Wolf *et al. Environmental Sciences Europe* (2025) 37:13
https://doi.org/10.1186/s12302-024-01050-0

Environmental Sciences Europe

RESEARCH

Open Access

iMulch: an investigation of the influence of polymers on a terrestrial ecosystem using the example of mulch films used in agriculture

Carmen Wolf[1*], Mike Wenzel[1], Björn Fischer[2], Ralf Bertling[3], Erich Jelen[3], Dieter Hennecke[4], Karlheinz Weinfurtner[4], Martina Roß-Nickoll[5], Henner Hollert[4,5,7], Antonia Weltmeyer[5], Kristina Bitter[6], Pauline Ruiz[8], Dusica Banduka[8], Jochen Tuerk[1] and Lars M. Blank[6*]

Abstract

Background This article provides an overview of the iMulch joint project, which analysed the use of polyethylene (PE) and biodegradable mulch films made of a polylactide (PLA) and polybutylene adipate terephthalate (PBAT) on agricultural land as a source of microplastic. The development of a detection methodology using Raman spectroscopy and thermo-extraction desorption gas chromatography mass–spectrometry (TED-GC–MS), the adsorption behaviour, ageing in drainage water and soil, their transport behaviour in lysimeters, ecotoxicity, uptake in plants, a life cycle assessment (LCA) and upcycling were considered.

Results The PE film tested showed hardly any degradation or fragmentation during the ageing tests. The biodegradable films showed incipient degradation after 8 weeks in drainage water and initial degradation after 12 weeks in soil ageing experiments. Additionally no degradation could be detected in the lysimeter test within the 24 months analysed. The biodegradable films could be metabolized in laboratory tests with some microorganisms present in the soil. This indicates that these films can be degraded in the environment if the conditions for degradation are optimal. No microorganisms or fungi that could degrade the PE film within a respective period of time were detected in the soil. Adsorption of the tested substances was not observed. Incorporated in soil, mulch film microplastic showed retention of extractable pesticides. In the ecotoxicological tests, both film types showed no acute toxic effects in the earthworm *Eisenia fetida* and the springtail *Folsomia candida*. Endocrine activity was observed in eluate samples from both films. However, aged films showed fewer effects than non-aged films.

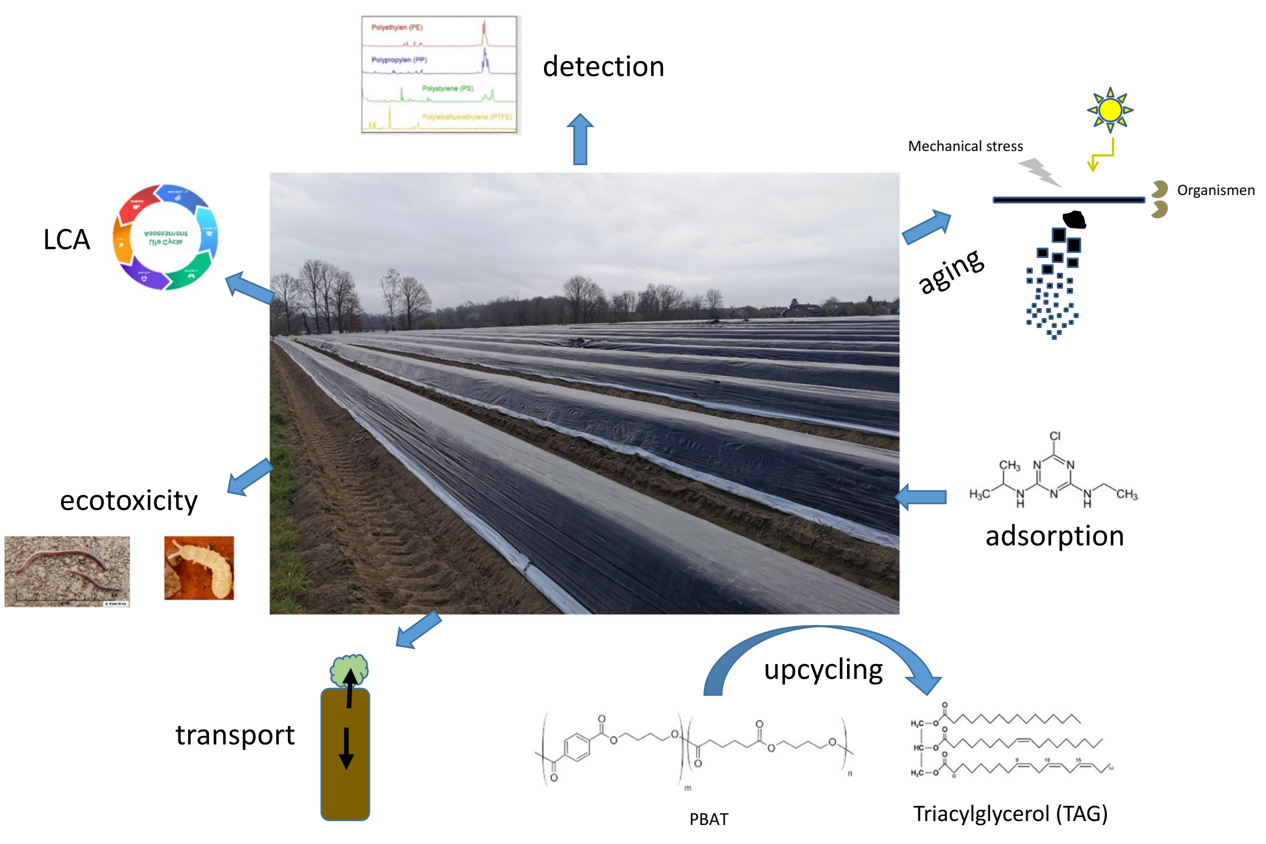
Conclusion Both types of film show no transport or degradation in the tests under real conditions, which means that they remain in the upper soil layer, where they are available to soil organisms and can lead to high concentrations in the future. As the biodegradable film could be degraded, at least under ideal conditions, we recommend its use. However, proof of degradation must first be verified under real field conditions. In addition, we recommend the use of thicker conventional mulch films to minimize the emission of plastic particles. For this purpose, a minimum lower limit for the material thickness should be defined.

Keywords Microplastic, Agriculture, Mulch films, Soils, Biodegradable plastic

---

*Correspondence:
Carmen Wolf
wolf@iuta.de
Lars M. Blank
lars.blank@rwth-aachen.de
Full list of author information is available at the end of the article

Graphical Abstract



Background

The production of plastic increased during the last decades from 1.7 million t in 1950 to about 400 million t in 2022 worldwide [1]. Clearly, with the myriad of applications no plastic-free future can be envisaged. However, due to the high production volumes and the unclear end-of-life, plastics are a major challenge, as acknowledged by the UN. In countries with suboptimal waste logistics, the challenge is omnipresent, while in countries with good waste logistics, the impact on organisms and whole ecosystems is more subtle. Plastic can enter the environment by wear and tear and littering or by purpose, as seen by the ever-growing use of plastic in the agricultural sector.

Mulch films are an example of agricultural plastics, with increasing applications. They serve to regulate temperature and moisture in the soil, prevent weed growth and soil erosion during heavy rainfall, and protect crops from pests and predators [2]. It is expected that the use of mulch films in agriculture will increase in the course of climate change and the targeted reduction in the use of pesticides [3]. In Europe, about 4270 km^2 of agricultural land is covered with mulch films, with polyethylene (PE) as the dominant material. PE mulch film is often modified with additives such as plasticizers, UV stabilizers, and colour pigments to achieve favourable properties [2]. Most mulch film is removed from the field after use. However, the thinner the film, the more difficult and time-consuming it is to remove the entire mulch film from the field after the harvest cycle [4]. For this reason, fragments of mulch film are intentionally or unintentionally left in the field and can break down into microplastic fragments.

While unintentional emissions cannot be directly regulated, the intentional use of plastic products in the environment can be regulated if necessary. Therefore, it is important to investigate these uses, identify, and quantify possible emissions of microplastic. Finally, a risk assessment of the technology comparing the advantages and disadvantages is required.

Whether the use of mulch films on agricultural land can have a negative effect on the soil ecosystem was analysed from various perspectives in the joint project "iMulch—an investigation of the influence of polymers on a terrestrial ecosystem using the example of mulch films used in agriculture". The project is a Leitmarkt. NRW project and was funded by the European Regional Development Fund (ERDF) "Investments in Growth and Employment".

The joint project was carried out and supported by a consortium consisting of six active project partners and three associated partners. In addition to the Institut für Umwelt & Energie, Technik & Analytik e.V. (IUTA), the two Fraunhofer Institutes IME and UMSICHT, RWTH Aachen University with the Institutes for Environmental Research (IFER) and the Institute for Applied Microbiology (iAMB), the company FISCHER GmbH and the nova-Institut were involved. Associated partners were the Research Institute Plastics and Recycling (FkuR), the company BASF and the Federal Environment Agency. There was also a lively exchange with the NRW Chamber of Agriculture.

The project analysed a PE film and two biodegradable plastic films [PLA/PBAT blend—polylactide (PLA) and fossil-based but biodegradable polybutylene adipate terephthalate (PBAT)] provided by the associated partners. The comparison between the films also served to analyse whether and to what extent biodegradable films are ecologically advantageous compared to conventional films.

