Production and characterisation of environmentally relevant microplastic test materials derived from agricultural plastics

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HIGHLIGHTS

- Environmentally relevant microplastic test materials are need to advance research.
- Five large batches of microplastic test materials were produced from mulching films.
- Thorough characterisation provides an in-depth description of particle properties.
- Material composition and selected method can introduce challenges for micronisation.
- Trade-offs exist between obtaining certain particle properties and associated costs.

GRAPHICAL ABSTRACT

ABSTRACT

Soil environments across the globe, particularly in agricultural settings, have now been shown to be contaminated with microplastics. Agricultural plastics – such as mulching films – are used in close or direct contact with soils and there is growing evidence demonstrating that they represent a potential source of microplastics. There is a demand to undertake fate and effects studies to understand the behaviour and potential long-term ecological risks of this contamination. Yet, there is a lack of test materials available for this purpose. This study describes the manufacture and characterisation of five large (1–40 kg) batches of microplastic test materials derived from agricultural mulching films. Batches were produced from either polyethylene-based conventional mulching films

or starch-polybutadiene adipate terephthalate blend mulching films that are certified biodegradable in soil. Challenges encountered and overcome during the micronisation process provide valuable insights into the future of microplastic test material generation from these material types. This includes difficulties in micronising virgin polyethylene film materials. All five batches were subjected to a thorough physical and chemical characterisation - both of the original virgin films and the subsequent microplastic particles generated - including a screening for the presence of chemical additives. This is a critical step to provide essential information for interpreting particle fate or effects in scientific testing. Trade-offs between obtaining preferred particle typologies and time and cost constraints are elucidated. Several recommendations emerging from the experiences gained in this study are put forward to advance the research field towards greater harmonisation and utilisation of environmentally relevant test materials.

1. Introduction

Microplastic test materials can be defined as standardised and well-characterised batches of particles available for use in scientific testing (Emons et al., 2006; Linsinger et al., 2001). Test materials are necessary for quality assurance and quality control (QA/QC) of both scientific experimentation and certification of technologies. They are a crucial component in facilitating cross comparability of analyses and hazard assessment: they are needed to verify the efficacy of analytical methods (e.g. through recovery testing or as analytical standards) and for the investigation of environmental processes or hazards (Uriano et al., 1977). Existing, available test materials are scarce (Seghers et al., 2022) and, as such, there is an urgent demand for the production of batches of test materials from the research field.

The lack of available test materials links to several critical challenges associated with their design and production. First, microplastics found in the environment vary widely in their properties, including size, shape, polymer type, additive content, and extent of ageing (Binda et al., 2024; Rochman et al., 2019). Nearly infinite combinations of these variables are possible, and a classification system that can feasibly account for all dimensions of this variability does not yet exist. These various particle characteristics all have the potential to influence the fate and impact of microplastics in the environment. To effectively test these processes and effects, reference particles are needed that represent different particle typologies, relevant to the specific aims and scope of experimentation. There is a need to better define what represents environmentally relevant test materials and then consistently produce batches of materials embodying those characteristics (Kokalj et al., 2024).

Several challenges have been identified related to the generation of microplastic particles. A number of approaches to manufacture particles have been used thus far, including both direct synthesis of microplastic particles (e.g. Dong et al., 2020; Mitrano et al., 2019; Schmiedgruber et al., 2019) and generation of small particles from larger plastic pieces, for example by cutting (e.g. Mossotti et al., 2021; Welden and Cowie, 2016), (cryo)milling (e.g. McColley et al., 2023; Ogonowski et al., 2016; Sarkar et al., 2021; Seghers et al., 2022), sonicating (e.g. von der Esch et al., 2020), and washing of textiles (e.g. Hurley et al., 2018; Schell et al., 2022). These production techniques are, however, typically limited in their capacity to generate large volumes of small and homogeneous particles. There is an urgent demand in the microplastic research field to increase harmonisation. Consolidating techniques and quality assurance in the production of test materials is key to address several important aspects of this demand. In particular, greater harmonisation can be facilitated by the production of sufficiently large batches of test materials which can fulfil several scientific studies, instead of each study producing a new batch of material which are then potentially poorly comparable. This is especially the case for hazard assessments where large batch experiments, mesocosm or field scale studies require a considerable volume of test materials.

