liang, Z., Sun, A., Lu, S., Ren, S., Zhu, D., Wei, S., Chen, L., Ding, J., 2024. Stress of soil moisture and temperature exacerbates the toxicity of tire wear particles to soil fauna: tracking the role of additives through host microbiota. J. Hazard. Mater. 480, 135995. https://doi.org/10.1016/j.jhazmat.2024.135995.

Ma, Jie-wen, Wu, Y., Xu, C.-L., Luo, Z., Yu, R., Hu, G., Yan, Y., 2023a. Inhibitory effect of polyethylene microplastics on roxarsone degradation in soils. J. Hazard. Mater. 454, 131483. https://doi.org/10.1016/j.jhazmat.2023.131483.

Ma, Jing, Xu, M., Wu, J., Yang, G., Zhang, X., Song, C., Long, L., Chen, C., Xu, C., Wang, Y., 2023b. Effects of variable-sized polyethylene microplastics on soil chemical properties and functions and microbial communities in purple soil. Sci. Total Environ. 868, 161642. https://doi.org/10.1016/j.scitotenv.2023.161642.

Ma, R., Xu, Z., Sun, J., Li, D., Cheng, Z., Niu, Y., Guo, H., Zhou, J., Wang, T., 2023c. Microplastics affect C, N, and P cycling in natural environments: highlighting the driver of soil hydraulic properties. J. Hazard. Mater. 459, 132326. https://doi.org/10.1016/j.jhazmat.2023.132326.

Massaccesi, L., Marabottini, R., Feudis, M. De, Leccese, A., Poesio, C., Marinari, S., Moscatelli, M.C., Agnelli, A., 2025. Impact of high-density polyethylene (HDPE) microparticles on soil physical-chemical properties, CO2 emissions, and microbial community in a two-year field trial. STOTEN 966, 178703. https://doi.org/10.1016/j.scitotenv.2025.178703.

Meizoso-Regueira, T., Fuentes, J., Cusworth, S.J., Rillig, M.C., 2024. Prediction of future microplastic accumulation in agricultural soils. Environ. Pollut. 359, 124587. https://doi.org/10.1016/j.envpol.2024.124587.

Meng, F., Yang, X., Riksen, M., Geissen, V., 2022. Effect of different polymers of microplastics on soil organic carbon and nitrogen – a mesocosm experiment. Environ. Res. 204, 111938. https://doi.org/10.1016/j.envres.2021.111938.

Morris, E.K., Morris, D.J.P., Vogt, S., Gleber, S.-C., Bigalke, M., Wilcke, W., Rillig, M.C., 2019. Visualizing the dynamics of soil aggregation as affected by arbuscular mycorrhizal fungi. ISME J. 13, 1639–1646. https://doi.org/10.1038/s41396-019-0369-0.

Nizzetto, L., Futter, M., Langaas, S., 2016. Are agricultural soils dumps for microplastics of urban origin? Environ. Sci. Technol. 50, 10777–10779. https://doi.org/10.1021/acs.est.6b04140.

Oades, J., Waters, A., 1991. Aggregate hierarchy in soils. Soil Res. 29, 815. https://doi.org/10.1071/SR9910815.

PAPILLONS, 2024. Plastics in agriculture: impacts, lifecycles & long-term sustainability. URL. https://www.papillons-h2020.eu/.

Parada, A.E., Needham, D.M., Fuhrman, J.A., 2016. Every base matters: assessing small subunit rRNA primers for marine microbiomes with mock communities, time series and global field samples. Environ. Microbiol. 18, 1403–1414. https://doi.org/10.1111/1462-2920.13023.

Pfohl, P., Wagner, M., Meyer, L., Domercq, P., Praetorius, A., Hüffer, T., Hofmann, T., Wohlleben, W., 2022. Environmental degradation of microplastics: how to measure fragmentation rates to secondary Micro- and nanoplastic fragments and dissociation into dissolved organics. Environ. Sci. Technol. 56, 11323–11334. https://doi.org/10.1021/acs.est.2c01228.

Piehl, S., Leibner, A., Löder, M.G.J., Dris, R., Bogner, C., Laforsch, C., 2018. Identification and quantification of macro- and microplastics on an agricultural farmland. Sci. Rep. 8, 17950. https://doi.org/10.1038/s41598-018-36172-y.