In a first step, a detection methodology was developed, which included the establishment and validation of a sample preparation, analysis and evaluation method for TED-GC–MS and a suitable sample preparation and analysis method for Raman microspectroscopy (µRaman). This makes it possible to determine the concentration, size distribution, morphology and type of polymers in soils. Weathering and ageing of the films, transport behaviour and possible ecotoxicity were also investigated. A life cycle assessment was also part of the project scope, as was the attempt to identify an alternative recycling path through upcycling. Recommendations were derived from the results in order to reduce future emissions from mulch films into the environment. In this article, an overview of the overall strategy and the results is presented, while we refer to specific references for details.

Materials and methods

Test fields

As part of the project, in addition to spiked reference soils, real soils should also be sampled and measured in order to be able to estimate the concentration of microplastics in real cultivated soils. In addition, these samples should be used to test the hypothesis that the form of cultivation has an influence on the concentration of microplastics in the soil.

Unfortunately, it was not possible to find suitable fields within the scope of the project, so the real samples were taken from test fields of the NRW Chamber of Agriculture.

One advantage of these fields was that the cultivation history of these fields over the last 10 years was known, so that suitable fields could be selected to test the hypothesis. The fields are cultivated normally but are smaller in comparison to non-test fields. As a result, it is easier for farmers to find and manually remove any film residues after use. These fields therefore represent a best-case scenario.

Sampling

The sampling was carried out in accordance with the soil science mapping guidelines for nutrients. For this purpose, up to 5 kg of soil of the first 30 cm of the soil was collected using a Pürkhauer sampler. The sampling was carried out diagonally across the surface of the field. The soil was collected in a metal bucket.

If plastic particles were sighted on the surface of the soil during sampling, this was noted and collected.

The soils samples were mixed and one part of the sample was characterized for the soil characteristics the other part was used for the microplastic analysis.

Microplastic analysis

A sample preparation and detection method was developed with which microplastic could be detected in soils, primarily PE and PLA/PBAT. A combination of spectroscopy and mass spectrometry seemed to be ideal to gain information about the size, shape, particle number and mass quantity of microplastics. A sample preparation method was established for both TED-GC–MS and µRaman.

For thermoanalytical methods like pyrolysis or TED-GC–MS the reduction of matrix is often not necessary. However, a microplastic enrichment step through the application of a sample preparation method can be helpful to reach the limit of quantification (LOQ) of the measurement systems. The LOQ was 1 µg/g for PE and 0.1 µg/g for PLA/PBAT.

For spectroscopic methods a reduction of the matrix is mandatory, since single particles need to be analysed. However, lower amounts of polymer particles can be detected by using these techniques. Due to these two different preparation methods were established.

A summary of the developed sample preparation workflows can be seen in Fig. 1. The workflow concerning Raman microspectroscopy analysis is already published

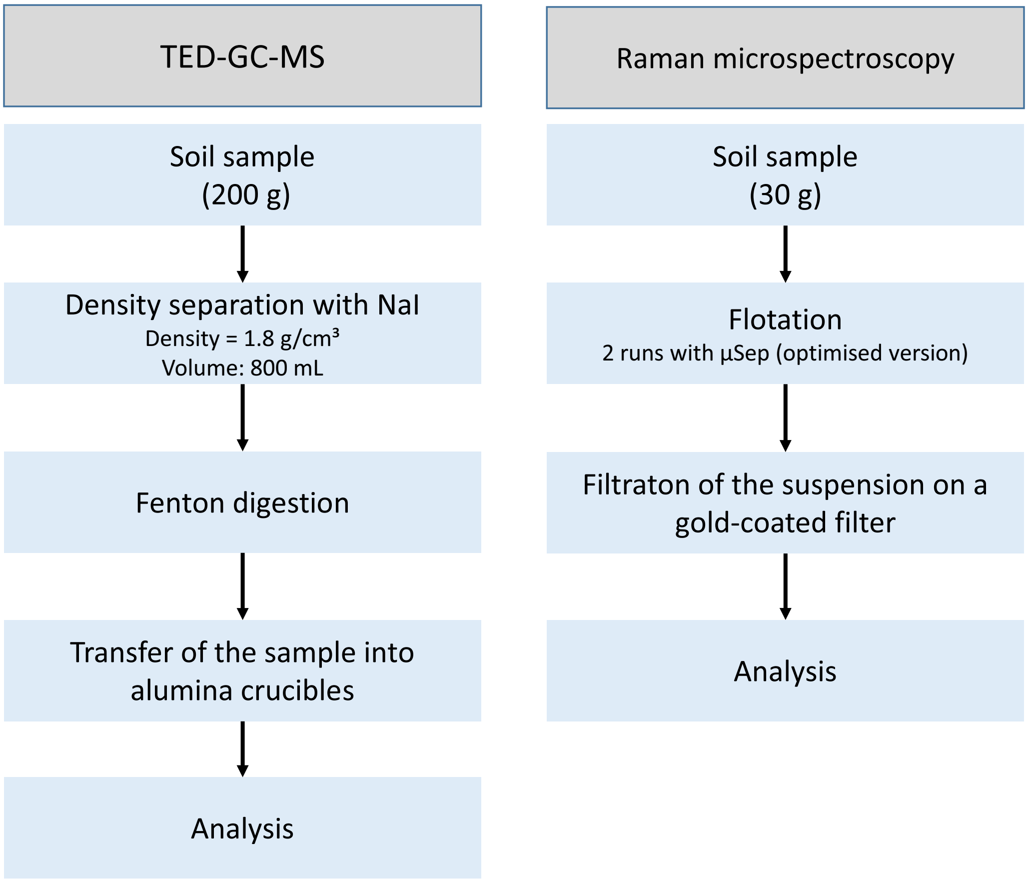


Fig. 1 Workflow for the sample preparation methods for TED-GC–MS and Raman microspectroscopy

and can be seen in detail within the study of Wenzel et al. [5].

The recovery rates for the sample preparation method for the TED-GC–MS method was investigated using the Reference soil A01 and the two different mulch film types (PE and biodegradable film—PLA/PBAT). An average recovery rate of 93% for PLA/PBAT and 109% for PE was determined. Recovery rates for the Raman microspectroscopy workflow was analysed by spiking reference particles to the systems. For polyethylene terephthalate (PET), a recovery rate of 77% ± 11%, for polystyrene (PS), 42% ± 11%, and for polypropylene (PP) 65% ± 12% was detected. A detailed description can be found elsewhere [5].

Sample preparation

Density separation

Density separation is based on increasing the density of the medium used by mixing water with a salt, e.g. sodium chloride (NaCl–water mixture 1.19 g/cm^3) or sodium iodide (NaI–water mixture 1.8 g/cm^3). Heavy matrix components sink when a sample is added, while the light fraction containing microplastic floats up, which can be separated and filtered.

In order to achieve a sufficiently large density difference between polymer and solution, sodium iodide was used. For this purpose, 200 g of the soil was mixed with 800 ml of a saturated sodium iodide (NaI) solution and transferred to a 1000-ml beaker. The soil–NaI mixture was stirred for 5 min and covered with aluminium foil. After standing for 1 h, the sample was stirred again for 5 min and then left to stand for 24 h.

The supernatant was collected with a test sieve with a 50 µm mesh size and filtered on a 47 mm cellulose filter by washing with a Tween/ultrapure water solution by vacuum filtration. The separated matrix was transferred to a petri dish and was homogenized afterwards. To reduce the organic-rich components of the sample, the homogenized sample is digested using Fenton's digestion.

Fenton's digestion

Fenton's digestion is based on oxidizing the organic matrix components, catalysed by iron II in an acidic medium. The combination of $FeSO_4$ (solution) and hydrogen peroxide (H_2O_2) produce hydroxyl radicals, needed to degrade the organic components. Fenton digestion has already been used in own work within the BMBF project SubμTrack for matrix reduction concerning wastewater samples [6] and adapted to the matrix soil for the iMulch project. Compared to the implementation according to Al Azzawi et al. [7], the volumes of the reagents used were adjusted to the sample quantity. The samples should be is completely wetted with the catalyst solution to ensure a homogeneous reaction. The volumes of hydrogen peroxide were therefore adjusted concerning the volume needed of the catalyst solution. After the Fenton digestion, the sample was ground with an agate grinder and an aliquot of 10 mg was taken for TED-GC–MS analysis.

Flotation

The basic concept of flotation was adapted from an enrichment step in mineral processing, where rising air bubbles in water are suitable for separating ore particles from stone. The air bubbles adhere to the surface of hydrophobic particles. Therefore, the air bubbles can serve as vertical transporters for hydrophobic particles [8, 9]. Due to the hydrophobic properties of most polymers, this process also seemed to be a suitable enrichment step for the analysis of microplastic in environmental samples. A detailed description of the method can be found elsewhere [5]. Briefly, the flotation apparatus designed by Renner et al. [10] for sediments was adapted to the matrix soil. However, in this project an improved version of the apparatus was used, which was completely made from glass. The used separator (named μSEP) is designed as a closed circuit, i.e. at the bottom of a separation column all particles enter a thin bypass which is used for the vertical transport of the whole sample and the subsequent reintroduction into the separation column. This transport is realized by a strong stream of large air bubbles. In contrast to the fine stream of air bubbles in the separation column, these large air bubbles in the transport bypass adapt to the column diameter and ensure that all particles are forced vertically in a non-destructive manner. The closed loop allows for multiple microplastic–air bubble interaction cycles within the separation column, which can improve separation. In addition, the outlet, diffuser and gas/liquid separator can be removed or replaced to adjust system parameters and clean or replace defective parts. The hydrophobic microplastic particles rise via the air bubble flow and pass the outlet, where they are collected at an overflow on a filter.