Within the microplastic research field, agriculture practices and, in particular, the use of agricultural plastics has emerged as an important topic (Hurley and Nizzetto, 2018; Ng et al., 2018; Tian et al., 2022). Agricultural plastics are often used in direct contact or close proximity with the environment and are subject to exposure to environmental conditions – such as direct solar radiation, fluctuating temperatures, and mechanical stress – for the duration of their use, which may initiate the release of microplastic particles to the environment. Emerging monitoring studies have already identified residues from agricultural plastics in soil environments across the globe (e.g. Huang et al., 2020; Katsumi et al., 2021; Li et al., 2022; Ranneklev et al., 2019; van Schothorst et al., 2021). There is an urgent need to characterise the potential risks posed by this microplastic contamination to environmental health, including on soil quality and the long term sustainability of agricultural production (Steinmetz et al., 2016). At present, no microplastic test materials based on agricultural plastics are currently commercially available. There is, therefore, a need to generate environmentally relevant microplastic particles from these source materials to supply scientific testing.

Based on a scoping of agricultural plastic products used in Europe, mulching films were identified as the target product for microplastic test material production. Mulching films are used widely across the globe for the cultivation of a variety of crop types as a means of increasing yields. This is achieved by modulating soil temperature and moisture and reducing pressures from weeds or pests (Steinmetz et al., 2016). These films are applied in direct contact with soils and may degrade and/or fragment during use or once they are removed from fields (in the case of conventional, non-biodegradable polymer types) as a result of weathering and mechanical stress (Huang et al., 2020; Steinmetz et al., 2016). Mismanagement of mulching film waste – often linked to challenges associated with waste handling – has also been observed in agricultural settings (Briassoulis, 2023), where this mismanagement can also lead to the release of fragments to soils. Thus, mulching films likely represent an important potential source of microplastics to soils and the generation of test materials derived from mulching films represent a priority for assessing the potential risks posed to soil or environmental health.

Polyethylene (PE) is the most common synthetic polymer used in the production of conventional agricultural mulching films (Sarkar et al., 2018). A significant proportion is specifically linear low density polyethylene (LLDPE), which is preferred based on its puncture resistance, tensile strength, and resistance to solar radiation and low temperature conditions (Espí et al., 2006). Microplastics derived from LLDPE mulching has been identified in farmed soils in China, where mulching films were used (He et al., 2018a). A longer list of studies have observed mulching film fragments in agricultural soils, but do not differentiate LLDPE from the broader class of PE (Huang et al., 2020; Liu et al., 2018; Wang et al., 2021; Zhou et al., 2019).

Blends of starch and polyester of various grades are now commercially available and used to produce mulching films (Serrano-Ruiz et al., 2021). These materials are biodegradable in soil and certified according to relevant international standard specifications (EN 17033, 2018; ISO 23517, 2021). The biodegradation of selected biodegradable mulching films in soil has also been investigated under laboratory and experimental field conditions (Briassoulis, 2007; Rudnik and Briassoulis, 2011). Starch-polybutadiene adipate terephthalate (PBAT) blends are commonly used to produce biodegradable mulching films. PBAT represents the largest share of the global production capacity for bioplastics, and this is expected to continue to expand in the coming years (European Bioplastics, 2022). Biodegradation of PBAT, and mulching

films that contain PBAT, has been demonstrated in test soils (Souza et al., 2019; Zumstein et al., 2018). The time for complete degradation can, however, vary in different climates and under real field conditions (Sintim et al., 2020); hence, residues from these films can transitorily accumulate in soils. Microplastics derived from starch-PBAT blend mulching films have, for example, been observed in farm soils in Norway (Ranneklev et al., 2019).

This study aims to address this specific demand by producing several batches of microplastic test materials derived from relevant agricultural plastic products selected for their likelihood to act as a source of microplastics to the environment. This paper describes the challenges encountered in generating batches of microplastic particles and approaches to overcome these, towards a manufacturing method capable of producing large batches (in the order of tens of kilograms) of test materials. This is supplemented by a thorough physical and chemical characterisation of these materials and the microplastic particle produced.