Plastics Europe, 2022. Plastics - the Facts 2021, an Analysis of European Plastics Production, Demand and Waste Data, Report 2021.

Qi, Y., Ossowicki, A., Yang, X., Huerta Lwanga, E., Dini-Andreote, F., Geissen, V., Garbeva, P., 2020. Effects of plastic mulch film residues on wheat rhizosphere and soil properties. J. Hazard. Mater. 387, 121711. https://doi.org/10.1016/j.jhazmat.2019.121711.

Qiu, Y., Zhou, S., Zhang, C., Zhou, Y., Qin, W., 2022. Soil microplastic characteristics and the effects on soil properties and biota: a systematic review and meta-analysis. Environ. Pollut. 313, 120183. https://doi.org/10.1016/j.envpol.2022.120183.

Quast, C., Pruesse, E., Yilmaz, P., Gerken, J., Schweer, T., Yarza, P., Peplies, J., Glöckner, F.O., 2012. The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. Nucleic Acids Res. 41, D590–D596. https://doi.org/10.1093/nar/gks1219.

Quince, C., Lanzen, A., Davenport, R.J., Turnbaugh, P.J., 2011. Removing noise from pyrosequenced amplicons. BMC Bioinf. 12, 38. https://doi.org/10.1186/1471-2105-12-38.

R Core Team, 2024. R: a Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/.

Rauscher, A., Meyer, N., Jakobs, A., Bartnick, R., Lueders, T., Lehndorff, E., 2023. Biodegradable microplastic increases CO2 emission and alters microbial biomass and bacterial community composition in different soil types. Appl. Soil Ecol. 182, 104714. https://doi.org/10.1016/j.apsoil.2022.104714.

Reay, M.K., Greenfield, L.M., Graf, M., Lloyd, C.E.M., Evershed, R.P., Chadwick, D.R., Jones, D.L., 2023. LDPE and biodegradable PLA-PBAT plastics differentially affect plant-soil nitrogen partitioning and dynamics in a Hordeum vulgare mesocosm. J. Hazard. Mater. 447, 130825. https://doi.org/10.1016/j.jhazmat.2023.130825.

Ren, X., Tang, J., Liu, X., Liu, Q., 2020. Effects of microplastics on greenhouse gas emissions and the microbial community in fertilized soil. Environ. Pollut. 256, 113347. https://doi.org/10.1016/j.envpol.2019.113347.

Rillig, M.C., Kim, S.W., Zhu, Y.-G., 2024. The soil plastisphere. Nat. Rev. Microbiol. 22, 64–74. https://doi.org/10.1038/s41579-023-00967-2.

Rillig, M.C., Lehmann, A., De Souza Machado, A.A., Yang, G., 2019. Microplastic effects on plants. New Phytol. 223, 1066–1070. https://doi.org/10.1111/nph.15794.

Rillig, M.C., Hoffmann, M., Lehmann, A., Liang, Y., Lück, M., Augustin, J., 2021. Microplastic fibers affect dynamics and intensity of CO2 and N2O fluxes from soil differently. Microplastics Nanoplastics 1, 3. https://doi.org/10.1186/s43591-021-00004-0.

Ros, G.H., Hanegraaf, M.C., Hoffland, E., Van Riemsdijk, W.H., 2011. Predicting soil N mineralization: relevance of organic matter fractions and soil properties. Soil Biol. Biochem. 43, 1714–1722. https://doi.org/10.1016/j.soilbio.2011.04.017.

Sander, M., Weber, M., Lott, C., Zumstein, M., Künkel, A., Battagliarin, G., 2023. Polymer biodegradability 2.0: a holistic view on polymer biodegradation in natural and engineered environments. In: Künkel, A., Battagliarin, G., Winnacker, M., Rieger, B., Coates, G. (Eds.), Synthetic Biodegradable and Biobased Polymers, Advances in Polymer Science. Springer International Publishing, Cham, pp. 65–110. https://doi.org/10.1007/12_2023_163.