Detection

Specific software programmes were used for the individual analysis procedures. Details on the data analysis are listed in the individual associated publications.

TED-GC–MS

In the context of TED-GC–MS analysis, the employed analytical methodology is firmly established and was concurrently utilized in previous studies of the authors [5]. Pyrolysis of samples was conducted utilizing a thermogravimetric furnace (TGA 2, Mettler Toledo GmbH, Gießen, Germany). Within this process, the sample of interest was situated in a 600-μl alumina crucible (Mettler Toledo GmbH, Gießen, Germany). Subsequent to pyrolysis, the resultant pyrolysis products underwent transfer through an interface to an adsorption unit employing a nitrogen flow. The adsorption transpired onto a solid-phase stir bar adsorber constructed from polydimethylsiloxane.

The automation of this process involved the utilization of a MultiPurposeSampler autosampler robot, facilitating the transfer of the adsorber to the thermal desorption unit. All instrumental components, including the interface, adsorber, adsorption and desorption units, as well as the MultiPurposeSampler, were procured commercially from Gerstel GmbH & Co KG, Mülheim an der Ruhr, Germany. Subsequently, the desorption of pyrolysis products ensued, followed by cryofocusing (CIS4, Gerstel GmbH & Co KG, Mülheim an der Ruhr, Germany), and injection into the gas chromatograph (GC7890, Agilent, Santa Clara, California, USA). Post chromatographic separation, the pyrolysis products were subjected to analysis using a mass spectrometer (5977B MSD, Agilent). Comprehensive details pertaining to the setup parameters are delineated in prior studies authored by the investigators. For identification and quantification, well-known marker compounds of the polymers at its specific retention times were used.

Raman microspectroscopy

A detailed description of the analytical method used in this study can be referenced in a previous publication by the researchers [5]. In summary, particle analysis was performed utilizing an alpha300 R confocal Raman microscope (WITec, Ulm, Germany). Initially, a light microscopic darkfield image of the filter surface was acquired using a Zeiss EC Epiplan HD 20×/0.5 NA microscope objective. Following image acquisition, WITec ParticleScout (version 5.3.18.110) was used for image processing, whereby bright appearing particles were demarcated against the dark background by automated determination of a brightness threshold executed by the software.

The alpha300 R configuration included a WITec UHTS 300 spectrometer and an Andor iDus Deep Depletion charge-coupled device (CCD) camera. A laser with a wavelength of 532 nm was applied for particle measurements. Raman spectra of the particles were acquired using a Zeiss EC Epiplan-Neofluar DIC 50x/0.8 NA microscope objective with a laser power of 2 mW on the sample. Particle positioning in the laser focus was automated using coordinates derived from the particle masking, and the focus was precisely adjusted to the surface via spectral autofocus. Subsequently, a Raman spectrum was recorded for each particle. The particles were identified by an automated spectral comparison using a self-generated spectral database that was precisely matched to the composition of terrestrial soils.

Environmental fate

UV aging

In the field, mulch films are spread on the fields for several months. The films are then exposed to various abiotic factors such as wind, temperature and UV light, which causes ageing of the films. In order to test if the toxicity of fresh versus aged films differs, the three tested films were artificially aged with a light source in a climate chamber.

The films were placed in glass trays filled with reference soil A01 and irradiated with an artificial light source (burner). A burner simulating the spectrum of the sun was used for the ageing tests. The burner was lent by Larsol GmbH, it is a Yuma HMI1200 DS SOLAR.

The ambient temperature and humidity were kept constant. The films were aged for 4 weeks at constant temperature (42 °C) and humidity (approx. 25% relative humidity) to simulate a 6-month period of use (April–September). The pyranometer and the black standard thermometer were fixed at the height of the film to determine the temperature and radiation intensity at film level and to adjust them if necessary. The black standard temperature was 60 °C.

After ageing, the films were weighed and then stored airtight in amber glass bottles, all films showed a significant reduction in weight. Part of the films were sent to the partner for ecotoxicological studies.

Aging in drainage water

In the iMulch project, no pure plastic degradation tests were deliberately carried out, as there are already many publications on this subject. Also, the project duration would not have been sufficient for degradation of the mulch films, as complete degradation in water can take years. Rather, weathering and alteration of mulch film particles were focused on, as these factors directly affect emission, dispersion, and fate of the particles.

Weathering tests of square and powdered test materials from petroleum and biodegradable mulch films were conducted in the aqueous medium with a laboratory wastewater treatment plant (Fig. 2) in two experiments, each lasting approximately 3 months. Test materials and aqueous medium were examined before and after the

Fig. 2 Laboratory wastewater treatment plant type KLD4N/SR

runtime by digital microscopy, spectrometry and pycnometry, among other methods.

Ageing on soils under laboratory conditions

A soil test stand for weathering mulch films under natural conditions in the laboratory was established (Fig. 3). Preliminary tests were carried out in a prototype (self-built). This has shown that the observation period lasts longer than initially expected due to accelerated climate chamber tests, as the samples were weathered on the ground under almost "normal" ambient conditions in terms of soil moisture and air temperature. Therefore, a duration of at least 16 weeks had to be assumed. To determine the influence of soil type, soil moisture and UV irradiation, appropriate sensors were tested and selected for the construction of the new test stand. The system concept was modified in that a drainage floor was installed to avoid water stagnation. In addition, the number of registers in the new test stand was increased to six. This makes it possible to test different soil and film types at the same time. Here the test were conducted with one soil type and the three mulch films (two biodegradable and one PE Film).

The six registers of the soil test stand were filled with a reference soil Refesol 01-A. RefeSol are soils recognized by the Federal Environment Agency (UBA) for test procedures in accordance with the Federal Soil Protection Act/Ordinance. Refesol 01-A is a Dystric cambisol, the soil is medium acid and very light humic and shows an effective cation exchange capacity of 11.6 mmolc/kg. The individual registers were each fitted with six drippers for irrigation and a capacitive moisture sensor to control the irrigation using a solenoid valve. A UV lamp for irradiation was positioned centrally above every two registers. A lamp system from Vilber (vilber.com) was used that has a similar spectrum to the UV-A lamps from the measuring device manufacturers ATLAS and QLab (pausch.com).

The radiation output is determined in one register using a UV sensor as an example. Two pieces of film of a mulch film type were placed in each of two registers for weathering.

During the test operation, data on humidification and ambient temperature were continuously recorded and photographic documentation of the samples was carried out. The moisture content of the soil was also checked using an external capacitive measuring device and the amount of water was determined manually. During irrigation, it was noticeable that the water remained on the surface of the soil for a long time before it seeped away. Samples were taken before and after the test to evaluate the test regime. Soil samples were taken at a depth of up to 5 cm, up to 10 cm and down to the bottom of the register.

The soil samples were subjected to a simplified leaching test. For this purpose, 100 ml of water was added to each sample in a beaker and stirred at room temperature for approx. 30 to 60 min until a homogeneous dispersion was formed. The filtrate was then filtered, concentrated, and analysed using infrared spectroscopy (IR). No evidence of films could be found in the filtrates.

The mulch films were examined before and after the test using various analysis methods. Fragmentation was investigated using digital microscope images and scanning electron microscopy (SEM) analysis. The change in the polymer was measured using differential scanning calorimetry (DSC) and thermogravimetry (TG) for the change in thermal parameters such as mass loss, melting

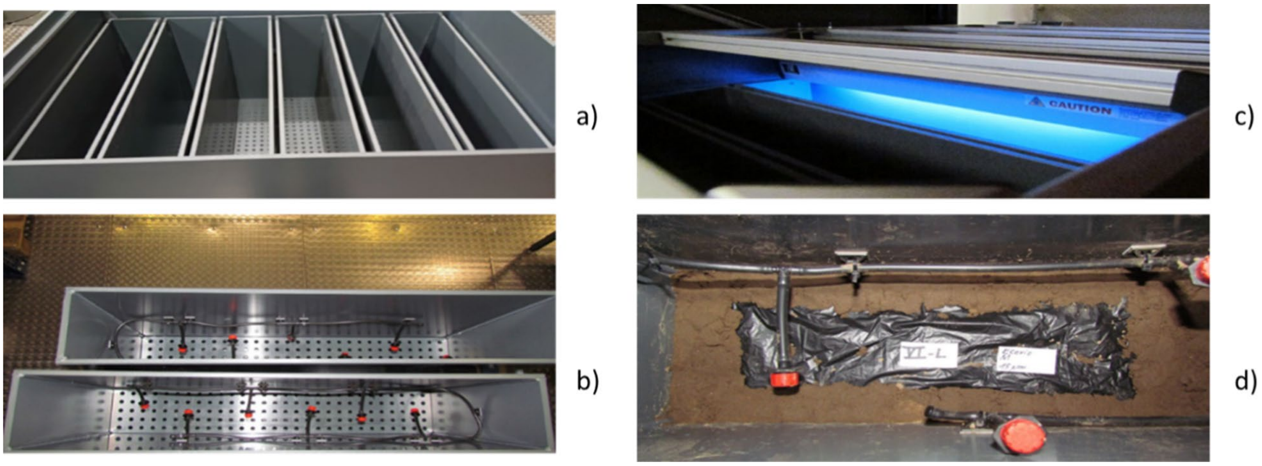


Fig. 3 System components of the soil test stand (**a**) registers which can be filled with different type of soils, **b** Photo of the irrigation system with which the moisture in the soil can be adjusted, **c** UV-lamp system for illumination, **d** photo of the test stand with mulch film

range, crystallization, and glass transition. IR was used to detect the change in molecular parameters.