2. Production of microplastic test materials

2.1. Materials

Rolls of agricultural mulching film composed of LLDPE and starch-PBAT blend were purchased from different manufacturers in Italy and Greece for the purpose of generating microplastic particles. Their characteristics are described in Table S1. Black mulching films with a similar thickness (approximately 15 μm) were selected based on the prevalence of their use in agricultural production in Europe. An additional roll of black mulching film based on low density polyethylene (LDPE) was purchased from Germany to address challenges that emerged during grinding (Section 2.2).

2.2. Micronisation method

The potential for generating microplastic fragments was first investigated through a series of laboratory scale grinding tests using a 6875 Freezer/Mill High Capacity Cryogenic Grinder (SPEX Sample Prep, USA) at the Norwegian Institute for Water Research. Prior to grinding, films were shredded using a benchtop shredder (SHR3D IT, 3DEVO, The Netherlands). Shredded films were placed inside steel grinding vials (capacity: 125 ml) and placed into the cryomill for 3×8 min cycles. Grinding is achieved by agitation of the vial and the presence of a steel bar inside the chamber. Cryogenic conditions are initiated through a pre-cooling phase and maintained by continuous immersion in liquid nitrogen. These tests revealed that i) the capacity and time/cost effectivity of laboratory-scale cryo-micronisation does not facilitate the production of sufficiently large batches of microplastic test materials (i. e. when the requirement is to produce >5 kg); and ii) that laboratory cryomilling equipment does not effectively grind the selected virgin LLDPE-based mulching films, bearing characteristics common to materials available across the European market. Whilst the laboratory grinding tests yielded a batch of starch-PBAT blend particles with a sufficiently fine particle size distribution (M-BIOIT-15-black-A0), the shredded LLDPE that came out of the cryomill resembled the same shredded pieces added, i.e. with no further reduction in size, despite several rounds of cool and grinding. Additional experiments applying physical and chemical stress to pieces of shredded LLDPE yield no further improvements in grinding, including heating to 60 °C and snap freezing in liquid nitrogen and exposure to 30 % H_2O_2 and 10 % KOH.

In order to overcome the limitations identified for laboratory-scale grinding, it was necessary to use commercial industrial-scale cryomilling facilities and simultaneously investigate different lines of innovation and development. Two strategies were followed: 1) virgin films of LLDPE and starch-PBAT blend were shredded and sent to commercial cryomilling facilities; and 2) virgin films were subject to one cycle of reprocessing, representing "artificial" recycling, which included melting

of the film and re-extrusion to form pellets. Recycled film pellets retain the same chemical composition as the virgin films, they have a lower stability and flexibility compared to the new polymer and therefore more closely mimic the properties of films aged under field conditions.

Industrial-scale cryomilling of virgin films encountered some challenges. Several micronisation companies expressed doubts regarding successful processing of the products into a relevant particle size distribution. The first company to indicate a potential to grind the mulching films found that their system provided insufficient cooling and that the particles were affected by static charge, which prevented them from being cleared from the machine. These particles warmed up and melted, and so it was necessary to halt all further grinding to prevent damage to the machine. A second company was identified and provided with the insights gained from the first experience. Their setup provides better control over cooling and uses different screens and suction to clear residual particles and prevent melting. A large batch of starch-PBAT blend microplastics was generated from rolls of virgin film (M-BIOEL-15-black-A0) using a counter-current pin mill with liquid nitrogen as a coolant. Milled particles were screened using a 300 μm mesh and all larger particles were re-entered into the cryomill until >96 % of particles were <300 μm. Particles were collected in paper bags for storage and shipment. The same issues observed for LLDPE from the in-house grinding tests were observed in the industrial-scale cryomilling: the company tested several configurations of their equipment but no effective micronisation of the shredded films was achieved.

Grinding of recycled film pellets was successful. A third company was used for this task, which had smaller cryomilling equipment but with a R&D unit to test grinding of different materials. Sufficient cooling was again identified as an issue that needed to be resolved for the pellets, as for the films. However, the company was able to successfully produce two large batches of microplastic test materials (>30 kg): one starch-PBAT blend (M-rBIO-black-A0) and one LLDPE (M-rPE-black-A0). The equipment passed the milled particles through a 1000 μm screen to ensure 90 % of the particle size distribution was below <1000 μm.