Schell, T., Rico, A., Vighi, M., 2020. Occurrence, fate and fluxes of plastics and microplastics in terrestrial and freshwater ecosystems. In: De Voogt, P. (Ed.), Reviews of Environmental Contamination and Toxicology Volume 250, Reviews of Environmental Contamination and Toxicology. Springer International Publishing, Cham, pp. 1–43. https://doi.org/10.1007/398_2019_40.

Scheurer, M., Bigalke, M., 2018. Microplastics in Swiss floodplain soils. Environ. Sci. Technol. 52, 3591–3598. https://doi.org/10.1021/acs.est.7b06003.

Schiedung, H., Bornemann, L., Welp, G., 2017. Seasonal variability of soil organic carbon fractions under arable land. Pedosphere 27, 380–386. https://doi.org/10.1016/S1002-0160(17)60326-6.

Schweizer, S.A., Bucka, F.B., Graf-Rosenfellner, M., Kögel-Knabner, I., 2019. Soil microaggregate size composition and organic matter distribution as affected by clay content. Geoderma 355, 113901. https://doi.org/10.1016/j.geoderma.2019.113901.

Shi, J., Wang, J., Lv, J., Wang, Z., Peng, Y., Wang, X., 2022. Microplastic presence significantly alters soil nitrogen transformation and decreases nitrogen bioavailability under contrasting temperatures. J. Environ. Manag. 317, 115473. https://doi.org/10.1016/j.jenvman.2022.115473.

Siddiqui, S.A., Singh, S., Bahmid, N.A., Shyu, D.J.H., Domínguez, R., Lorenzo, J.M., Pereira, J.A.M., Câmara, J.S., 2023. Polystyrene microplastic particles in the food chain: characteristics and toxicity - a review. Sci. Total Environ. 892, 164531. https://doi.org/10.1016/j.scitotenv.2023.164531.

Šmídová, K., Selonen, S., Van Gestel, C.A.M., Fleissig, P., Hofman, J., 2024. Microplastics originated from agricultural mulching films affect enchytraeid multigeneration reproduction and soil properties. J. Hazard. Mater. 479, 135592. https://doi.org/10.1016/j.jhazmat.2024.135592.

Soinne, H., Keskinen, R., Räty, M., Kanerva, S., Turtola, E., Kaseva, J., Nuutinen, V., Simojoki, A., Salo, T., 2021. Soil organic carbon and clay content as deciding factors for net nitrogen mineralization and cereal yields in boreal mineral soils. Eur. J. Soil Sci. 72, 1497–1512. https://doi.org/10.1111/ejss.13003.

Su, P., Gao, C., Zhang, X., Zhang, D., Liu, X., Xiang, T., Luo, Y., Chu, K., Zhang, G., Bu, N., Li, Z., 2023. Microplastics stimulated nitrous oxide emissions primarily through denitrification: a meta-analysis. J. Hazard. Mater. 445, 130500. https://doi.org/10.1016/j.jhazmat.2022.130500.

Sun, X., Zhang, X., Xia, Y., Tao, R., Zhang, M., Mei, Y., Qu, M., 2022. Simulation of the effects of microplastics on the microbial community structure and nitrogen cycle of paddy soil. Sci. Total Environ. 818, 151768. https://doi.org/10.1016/j.scitotenv.2021.151768.

Thompson, R.C., Courtene-Jones, W., Boucher, J., Pahl, S., Raubenheimer, K., Koelmans, A.A., 2024. Twenty years of microplastic pollution research—what have we learned? Science 386, eadl2746. https://doi.org/10.1126/science.adl2746.

Wang, F., Wang, Q., Adams, C.A., Sun, Y., Zhang, S., 2022. Effects of microplastics on soil properties: current knowledge and future perspectives. J. Hazard. Mater. 424, 127531. https://doi.org/10.1016/j.jhazmat.2021.127531.

Weithmann, N., Möller, J.N., Löder, M.G.J., Piehl, S., Laforsch, C., Freitag, R., 2018. Organic fertilizer as a vehicle for the entry of microplastic into the environment. Sci. Adv. 4, eaap8060. https://doi.org/10.1126/sciadv.aap8060.

White, T.J., Bruns, T., Lee, S., Taylor, J., 1990. 38 - amplification and direct sequencing of fungal ribosomal rna genes for phylogenetics. In: Innis, M.A., Gelfand, D.H., Sninsky, J.J., White, Thomas J. (Eds.), PCR Protocols. Academic Press, San Diego, pp. 315–322. https://doi.org/10.1016/B978-0-12-372180-8.50042-1.