Lysimeter experiments

For the lysimeter studies the ^{14}C-radioactive labelled polymers were synthesized, ^{14}C-PBAT and ^{14}C-PE were synthesized. The polymers were cryo-milled and the resulting particles were spiked on the top 5 cm of undisturbed outdoor lysimeter. The lysimeter experiments were run for 24 months. Different plants were cultivated on the lysimeters according to normal agricultural practice. The transport of the polymer particles or degradation products in the lysimeters as well as the uptake into the plants were investigated.

Carrier function, adsorption of contaminants on the polymer surface

A frequently discussed problem with the adsorption of substances is that the non-toxic sorbent can serve as a vector for sorbed, toxic substance. On the other hand, it can be of great advantage if the concentration of the toxic substances is reduced by the sorption, provided that desorption does not occur again downstream.

Therefore, it is important to better understand both the adsorption and desorption processes of the films.

Sorption studies are mainly conducted in batch experiments [11]. The batch approach is widely used because of its simplicity [12]. It requires that the sorbate (target compound) and the sorbent are mixed in a liquid matrix [12]. Once sorption equilibrium is reached, the equilibrium concentration of the remaining sorbate in the liquid phase can be determined using various detection methods that depend on the sorbate, such as UV–vis spectrometry or mass spectrometry (MS)-based methods. The sorbate concentration in or on the solid phase is determined by a mass balance calculation using the initial spiked mass and the equilibrium concentration in the liquid phase. For the batch method, an established method based on OECD TG106 [13] was adapted for the adsorption experiments in this project.

The adsorption behaviour of the three films [conventional PE film and two biodegradable (PLA/PBAT) films (biodegradable 1 and biodegradable 2)] was tested according to the procedure shown in Fig. 4. The pesticides atrazine, tebuconazole and thiacloprid and the heavy metal copper were selected as adsorbates. For the tests, a constant mass (5 mg) of the films was mixed with a certain concentration of the substance under investigation and shaken with water at a constant temperature (21 °C) for a certain time (24 h, 48 h, 1 week, 2 weeks). After the selected time points, the liquid was removed with a pipette and then analysed for the pesticide concentration using liquid chromatography–tandem mass spectrometry (LC–MS/MS) or for the copper concentration using inductively coupled plasma mass spectrometry (ICP-MS). To prevent the films from floating, they were weighted down with glass beads.

The measured concentration in the liquid provides information about the amount of the substance adsorbed on the substrate surfaces.

Ecotoxicity

Potential ecotoxicological effects of mulch films were investigated in a bidirectional approach, considering exposition in water and soil, whereby the focus was set on terrestrial exposure scenarios.

In the aquatic approach, films were eluted in water over 1 d and 30 d to simulate leaching and thus exposure in aquatic system. They were tested in an aquatic biotest battery including the micronucleus assay [14], the ER-CALUX assay [15], the micro-EROD assay [16] and previous cytotoxicity tests in order to estimate exposure concentrations for the mechanism-specific assays without acute toxicity.

For terrestrial investigations, films were cryoground to microplastic particles of 50–1000 µm size range and evenly distributed in soil to assess effects of possibly remaining microplastic particles in soil on native species. Reproduction assays were conducted with the earthworm *Eisenia fetida* and the springtail *Folsomia candida* in different concentrations up to 5 and 10 mg/g soil dry weight, respectively, according to the OECD guidelines 222 [17] and 232 [18]. Survival, weight change, reproductive

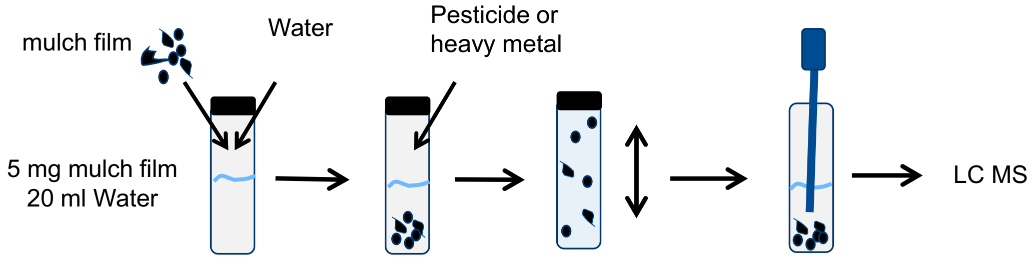


Fig. 4 Simplified scheme of the adsorption tests

output, and hatching delay were investigated for the earthworm and reproductive output for springtails.

Further, biomarker measurements of catalase (CAT), Glutathione-*S*-transferase (GST) and acetylcholine esterase (AchE) activities and oxidative stress were conducted for earthworms exposed for 4 weeks and also after a subsequent recovery time in uncontaminated soil for another 4 weeks according to the methods detailed in Lackmann et al. [19].

Additionally, polyethylene film particles were mixed with established pesticides based on active ingredients tebuconazole and thiacloprid and tested in acute mortality tests with *Eisenia fetida* to observe the particle's impact on pesticide exposure and availability to organisms. Investigated concentrations ranged from 50 to 300 mg/kg$_{dw}$ (tebuconazole) and 0.5 to 500 mg/kg$_{dw}$ (thiacloprid).

Life cycle analysis (LCA)

A life cycle analysis (LCA) according to ISO 14040 [20] and ISO 14044 [21] was carried out to assess the environmental impact of a biodegradable mulch film in comparison to a conventional non-biodegradable mulch film. Potential environmental hotspots were identified within the life cycle. The results of this LCA have not been critically reviewed.

As a functional unit, 1 ha of mulch film for the cultivation of courgettes was selected for both types of mulch film. A conventional one based on polyethylene (PE) with a layer thickness of 30 µm and a biodegradable one based on bio-based polylactide (PLA) and fossil-based polybutylene adipate terephthalate (PBAT) with a layer thickness of 10 µm were compared.

A cradle-to-grave study was conducted and covered material production, transport between processing plants, compounding, extrusion of the film and various end-of-life (EoL) treatment options. At EoL, the biodegradable film remains in the field and biodegrades there whereas for the conventional film, two types of EoL were considered, namely mechanical recycling and incineration with energy recovery. The PE are collected, cleaned and dried before being incinerated or recycled. This study assessed a scenario where 80% of the PE film are incinerated and 20% are mechanically recycled.

The value chain definition and the inventory data are based on LCA databases (Ecoinvent 3.6), literature research [22–25] and communication with different experts on biodegradable compounds, mulch films and recycling. Therefore, this study compares two generic mulch films and is not specifically representative of commercially available mulch films.

For the LCA, the EF 3.0 (Environmental Footprint 3.0) was used as the impact category set [26]. In this publication, only the results for Global Warming Potential (GWP100) are reported [27].

Biogenic carbon uptake from PLA for the biodegradable mulch films, as well as credits from energy and material recovery at the end of life of the PE mulch films are considered and showed separately. The biogenic carbon embedded in PLA is released as biogenic carbon emissions to the atmosphere during biodegradation.

Can the plastic be kept in the loop? Upcycling as potentially sustainable end-of-life option

The use of low value mixed plastic fractions or contaminated plastics is a challenge in countries, in which waste logistics is implemented at a quality that avoids to the large extend littering in to the environment. Besides incineration, a clearly beneficial technology compared to landfilling, few ideas exist on how one could use these mixed plastic fractions. Depending on the quality of the plastics, pyrolysis as discussed for decades and now brought to industrial scale. Alternatively, catalytic degradation of plastic to the monomers followed by subsequent use of these chemical mixtures as carbon source for microbes is discussed by [28]. Examples include the enzymatic degradation of PET and subsequent utilization of ethylene glycol and terephthalic acid by engineered *Pseudomonas* species to rhamnolipids [29] or the pyrolysis of PE to alkanes, followed by chemical oxygenation. The latter product was a superior substrate for PHA synthesis by Pseudomonas [30]. More complex is the utilization of PU monomers, as showcased by Utomo et al. In iMulch, we hence investigated if upcycling rather than leaving the plastic in the soil, could contribute to an improved material use. As an alternative end-of-life scenario, we discussed, if the addition of microbes that can utilize the plastic monomers and may even have a hydrolytic activity on the mulch film, as discussed elsewhere [31], would help to reduce the time of (micro)plastic in the field. The first step was to identify microorganisms, which are able to metabolize the PLA/PBAT mulch film monomers lactic acid, adipic acid and 1,4-butanediol, terephthalic acid. This was done by isolating microorganisms from several field soil samples as well as by a literature study on potential promising organism groups and respective screening of the institute's strain collection. The entirety of bacteria in the field soil samples was analysed by 16S rRNA Illumina amplicon sequencing. Microbial phenotypes on the monomers in pure culture was confirmed by simultaneously measuring microbial growth photometrically and the dissipation of mulch film monomers from the cultivation supernatant via high-performance liquid chromatography. Further experiments aimed at the microbial conversion of mulch film monomers into molecules of value. Finally, a microbe was identified that not only could natively use all four PLA/PBAT monomers, but also showed low hydrolytic activity on the mulch film directly (analysis via CO_2-emission, SEM, EDX and enzymatic assays). The conditions of the microbial cultivation such as preculture, temperature, pH, and oxygen supply were optimized.