In order to address the lack of PE-based particles derived from virgin films, a further round of laboratory scale grinding tests was conducted. Based on the experiences gained from the many laboratory and commercial grinding rounds, a new mulching film was obtained. This roll was purchased from Germany and also represents a common black mulching film typically used in agricultural production; however, it is composed of LDPE and has a greater thickness (approximately 45 μm versus 15 μm), which was thought to improve the chances of successful micronisation. LDPE films were first pre-cut using a guillotine paper cutter and cryomilled using a Retsch ZM 300 ultra-centrifugal mill (Retsch, Germany) at the University of Bayreuth. The LDPE strips were pre-cooled in liquid nitrogen before being added into a stainless streel sample cassette. To increase the efficacy of micronisation, additional mechanical stress was applied: a spoonful of dry ice was added to the sample cassette. Grinding was performed at a speed of 18,000 rpm and the temperature was maintained at <15 °C for the duration of grinding. The cryomill was fitted with a 350 μm ring sieve to screen particles below this size. With this technique, grinding is achieved based on sheer stress between the ring sieve and the rotating rotor. Milled particles were transferred into LDPE bags for storage and shipment. Due to the laboratory scale of this grinding, a small batch (approximately 2 kg) of LDPE microplastic fragments was produced (M-PEDE-45-black-A0).

The culmination of these grinding trials and efforts yielded a total of five batches of microplastic test materials for scientific testing. Table 1 provides a list of these batches with an assigned test material code based on the PAPILLONS project (PAPILLONS, 2024) agricultural plastic identifier scheme, which identifies the material as mulching film (M), describes the material type (biodegradable or polyethylene-based), film thickness and colour and extent of artificial ageing. Particles produced from artificially recycled film pellets are identified by 'r'. These codes are used henceforth to denote each batch.

Table 1

Summary of the five batches of microplastic test materials generated in the PAPILLONS project, including the approximate size of each batch in weight.

Test material code	Primary polymeric composition	Material	Approx yield
M-BIOIT-15-black-A0	Starch-PBAT blend	Virgin films	1 kg
M-BIOEL-15-black-A0	Starch-PBAT blend	Virgin films	37 kg
M-rBIO-black-A0	Starch-PBAT blend	Recycled film pellets	40 kg
M-rPE-black-A0	LLDPE	Recycled film pellets	40 kg
M-PEDE-45-black-A0	LDPE	Virgin films	2 kg

3. Characterisation of test materials

3.1. Methodology

3.1.1. Determination of mechanical and radiometric properties of the virgin films

The virgin films used to generate microplastic test materials were first subject to a thorough characterisation to investigate their physical properties, including their strength and ductility, which relate to their potential to generate microplastics under real field conditions. Mechanical properties of the five virgin films were assessed at the Laboratory of Farm Structures at the Agricultural University of Athens. The film thickness was measured using a micrometer (byko-test 4500 coating thickness gauge; BYK, Germany) and the tensile properties were measured using an Instron series 5900 dynamometer (Instron, USA) in accordance with the ISO 527-1:2018 test standard (ISO 527-1, 2019; ISO 527-3, 2018). Measurements were taken at 23 ± 1 °C and 50 % relative humidity, in both machine and transversal direction.

Puncture tests were performed by the Institute of Polymers, Composites and Biomaterials of the National Research Council Italy to evaluate the resistance of the mulching films against the pressure exerted by spontaneous weeds. Briefly, 40 mm diameter circular specimens were die cut from the films using a stainless steel hollow punch (Bohem). The film specimens were then sealed to the open mouth of an aluminium test cup using an aluminium threaded flanged ring with two rubber gaskets to secure the specimen in place. The cup assembly was gripped to the lower specimen holder of an Instron 4023 dynamometer (Instron, USA). The film specimen was then punctured from above by a steel probe mounted onto the moving crosshead of the instrument until rupture of the material occurred. A 14 mm diameter spherical probe moving down at a fixed rate of 2 mm min^{-1} was used. The applied load was recorded as a function of the displacement. All the mechanical analyses were performed on at least three specimens of each film type, at room temperature and 50 % relative humidity.