Wrigley, O., Braun, M., Amelung, W., 2024. Global soil microplastic assessment in different land-use systems is largely determined by the method of analysis: a meta-analysis. Sci. Total Environ. 957, 177226. https://doi.org/10.1016/j.scitotenv.2024.177226.

Xiang, Y., Peñuelas, J., Sardans, J., Liu, Y., Yao, B., Li, Y., 2023. Effects of microplastics exposure on soil inorganic nitrogen: a comprehensive synthesis. J. Hazard. Mater. 460, 132514. https://doi.org/10.1016/j.jhazmat.2023.132514.

Yang, B., Wu, L., Feng, W., Lin, Q., 2024. Global perspective of ecological risk of plastic pollution on soil microbial communities. Front. Microbiol. 15, 1468592. https://doi.org/10.3389/fmicb.2024.1468592.

Yang, C., Yang, R., Feng, Y., Wang, Y., Zou, Q., Song, J., Duan, J., Li, H., Gao, X., Chen, M., Zhang, H., 2025. Microplastics affect organic nitrogen in sediment: the response of organic nitrogen mineralization to microbes and benthic animals. J. Hazard. Mater. 485, 136926. https://doi.org/10.1016/j.jhazmat.2024.136926.

Yu, Y., Battu, A.K., Varga, T., Denny, A.C., Zahid, T.Md, Chowdhury, I., Flury, M., 2023. Minimal impacts of microplastics on soil physical properties under environmentally relevant concentrations. Environ. Sci. Technol. 57, 5296–5304. https://doi.org/10.1021/acs.est.2c09822.

Zeb, A., Liu, W., Ali, N., Shi, R., Wang, Q., Wang, J., Li, J., Yin, C., Liu, Jinzheng, Yu, M., Liu, Jianv, 2024. Microplastic pollution in terrestrial ecosystems: global implications and sustainable solutions. J. Hazard. Mater. 461, 132636. https://doi.org/10.1016/j.jhazmat.2023.132636.

Zhang, X., Li, Y., Lei, J., Li, Z., Tan, Q., Xie, L., Xiao, Y., Liu, T., Chen, X., Wen, Y., Xiang, W., Kuzyakov, Y., Yan, W., 2023. Time-dependent effects of microplastics on soil bacteriome. J. Hazard. Mater. 447, 130762. https://doi.org/10.1016/j.jhazmat.2023.130762.

Zhang, X., Wu, D., Jiang, X., Xu, J., Liu, J., 2024a. Source, environmental behavior and ecological impact of biodegradable microplastics in soil ecosystems: a review. Rev. Environ. Contam. Toxicol. 262, 6. https://doi.org/10.1007/s44169-023-00057-7.

Zhang, Z., Peng, L., Gao, W., Wu, D., Zhang, W., Wang, X., Liu, H., Li, Q., Fan, C., Chen, M., 2024b. PBAT microplastics exacerbates N2O emissions from tropical latosols mainly via stimulating denitrification. Chem. Eng. J. 495, 153681. https://doi.org/10.1016/j.cej.2024.153681.

Zhao, T., Lozano, Y.M., Rillig, M.C., 2021. Microplastics increase soil pH and decrease microbial activities as a function of microplastic shape, polymer type, and exposure time. Front. Environ. Sci. 9, 675803. https://doi.org/10.3389/fenvs.2021.675803.

Zhu, F., Zhu, C., Wang, C., Gu, C., 2019. Occurrence and ecological impacts of microplastics in soil systems: a review. Bull. Environ. Contam. Toxicol. 102, 741–749. https://doi.org/10.1007/s00128-019-02623-z.

Zumstein, M.T., Schintlmeister, A., Nelson, T.F., Baumgartner, R., Woebken, D., Wagner, M., Kohler, H.-P.E., McNeill, K., Sander, M., 2018. Biodegradation of synthetic polymers in soils: tracking carbon into CO_2 and microbial biomass. Sci. Adv. 4, eaas9024. https://doi.org/10.1126/sciadv.aas9024.