Results

Detection method

In a first step, a detection methodology was developed, which included the establishment and validation of a sample preparation, analysis and evaluation method for thermo-extraction desorption (TED)-GC–MS on the one hand and a suitable sample preparation and analysis method for Raman microspectroscopy on the other.

The Raman microspectroscopic method was successfully used for particle characterization of soil samples. The workflow could be (partially) automated, so that in addition to the particle identification, statements could be made about the material and particle size distribution as well as the particle shape. Small amounts of microplastics of different types could be detected in all soil samples investigated. However, the (time) limiting factor in the use of this measurement technique is still the high matrix load of the soil samples. In the course of the project, the matrix separation could be improved significantly, but the high heterogeneity of terrestrial soil matrix is still a big problem. Currently, 200 particles need to be spectroscopically measured in order to find one plastic particle. For more details, please refer to [5].

Concentration in field samples

With the established TED-GC–MS method, different soil samples, for which the type of cultivation was known, were analysed. The hypothesis was that the different forms of cultivation, cultivation with/without mulch film, with biodegradable mulch film and conventional PE film would lead to different concentrations of PE or PLA/PBAT in the soil. However, this could not be confirmed by the measurements.

It was also shown that the two sampling campaigns show different results, indicating an inhomogeneous distribution of the microplastic on the field (Table 1).

Table 1 Concentration of PE and PLA/PBAT from the different fields measured with TED-GC–MS

Field	µg PE/g soil	µg PLA/PBAT/g soil
Field with biodegradable film_A	< 1	0.3
Field with biodegradable film_A2	4.4	< 0.1
Field with biodegradable film_B	5.2	0.5
Field without film_A	< 1	< 0.2
Field without film_A2	1.4	< 0.3
Field without film_B	< 4	0.5
Field with mulchfilm/strawberries_A	< 1	< 0.1
Field with mulchfilm/strawberries_B	< 1	2.2
Field close to motorway service station_A	< 1	< 0.1
Field with asparagus film_A	< 1	0.2
Field with asparagus film_B	< 1	0.4
Reference soil RefesolA01	8.2	0.4

All soils are Luvisol with a pH between 6.4 and 6.7. The last field shows a pH of 7.5.A = sampling in March 2021, B = sampling June 2021, A2 samples were shipped in plastic foil made of PE

Environmental fate studies

Ageing in drainage water

Ageing tests with conventional (PE) and biodegradable mulch films were carried out in a laboratory-scale test sewage treatment plant, which was filled with drainage water from an arable field. The PLA/PBAT mulch films showed clear deterioration after only a few days. At the end of the tests after 8 weeks cracks and holes were evident, but no complete degradation. However, fouling and weathering occurred on all mulch films (Fig. 5). Both film types showed a significant increase in density over time due to fouling, which causes film fragments to sediment in water bodies. Additionally the FTIR spectra changed over time, especially for the *biodegradable* film fragments, due to fouling, which can make it difficult to clearly identify the polymers. Using 10% HCl, this process is reversible, so HCl treatment is recommended prior to spectrometric studies.

Aging in soil test stand

The weathering of mulch films was additionally investigated in a soil test stand, here more naturally like conditions could be tested compared to a climate chamber test system. After about six to eight weeks, the first changes in the bio degradable films were visible (cracks, holes). By the end of the test, no complete degradation of the biodegradable films could be observed and the PE Film shows no degradation at all. The following Fig. 6 shows the results of the photo documentation.

This observation was confirmed in the SEM images (not shown). Here fragmentation was observed with cracks and holes for the biodegradable films and no degradation for the PE film.

However, no significant change over time was observed in the IR spectra and in the thermal analysis, also indicating a low degradation of the material at all.

The degree of soil mass on the biodegradable film was greater than in the PE film. This “contamination” influences the IR spectroscopy as bands are covered by the

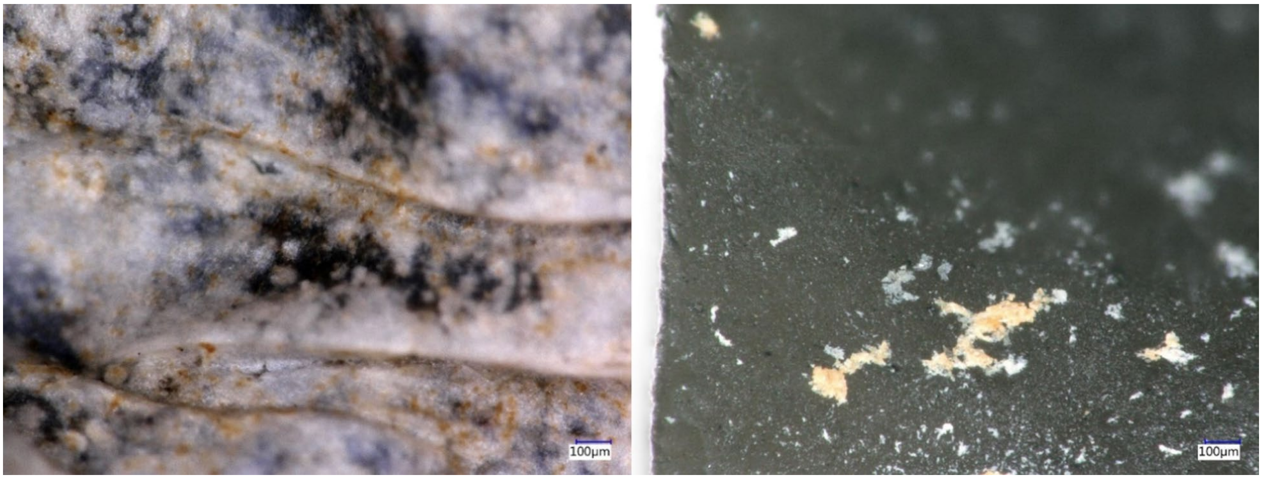


Fig. 5 Fouling and weathering of biodegradable (l.) and PE (r.) mulch film fragments

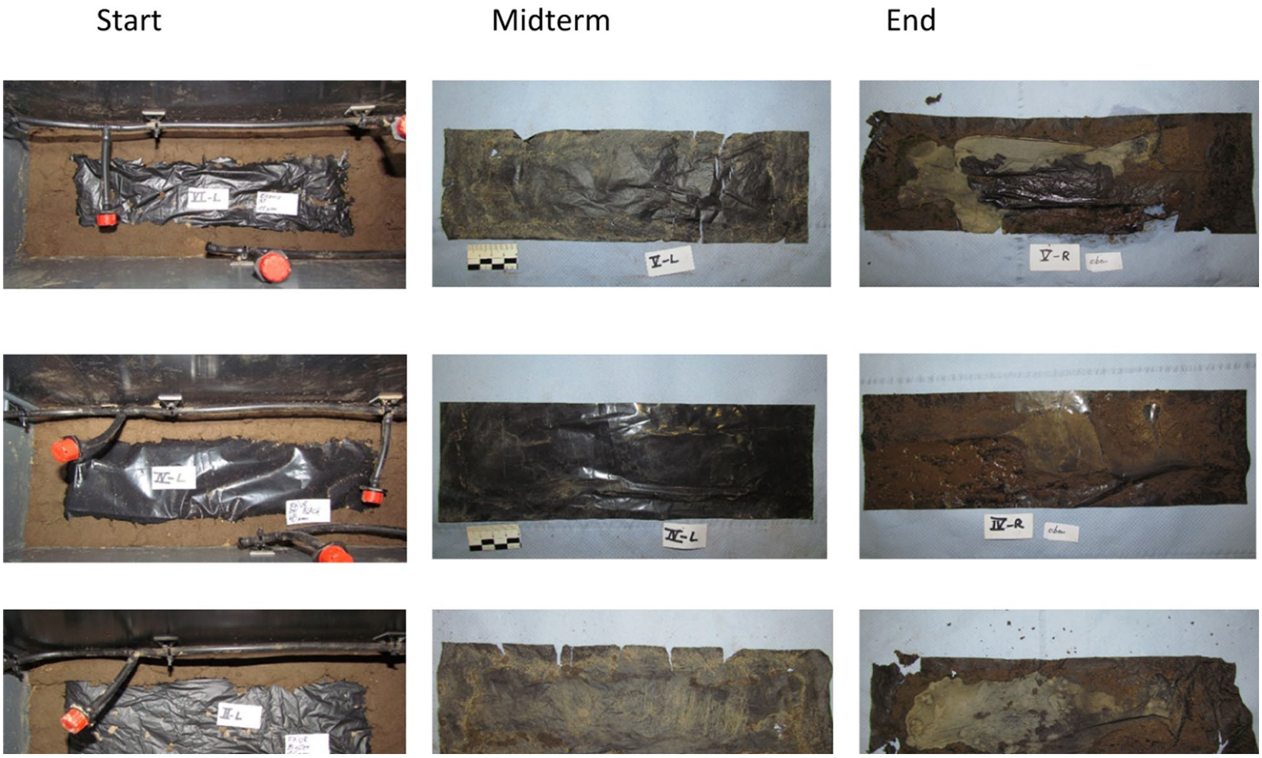


Fig. 6 Photo-documentation (from top to bottom biodegradable 1-15 µm, PE black—40 µm, biodegradable 2-16 µm)

soil material and can therefore not be evaluated. In the DSC curves, small changes can be seen in the crystallization and melting areas of the biodegradable films whereas no differences can be seen in the PE film.