Radiometric properties of virgin films were measured at the University of Bari. Five replicates of 40×40 mm strips of the film materials were prepared for the measurements. Spectral direct transmissivity was measured using a double beam ultraviolet-visible-near infrared (UV-VIS-NIR) spectrophotometer (Lambda 950, Perkin Elmer Instruments, Norwalk, CT, USA). Measurements were taken in the solar wavelength band (200–2500 nm) in steps of 10 nm using radiation with a direct perpendicular incidence. Spectral total transmissivity and reflectivity were measured by means of an integrating sphere (diameter 100 mm) used as the receiver of the spectrophotometer, with a double beam comparative method. Spectral diffuse transmissivity was calculated as the difference between total and direct transmissivity.

Spectral transmissivity and reflectivity of the virgin films were measured in the long wave infrared radiation (LWIR) range using a Fourier Transform Infrared (FT-IR) spectrophotometer (1760 X, Perkin Elmer Instruments, Norwalk, CT, USA) in steps of 4 cm^{-1}. Spectral transmissivity was measured using radiation with a direct perpendicular incidence, whilst reflectivity was measured at near normal incidence (10°).

The radiometric coefficients of the virgin films were calculated for different wavelength bands: the solar wavelength range (200–2500 nm), photosynthetic active radiation (PAR, 400–700 nm), and LWIR (7500–12,500 nm). The radiometric coefficients in the solar wavelength band were calculated as the weighted average value of the spectral transmissivity using the spectral distribution of the solar radiation at ground level as the weighting function. The transmissivity and reflectivity coefficients of the virgin films in the LWIR range were calculated as average values of the spectral transmissivity and reflectivity in the wavelength range from 7500 to 12,500 nm.

3.1.2. Analysis of the size and morphology of the obtained particles

The size and morphology of the particles obtained after cryomilling were examined using both light and electron microscopy. The particles were examined under a stereomicroscope (Nikon SMZ 745 T) at 10-50× magnification at the Norwegian Institute for Water Research. A series of micrograph images were taken using a camera accessory (Infinity 1) were taken by carefully spreading the particles across a white glass fibre filter paper to provide a high contrast background. The filters were first traversed under the microscope to separate any overlapping or clustered particles. Particle size was then assessed based on these images using ImageJ, by analysing the Feret's diameter of the particles using the Analyze Particles function. The measurements were calibrated using a measurement standard and the lower size limit was set to 10 μm based on the resolution of the camera.

Particle morphology and surface texture were further examined using scanning electron microscopy (SEM). A Philips XL-30 Field Emission Gun-Scanning Electron Microscope with a 20 keV beam under high vacuum conditions was used for this purpose. Plastic samples were placed on a standard aluminium stub sample holder (12.5 mm dia.) and attached on a conductive carbon tape. The samples were uniformly coated with a ~ 5 nm thick gold layer using a Cressington 108 auto vacuum sputter coater (Cressington, United Kingdom) before SEM analysis to make the plastic surface conductive and improve image quality.

3.1.3. Characterisation of chemical composition

The primary chemical composition of the material and subsequent microplastic particles was confirmed using attenuated total reflectance Fourier transform infrared spectroscopy (ATR-FTIR) at the Norwegian Institute for Water Research. Particles were placed onto a diamond ATR accessory on a PerkinElmer Frontier system and scanned at 4 cm^{-1} spectral resolution across a spectral range of 4000–650 cm^{-1}. Each spectrum was composed of a total of 12 co-scans. Background measurements were taken between each sample.

Chemical characterisation of additive content was performed through several approaches. Thermal desorption pyrolysis gas chromatography mass spectrometry (TD-Pyr-GC/MS) was used to identify volatile additives in the materials. Samples were analysed using a Multi-Shot Pyrolyzer (EGA/PY-3030D, Frontier Labs), connected to a GC system GC-2020 (Shimadzu Corporation), coupled with triple quadrupole mass spectrometry detector and electronic ionisation (70 eV) Mass Detector TQ8040 (Shimadzu Corporation) at the Institute of Polymers, Composites and Biomaterials of the National Research Council Italy. The gas chromatograph was equipped with an Ultra Alloy® Metal Capillary Column (Frontier Labs stationary phase 5 % di-phenyl-methylpolysiloxane, with an inner diameter of 250 μm, a film thickness of 0.25 μm and a length of 30 m). Interfaces of the pyr-GC and GC/MS were kept at 300 °C and 250 °C, respectively. The carrier gas was helium at a controlled flow of 1.78 ml min^{-1}. The split ratio was 1/50 of the total flux. Blank measurements were taken before each analysis, by placing an empty crucible in the furnace and performing thermal desorption or pyrolysis in the same conditions as described above.

Soxhlet extractions were also performed at the Institute of Polymers, Composites and Biomaterials of the National Research Council Italy. To identify non-volatile additives, 5 g samples of the virgin films were cut and placed in a Soxhlet for 48 h using isopropanol or methanol as the extracting solvent. The resulting solutions were concentrated and an aliquot was filtered and subjected to analysis. Chromatographic separation was carried out using an Agilent 1290 ultra-high-performance liquid chromatography (UPLC) system (Agilent Technologies, USA). A reversed-phase C18 analytical column of 2.1×100 mm and 1.8 μm particle size (Hypersil GOLD, Thermo scientific) was used. The injection volume was 5 μl and the flow rate was set at 0.6 ml min^{-1}. Mobile phases consisted of water (A) and methanol (B). The chromatographic run started with 20 min isocratic at 20 % of A and 80 % of B, followed by the first gradient to increase the percentage of B to 100 % at 1.0 min. This was held for 10 min before a second gradient decreased the percentage of B to 80 % at 1 min. The analysis was carried out using a quadrupole time-of-flight mass spectrometer: a 6500 Q-TOF LC/MS (Agilent Technologies, USA) equipped with a Dual Jet Stream electrospray ionisation (ESI) source operating in positive ionisation mode at the full scan. The ESI source settings comprised a gas temperature of 320 °C, gas flow of 8 l min^{-1}, nebuliser gas pressure of 35 psi, sheath gas temperature of 350 °C, sheath gas flow of 11 l min^{-1}, capillary voltage of 3500 V, nozzle voltage of 1000 V, and fragment voltage of 175 V. The instrument is equipped with an Ultraviolet (UV) Diode Array Detector (DAD) for the recognition of the analytes of interest that contain rings or double bonds which absorb UV light.

The occurrence of a list of 20 plastic additive compounds (Table S2) selected prior to the analysis was investigated using an UPLC 1290 Infinity LC (Agilent Technologies, USA) coupled to an Agilent Triple Quadrupole G6495 B (Agilent Technologies, USA) mass spectrometer equipped with an ESI source at the University of Chemistry and Technology, Prague. The analytical method is described in Dvorakova et al. (submitted), this issue. Briefly, one gram of the test material was extracted with ethyl acetate for four weeks. Ultrasound assisted extraction was performed for 2 h each day. To control for background contamination, a procedural blank (ethyl acetate only) was prepared for each batch of samples. A volume of 5 μl of obtained extracts was injected onto a BEH C18 column (Waters, USA) (100×2.1 mm; 1.7 μm), maintained at 40 °C, with 5 mM ammonium acetate in water (A) and methanol (B) as mobile phases. The gradient was as follows: 0.0–1.5 min: 10–50 % B at 0.3 ml min^{-1}; 1.5–4 min: 50–100 % B at 0.3 ml min^{-1}; 4–10 min: 100 % B at 0.4 ml min^{-1}; 10.0–10.1 min: 100–10 % B at 0.3 ml min^{-1}; and 10.1–12 min: 10 % B at 0.3 ml min^{-1}.

The metal ions content of the virgin films was investigated using inductively coupled plasma mass spectrometry (ICP-MS) analysis. The samples were first mineralised using an Anton-Paar Multiwave 3000 Microwave digestion system, following EPA Method 3052. A Perkin Elmer ICP-MS model Nexion 300× (Perkin Elmer Inc. Waltham, Massachusetts, U.S.A.) was used to measure metal ions content by following EPA Method 6020. The results were normalised based upon the blank mineralised sample, and values were reported in mg kg^{-1}. Each determination was performed in triplicate.