Lysimeter experiments

In order to analyse the transport and degradation behaviour of the polymers in the soil, mulch film particles with ^{14}C-radioactive labelling were synthesized, placed in topsoil and investigated in a lysimeter study for 24 month. Under realistic field conditions, no transport, no significant degradation and no uptake of the mulch film particles or their degradation products in plants could be detected.

Carrier function, adsorption of contaminants on the polymer surface

The results of the adsorption tests indicate that no detectable adsorption of the heavy metal copper was observed (data not shown). Additionally, among the three pesticides tested on the films (see Fig. 7) using the selected method, only the pesticide tebuconazole showed a slight tendency toward low adsorption on the biodegradable film. Given that negligible or minimal adsorption was detected for all the substances tested, no desorption tests were performed. Consequently, it can be concluded that the film fragments do not serve as carriers for the pesticides.

Ecotoxicity studies

The ecotoxicological effects of the mulch films on aquatic and terrestrial organisms were studied. In the terrestrial toxicity tests, no negative effects on soil organisms could be detected in the acute reproduction tests with the earthworm *Eisenia fetida* and the springtail *Folsomia candida*. Some biomarker analyses indicated slight concentration-dependent increase in earthworms. Further, the presence of microplastic seemed to lower the availability and, thus, toxicity of pesticides in acute testing.

In the investigation of effects on aquatic organisms, similar estrogenic effects in range of 8–10 17ß-Estradiol eq./mg mulch film in the ER CALUX assay could be detected for both film types (PE and PLA/PBAT blend) in different elution time frames, with the aged films showing a lower effect. No differences in effect strength between the film types could be shown. Detailed results will be given elsewhere [32–34].

Life cycle analysis (LCA)

As seen in Fig. 8, raw material sourcing has a significant impact on the cradle-to-gate results for both film types. For biodegradable films, the high impact of fossil-based PBAT production is mainly due to the high carbon footprint of adipic acid, linked to the release of N_2O during its synthesis. For PE films, PE production shows a high contribution to climate change. The energy contribution from the processing steps (compounding and extrusion) is also relevant, especially for PE films, as more material has to be processed to produce 1 ha of mulch film.

At the end of the life cycle of biodegradable mulch film, the film remains on the ground. The emissions of CO_2 released during biodegradation enter the atmosphere and contribute to climate change. The disposal of PE films is more complex requires several steps: they have to be collected, cleaned and dried, and then recycled or incinerated. Therefore, the end-of-life impact of PE films on climate change is higher than the end-of-life impact of biodegradable mulch films.

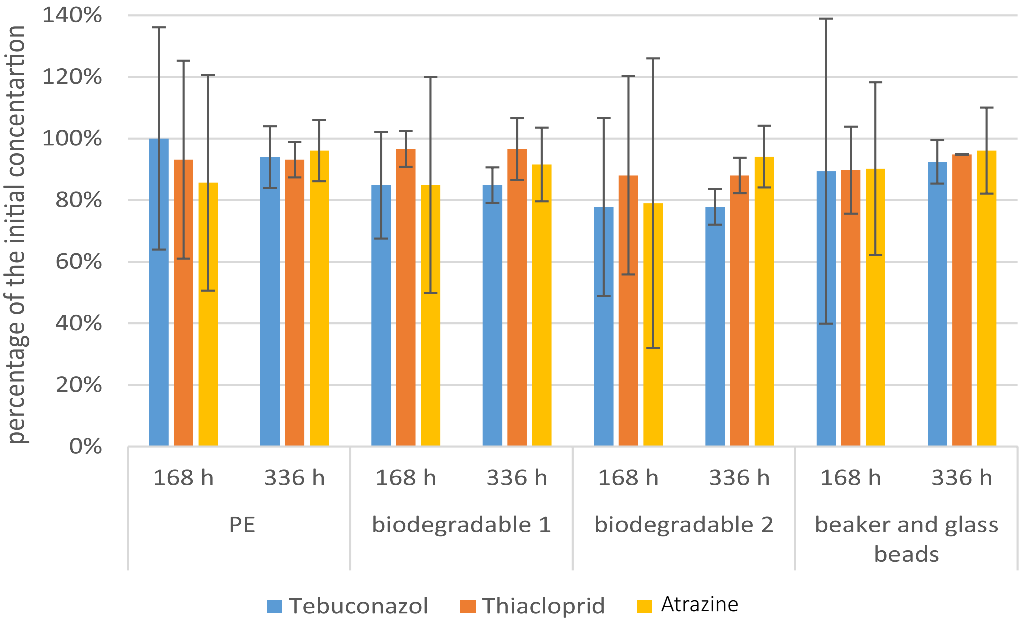


Fig. 7 Results of the adsorption experiments after 168 h and 336 h for the pesticides tebuconazole (blue), thiacloprid (orange) and atrazine (yellow). Concentration: 400 ng/ml. Shown is the percentage of the initial concentration in the liquid; *n* = 3

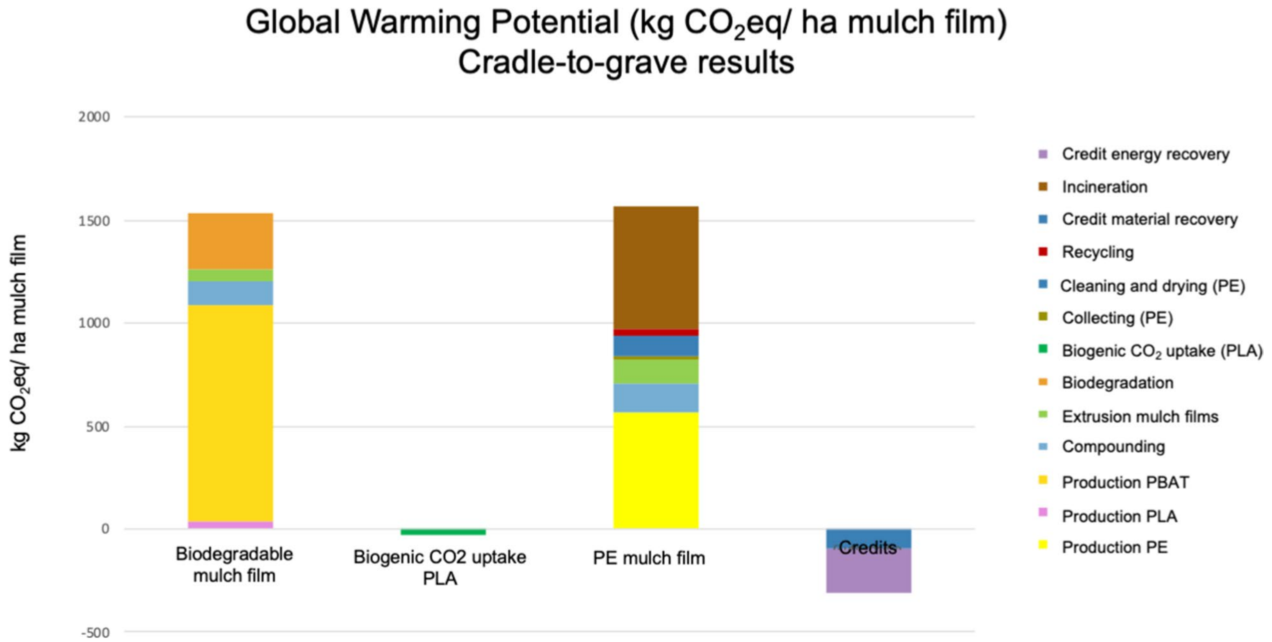


Fig. 8 Life cycle analysis (cradle-to-grave) for both mulch film types, PE mulch film and biodegradable mulch film. Shown is the global warming potential (kg CO_2eq/ha mulch film)

Additionally, the contribution of end-of-life processes (recycling and incineration) to the final LCA results strongly depends on the allocation methodology applied. For climate change impacts, PE mulch films perform better than biodegradable mulch films when energy recovery and material recycling credits are taken into account. If only the EoL burdens are taken into account, the two mulch films perform equally well.

Finally, potential for greenhouse gas emissions savings have been evaluated in the full LCA study, particularly with the use of bio-based adipic acid for the synthesis of PBAT and an increase in the recycled content of the PE film.

Upcycling as potentially sustainable end-of-life option

Another focus of the work was the biotechnological upcycling of the biodegradable PLA/PBAT- mulch films. Within the framework of the project, microorganisms able to utilize all of the monomers lactic acid, adipic acid, 1,4-butanediol and terephthalic acid and of the biodegradable mulch film were identified and a conversion to other molecules of value was demonstrated. Further, hydrolytic activity of the microbe directly on the mulch film could be shown. On the other hand, the conventional film consisting of PE, could not be degraded or transformed by the microorganisms. Detailed results are yet in preparation to be published [35, 36].

Discussion

A strength of this project is that not only an analysis method was developed, but also that various aspects of the life cycle and their impact were considered.

A sample preparation method for TED-GC–MS and Raman microspectroscopy was successfully established and various real soil samples were analysed. It was investigated whether a correlation with the form of cultivation could be detected. While it was possible to analyse six fields including the reference soil with the TED-GC–MS, only one field could additionally be analysed with Raman microspectroscopy. In general, the results show a wide variation, between the two sampling campaigns and the different soils from < LOD up to 8.5 mg/kg, which indicate a non-homogeneous distribution of the particles. The highest concentration is comparable to dimensions of concentrations found for other polymers by Müller et al. (2022) and Steinmetz et al. [37]. In a Study conducted by Steinmetz et al. [37] eight soils were studied for their plastic content using pyrolysis GC–MS. The highest concentrations for PE up to 35 mg/kg was found for a soil, which used 40 µm perforated PE film. In two other soils using this type of foil concentration from 4–7 mg/kg were detected. For fields using 50 µm foil no concentration about the limit of detection (1.5–2 mg/kg) could be detected.

On the fields tested in this project mulch film with a thickness of 50 µm was used, which may explain the results that no PE could be found in the field which was covered with mulch film.

Furthermore, the results of the TED-GC–MS analysis did not show any correlation with the type of cultivation and the concentration of PE or PLA/PBAT in the soil. For example, the highest concentrations of PE were found in soils that were cultivated with biodegradable mulch film. Although the origin of the PE cannot be clarified, it is conceivable that the PE was introduced by littering or atmospheric deposition.

Furthermore, it has to be noted, that the selected areas represent ideally cultivated areas, as these were very small and thus the farmers are more likely to notice when films remain on the field, whereupon they can then be collected by hand. These fields may present a best-case scenario. The sampling was carried out in accordance with the soil science mapping guidelines for nutrients as described above.

Repeat sampling of the same areas revealed significant discrepancies from the initial results, highlighting a clear need for improvement in the sampling process. At the time the iMulch project was conducted, there was limited guidance available on suitable sampling methods for microplastics, and current literature still lacks definitive recommendations for appropriate sampling techniques. However, the project results demonstrate the importance of sampling in microplastic analysis and indicate a pressing need for further research in this area.

Exemplary Raman images demonstrated the presence of microplastic particles in the soil samples. For the Raman spectroscopy beside the spiked soil samples for the method validation one real undoped soil sample was analysed. From this sample, 3906 particles were examined, revealing only 12 polymer particles: 1 PET, 3 PS, and 8 PTFE particles. Neither PE nor PBAT was detected. Given that the mulch films used were black colour, it is possible that these particles were overlooked during Raman microspectroscopy analysis.

Other studies have reported higher quantities of particles, particularly in regions of China. For example, Liu et al. found 62.5 particles/kg in deep farmland soils, 78 particles in shallow soils, and 1.9 particles in garden soils. Thinner mulch films are frequently employed in non-EU countries, with typical thicknesses in China ranging from 6 to 10 µm, while EU regulations require agricultural covers to be over 20 µm thickness [37, 38]. Thinner films fragment more quickly, due to this a greater number of meso- and microplastic fragments remaining in the soil. This discrepancy in mulch film thickness may help explain why studies in China report higher plastic concentrations in soils compared to western and northern Europe [39, 40].

The results of the aging studies have shown that the PE films show almost no aging, which also means that there is only fragmentation in the environment but the films cannot be degraded. Lysimeter experiments further suggested that PE fragments do not migrate through the soil and are unlikely to reach groundwater. Instead, they tend to accumulate in the upper soil layers, available for soil organisms living in that area. Since the particles do not degrade but continue to accumulate, an increasing exposure concentration is expected in future.

However, the aging studies have shown that under more natural conditions, even the films that are considered biodegradable require a significantly longer time to degrade and the results cannot be used to determine how long this period is. The PBAT particles were neither transported in the lysimeter tests nor taken up by the plants grown on the lysimeter. Furthermore they show no degradation under lysimeter conditions. The results for the upcycling experiments have shown that under ideal laboratory conditions, the PLA/PBAT films can be degraded by microorganisms. How long this process takes under natural conditions requires further research. But it remains to be noted that there is at least a chance that these particles can be degraded at some point in contrast to PE films.

In the ecotoxicological experiments the two film types (PE and *biodegradable* alternative) show more or less the same behaviour and exhibit no effects in the acute reproduction tests for the soil organisms. The results for the cell biotests indicate an endocrine effect for both film types, whereas aged films indicate a lower effect. However, more research and long-term studies are needed to enable a comprehensive assessment for soil and aquatic organisms, especially considering that an enrichment of the microplastic concentration in the upper soil layers is to be expected. The concentrations in which mulch films in particular are present in the environment or in wastewater ditches are not sufficiently documented at this time.

Summary and conclusion

In the framework of the project it could be shown that the PE could neither be degraded nor transported, even under ideal conditions no degradation could be detected.

However, the biodegradable films showed a similar pattern. The films could only be degraded under ideal conditions in the laboratory. In the life cycle assessment, PE mulch films perform better than biodegradable mulch films in terms of their impact on climate change when the credits for energy recovery and material recycling are taken into account.

In the ecotoxicological investigations, both types of film showed similar effects, whereby the aged films showed fewer effects than the non-aged films, which could indicate a possible involvement of additives used in both types of the films.

Results from the project suggest that degradation of biodegradable films only occurs in the laboratory under optimal conditions, while no degradation could be detected under the experimental conditions in the field. To reduce the input of plastic film fragments into the environment and thus make the use of mulch films lower in emissions following suggestions were derived.

PE films

- A minimum lower limit of material thickness should be defined in order to minimize possible losses when collecting the film.

Biodegradable films

- Increase the film thickness of biodegradable films.
- Collection of the film from the field after use. If individual fragments remain on the soil, there is at least a chance that they can be degraded after some time.

Proof of degradability under different realistic field conditions (lower temperature and humidity) long-term studies are lacking for a final assessment of the fate and impact of PE film or PLA/PBAT films.

In contrast to mulch films, the use of sewage sludge or compost, which can be contaminated with plastics, seems to lead to significantly higher concentrations in the soil.

Abbreviations

DSC	Differential scanning calorimetry
ICP-MS	Inductively coupled plasma mass spectrometry
LCA	Life cycle assessment
LC–MS	Liquid chromatography mass spectrometry
LC–MS/MS	Liquid chromatography tandem mass spectrometry
LOQ	Limit of quantification
NaCl	Sodium chloride
NaI	Sodium iodide
PHA	Polyhydroxyalkanoates
PE	Polyethylene
PET	Polyethylene terephthalate
PTFE	Polytetrafluoroethylene
PLA	Polylactide
PBAT	Polybutylene adipate terephthalate
PU	Polyurethane
PS	Polystyrol
SEM	Scanning electron microscopy
TED-GC–MS	Thermo extraction desorption gas chromatography mass spectrometry
TG	Thermogravimetry

Acknowledgements
The iMulch project was funded by the European Regional Development Fund (ERDF) "Investment for Growth and Employment". ERDF-0801177. We would like to thank the associated partners FKUR and BASF for providing the mulch films. We would like to thank the Landwirtschaftskammer NRW for its support with soil sampling.

Author contributions
Carmen Wolf, Antonia Weltmeyer and Kristina Bitter conceptualized and drafted the manuscript. All other authors helped to further elaborate the manuscript and contributed specific aspects. All authors read and approved the final manuscript.

Funding
Open Access funding enabled and organized by Projekt DEAL. The iMulch project was funded by the European Regional Development Fund (ERDF) "Investment for Growth and Employment". ERDF-0801177.

Data availability
No datasets were generated or analysed during the current study.

Declarations

Ethics approval and consent to participate
Not applicable.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

Author details
[1]Institut Für Umwelt & Energie, Technik & Analytik e.V. (IUTA), Duisburg, Germany. [2]FISCHER GmbH, Meerbusch, Germany. [3]Fraunhofer UMSICHT, Oberhausen, Germany. [4]Fraunhofer Institute for Molecular Biology and Applied Ecology IME, Schmallenberg, Germany. [5]RWTH Aachen University, Institute for Environmental Research IFER, Aachen, Germany. [6]RWTH Aachen University, Institute of Applied Microbiology iAMB, Aachen, Germany. [7]Goethe Universität, Department Evolutionary Ecology & Environmental Toxicology E3T, Frankfurt, Germany. [8]Nova-Institut GmbH, Huerth, Germany.

Received: 18 May 2024 Accepted: 30 December 2024

References

1. PlasticEurope (2022) Plastic-the facts 2022—an analysis of European plastics production, demand and waste data. Bruessels.PlasticEurope. https://plasticseurope.org/knowledge-hub/plastics-the-facts-2022/
2. Hofmann T, Ghoshal S, Tufenkji N et al (2023) 2023 Plastics can be used more sustainably in agriculture. Commun Earth Environ 4:332. https://doi.org/10.1038/s43247-023-00982-4
3. Le Moine B, Ferry X (2019) Plasticulture: economy of resources. Acta Hortic 1252:121–130. https://doi.org/10.17660/ActaHortic.2019.1252.17
4. Steinmetz Z, Wollmann C, Schaefer M, Buchmann C, David J, Tröger K, Munoz K, Frör O, Schaumann GE (2016) Plastic mulching in agriculture. Trading short-term agronomic benefits for long-term soil degradation? Sci Total Environ 550:690–705
5. Wenzel M, Fischer B, Renner G, Schoettl J, Wolf C, Schram J, Schmidt TC, Tuerk J (2022) Efficient and sustainable microplastics analysis for environmental samples using flotation for sample pre-treatment. Green Anal Chem 3:100044. https://doi.org/10.1016/j.greeac.2022.100044
6. Funck M, Al-Azzawi MSS, Yildirim A, Knoop O, Schmidt TC, Drewes JE, Tuerk J (2021) Release of microplastic particles to the aquatic environment via wastewater treatment plants: the impact of sand filters as tertiary treatment. Chem Eng J. https://doi.org/10.1016/j.cej.2021.130933
7. Al-Azzawi MSM, Kefer S, Weißer J, Reichel J, Schwaller C, Glas K, Knoop O, Drewes JE (2020) Validation of sample preparation methods for microplastic analysis in wastewater matrices—reproducibility and standardization. Water 12:2445. https://doi.org/10.3390/w12092445
8. Nwaila GT, Frimmel HE, Zhang SE, Bourdeau JE, Tolmay LCK, Durrheim RJ, Ghorbani Y (2022) The minerals industry in the era of digital transition: an energy-efficient and environmentally conscious approach. Resour Policy 78:102851. https://doi.org/10.1016/j.resourpol.2022.102851