3.2. Results and discussion

3.2.1. Mechanical and radiometric properties of the virgin films

The results of the tensile strength, puncture resistance, and radiometric tests performed on the virgin films are presented in Table 2. The tensile properties of conventional PE films with an average thickness of 15 μm (M-PEIT and M-PEEL) showed higher tensile strength in the machine direction (MD) and a lower strength in the transverse direction (TD). The corresponding elongation at break values are high, as expected for this ductile material. M-PEEL had higher elongation at break values in TD than in MD direction (usual for PE films) while M-PEIT exhibited the opposite behaviour. The third conventional mulch film, with an average thickness of 43 μm (M-PEDE), showed lower tensile

Table 2
Properties of the source material mulching films used for the generation of microplastic test materials. MD: machine direction; TD: transversal direction; PAR: photosynthetically active radiation; LWIR: long wave infrared radiation.

		Conventional			Biodegradable	
		M-PEIT	M-PEEL	M-PEDE	M-BIOIT	M-BIOEL
Tensile tests						
Thickness (μm)	MD	15.0 ± 5	15.4 ± 0.6	43 ± 0.6	9.0 ± 3	15.1 ± 0.8
	TD	–	15.0 ± 0.9	43.8 ± 0.7	–	14.7 ± 0.2
Tensile strength (MPa)	MD	40.8 ± 6.2	33.0 ± 4.9	24.6 ± 0.6	26.1 ± 2.0	29.3 ± 1.7
	TD	19.1 ± 2.3	28.7 ± 2.2	20.6 ± 0.9	16.1 ± 1.7	28.5 ± 3.7
Elongation at break (%)	MD	376.8 ± 3.7	400.0 ± 18.6	225.7 ± 15.8	300.1 ± 7.9	363.8 ± 35.1
	TD	322.3 ± 2.5	608.3 ± 20.0	654.6 ± 20.5	207.7 ± 6.8	567.2 ± 43.3
Puncture tests						
Maximum load (N)		20.6 ± 3.2	11.1 ± 4.1	22.0 ± 4.0	2.1 ± 0.4	10.6 ± 2.5
Displacement (mm)		24.3 ± 1.3	18.4 ± 8.3	18.8 ± 0.6	2.4 ± 0.5	15.6 ± 5.0
Energy at failure (J x 10^{-3})		284.1 ± 2.7	139.1 ± 30.0	217.2 ± 38.4	4.7 ± 0.7	143.3 ± 1.1
Radiometric tests						
Solar total transmissivity (%)		0.1	1.8	1.5	0.8	8.4
Solar diffuse transmissivity (%)		0.0	0.2	0.0	0.4	2.7
PAR total transmissivity (%)		0.0	0.4	0.0	0.1	3.7
PAR diffuse transmissivity (%)		0.0	0.1	0.0	0.1	2.5
LWIR transmissivity (%)		17.7	45.1	46.9	15.9	20.9
LWIR reflectivity (%)		5.3	4.1	4.8	4.4	6.5

strength values than the thinner films, whilst the elongation at break value in TD was much higher than the one in MD. The elongation at break value in MD was the lowest one among the three conventional PE films. The variation observed between the films is attributed to different compositions (e.g., LDPE versus LLDPE, as well as additives) and the processing conditions during film blowing.

The two biodegradable mulching films (M-BIOIT and M-BIOEL), with an average thickness of 9 and 15 μm, respectively, present a different behaviour. The thinner M-BIOIT had a very low tensile strength in the TD and relatively low elongation at break values, especially in the TD. The biodegradable 15 μm thick film (M-BIOEL) had good tensile strength comparable to that of a strong conventional film in both directions, and likewise, comparable high elongation at break values.

Puncture testing primarily involves tear and stiffness and measures the resistance of a test specimen to puncture. This represents an important property in flexible films. In general, the puncture properties of mulch films are proportional to the thickness of the specimens, with thicker films typically exhibiting a higher maximum puncture force and puncture elongation (Guo et al., 2023). The mechanical behaviour of the selected mulching films was recorded as the maximum load, as a function of the displacement (Schettini et al., 2008). The data obtained, alongside the energy to failure – calculated as the area under the experimental curves – are reported in Table 2. The PE films showed comparatively higher maximum load and displacement values compared to the biodegradable films. The specific reason for this cannot be ascertained from this dataset alone but has previously been linked to the differences in material composition and the regularity of macromolecular organisation (Schettini et al., 2008). Furthermore, as

expected the thickness of the specimens is an important factor in determining their puncture properties, as is demonstrated from the maximum load value of the M-PEDE film.