9. Song S, Zhang X, Yang B, Lopez-Mendoza A (2012) Flotation of molybdenite fines as hydrophobic agglomerates. Sep Purif Technol 98:451–455. https://doi.org/10.1016/j.seppur.2012.06.016
10. Renner G, Nellessen A, Schwiers A, Wenzel M, Schmidt TC, Schram J (2020) Hydrophobicity–water/air–based enrichment cell for microplastics analysis within environmental samples: a proof of concept. MethodsX 7:100732. https://doi.org/10.1016/j.mex.2019.11.006
11. Kah M, Zhang X, Jonker MTO, Hofmann T (2011) Measuring and modeling adsorption of PAHs to carbon nanotubes over a six order of magnitude wide concentration range. Environ Sci Technol 45(14):6011–6017. https://doi.org/10.1021/es2007726
12. Shahryari Z, Goharrizi AS, Azadi M (2010) Experimental study of methylene blue adsorption from aqueous solutions onto carbon nano tubes. Int J Water Resour Enviro Eng 2:16–28. https://doi.org/10.5897/IJWREE.9000021
13. OECD (2000) Test No. 106: adsorption—desorption using a batch equilibrium method. OECD guidelines for the testing of chemicals, Section 1. OECD Publishing, Paris. https://doi.org/10.1787/9789264069602-en
14. Reifferscheid G, Ziemann C, Fieblinger D, Dill F, Gminski R, Grummt HJ, Hafner C, Hollert H, Kunz S, Rodrigo G, Stopper H, Selke D (2008) Measurement of genotoxicity in wastewater samples with the in vitro micronucleus test—results of a round-robin study in the context of standardisation according to ISO. Mutat Res Genet Toxicol Environ Mutagen 649:15–27. https://doi.org/10.1016/j.mrgentox.2007.07.015
15. Kunz PY, Simon E, Creusot N, Jayasinghe BS, Kienle C, Maletz S, Schifferli A, Schonlau C, Ait-Aissa S, Denslow ND, Hollert H, Werner I, Vermeirssen ELM (2017) Effect-based tools for monitoring estrogenic mixtures: Evaluation of five in vitro bioassays. Water Res 110:378–388. https://doi.org/10.1016/j.watres.2016.10.062
16. Schiwy A, Brinkmann M, Thiem I, Guder G, Winkens K, Eichbaum K, Nusser L, Thalmann B, Buchinger S, Reifferscheid G, Seiler TB, Thoms B, Hollert H (2015) Determination of the CYP1A-inducing potential of single substances, mixtures and extracts of samples in the micro-EROD assay with H4IIE cells. Nat Protoc 10:1728–1741. https://doi.org/10.1038/nprot.2015.108
17. OECD (2016) Test No. 222: Earthworm reproduction test (*Eisenia fetida*/*Eisenia andrei*), OECD guidelines for the testing of chemicals, Section 2. Paris, OECD Publishing
18. OECD (2016) Test No. 232: collembolan reproduction test in soil, OECD guidelines for the testing of chemicals, section 2. Paris, OECD Publishing
19. Lackmann C, Velki M, Bjedov D, Ečimović S, Seiler T-B, Hollert H (2021) Commercial preparations of pesticides exert higher toxicity and cause changes at subcellular level in earthworm Eisenia andrei. Environ Sci Eur 33:12. https://doi.org/10.1186/s12302-021-00455-5
20. ISO (2006a) Environmental management—Life cycle assessment—principles and framework (ISO 14040:2006 Amd 1:2020). English version EN ISO 14040:2006 A1:2020
21. ISO (2006b) Environmental management—Life cycle assessment—requirements and guidelines (ISO 14044:2006 Amd 1:2020); German and English version EN ISO 14044:2006 A2:2020
22. Bauer (2021) Personal communication. 2021–02–24.
23. DIN EN 17033:2018-03 (2018) Deutsches Institut für Normung, Europäische Norm) Kunststoffe—Biologisch abbaubare Mulchfolien für den Einsatz in Landwirtschaft und Gartenbau—Anforderungen und Prüfverfahren; Deutsche Fassung EN 17033:2018. DIN EN (Ed.). https://www.din.de/de/mitwirken/normenausschuesse/fnk/veroeffentlichungen/wdc-beuth:din21:275787663
24. Nessi S, Sinkko T, Bulgheroni C, Garcia-Gutierrez P, Giuntoli J, Konti A, Sanye-Mengual E, Tonini D, Pant R, Marelli L, Ardente F (2021) Life Cycle Assessment (LCA) of alternative feedstocks for plastics production - Part 1: the Plastics LCA method, EUR 30725 EN, Publications Office of the European Union, Luxembourg, 2021, ISBN 978-92-76-38145-7, https://doi.org/10.2760/271095, JRC125046
25. Previero (2021) Personal e-Mail communication.
26. European Commission (2019) Environmental Footprint version 3.0. Impact assessment method of the European Environmental Footprint initiative.
27. IPCC (2013) Intergovernmental panel on climate change. fifth assessment report: climate change 2013. http://www.ipcc.ch/ipccreports/assessments-reports.htm.
28. Wierckx N, Prieto MA, Pomposiello P, de Lorenzo V, O'Connor K, Blank LM (2015) Plastic waste as a novel substrate for industrial biotechnology. Microb Biotechnol 8:900–903. https://doi.org/10.1111/1751-7915.12312
29. Tiso T, Narancic T, Wei R, Pollet E, Beagan N, Schröder K, Honak A, Jiang M, Kenny ST, Wierckx N, Perrin R, Avérous L, Zimmermann W, O'Connor K, Blank LM (2021) Towards bio-upcycling of polyethylene terephthalate. Metab Eng 66:167–178. https://doi.org/10.1016/j.ymben.2021.03.011
30. Guzik MW, Nitkiewicz T, Wojnarowska M, Sołtysik M, Kenny ST, Babu RP, Best M, O'Connor KE (2021) Robust process for high yield conversion of non-degradable polyethylene to a biodegradable plastic using a chemo-biotechnological approach. Waste Manag 135:60–69. https://doi.org/10.1016/j.wasman.2021.08.030.Epub2021Aug31
31. Utomo NRC, Li WL, Tiso T, Eberlein C, Doeker M, Heipieper HJ, Jupke A, Wierckx N, Blank LM (2020) Defined microbial mixed culture for utilization of polyurethane monomers. Sustain Chem Eng 8(47):17466–17474. https://doi.org/10.1021/acssuschemeng.0c06019
32. Weltmeyer et al (2024) Working title: effects of virgin and altered mulch film from biobased and petrol-based origin [manuscript in preparation]. Institute for Environmental Research, RWTH Aachen University
33. Weltmeyer A, Roß-Nicoll M (2024) Different mulch films, consistent results: soil fauna responses to microplastic. Environ Monit Assess 196:943. https://doi.org/10.1007/s10661-024-13096-x
34. Weltmeyer et al (2024) Working title: Interaction of microplastic with commonly applied pesticides in earthworm and bacteria [manuscript in preparation]. Institute for Environmental Research, RWTH Aachen University
35. Bitter K, Granderath-Miegel S, Utomo NRC, Pal U, van Haugwitz G, Wei R, Bornscheuer U, Blank LM (2024) Working title: PBAT-PLA mulch film upcycling: BIOTECHNOLOGICAL conversion with *Rhodococcus opacus* DSM43250 [manuscript in preparation]. Institute of Applied Microbiology, RWTH Aachen University
36. Bitter K, Granderath-Miegel S, Utomo NRC, Blank LM (2024) Working title: bioremediation of PBAT-PLA mulch film with *Rhodococcus opacus* DSM43250 [manuscript in preparation]. Institute of Applied Microbiology, RWTH Aachen
37. Steinmetz Z, Löffler P, Eichhöfer S, David J, Muñoz K, Schaumann GE (2022) Are agricultural plastic covers a source of plastic debris in soil? A first screening study, SOIL 8:31–47. https://doi.org/10.5194/soil-8-31-2022
38. EN 13655 (2018).EN 13655 Plastics - thermoplastic mulch films recoverable after use, for use in agriculture and horticulture. European Committee for Standardization, Brussels, BelgiumTechnical report
39. Liu EK, He WQ, Yan CR (2014) "White revolution" to "white pollution"—agricultural plastic film mulch in China. Environ Res Lett 9:091001. https://doi.org/10.1088/1748-9326/9/9/091001
40. Liu M, Lu S, Song Y, Lei L, Hu J, Lv W, Zhou W, Cao C, Shi H, Yang X, He D (2018) Microplastic and mesoplastic pollution in farmland soils in suburbs of Shanghai, China. Environ Pollut 242:855–862

Publisher's Note