The analysis of the data relative to the biodegradable films highlights a significantly lower mechanical performance ascribed to these materials with respect to PE. Generally, both displacement and maximum load are reduced. This is likely due to the different cohesive force of the biodegradable blend, made up of PBAT and thermoplastic starch (TPS), compared to the plain PE matrix. Furthermore, a large variability was noticed for the M-BIO films in terms of the maximum load and displacement. These results indicate degradation effects occurring under storage prior to (and following) purchase, which should be considered when biodegradable films are used in mulch applications. This variability indicative of degradation has already been highlighted in other studies (Hayes et al., 2012). Indeed, mulch films undergo ageing processes due to weathering agents (solar radiations, high air temperature and relative humidity, etc.) and the employment of chemical substances during the cultivation period (Giaccone et al., 2018). The combination of these effects with long storage time before applications could result in unpredictable premature failure of the films during use.

The values of the radiometric coefficients of the tested virgin mulching films in the solar (200–2500 nm), in the PAR (400–700 nm), and in the LWIR (7500–12,500 nm) ranges are shown in Table 2. The main radiometric requirement of a black mulching film is to be opaque to the PAR radiation – i.e. it must have a very low PAR total transmissivity coefficient – for the purpose of reducing weed development (Vox and Schettini, 2007). The conventional and biodegradable black mulching films tested showed very low PAR total transmissivity coefficients, ranging from 0.0 % (M-PEIT and M-PEDE) to 3.7 % (M-BIOEL). These coefficients show the high capacity of the tested materials to reduce weed growth, as required by this type of mulching application. The LWIR transmissivity coefficient strongly influences soil temperature in the case of black mulching films. The lower the LWIR transmissivity coefficient of the mulching film, the higher the soil temperature under the mulching film. Among all the tested materials, the M-BIOIT film was the most effective in decreasing heat losses by radiation with the lowest LWIR transmissivity coefficient.

3.2.2. Size and morphology of the obtained microplastic particles

The original material composition, the form of input material, and the mode of micronisation all appear to yield slight differences in the physical properties of microplastic particles generated. Fig. 1 presents micrograph images of the five different batches generated and Fig. 2 provides further elaboration on the morphology and size of the particles

with SEM imaging. A notable difference in morphology is between the batches produced directly from films and those generated from recycled film pellets. In two dimensions, there is a similar mixture of medium and low circularity particles between most of the individual batches (Fig. 1); yet, the SEM images reveal greater and more variable particle thickness and a greater irregularity in particle morphology in the third dimension for the recycled film pellets (i.e. M-rPE-black-A0 and M-rBIO-black-A0 versus M-PEDE-45-black-A0, M-BIOEL-15-black-A0, and M-BIOIT-15-black-A0; (Fig. 2). This highlights the influence of the source material (film versus pellet) in the final particle morphologies produced.

This difference in morphology is most apparent when comparing M-BIOEL-15-black-A0 with M-rPE-black-A0 or M-rBIO-black-A0. The film fragments produced by large scale commercial grinding (M-BIOEL-15-black-A0) retain their original thickness more effectively than is observed for laboratory grinding of virgin films (M-PEDE-45-black-A0 and M-BIOIT-15-black-A0), and more closely resemble film fragments (Fig. 2d,e). They are also substantially more angular in their morphology. Laboratory grinding of starch-PBAT blend films (M-BIOIT-15-black-A0) yielded particles that more closely resemble ground recycled film pellets (M-rBIO-black-A0). In addition, laboratory grinding resulted in a more irregular surface texture than was observed for commercial ground starch-PBAT blend films (Fig. 2f,i).

Another notable difference, particularly evident in Fig. 2, is between the two batches derived from recycled film pellets: M-rPE-black-A0 an M-rBIO-black-A0. Despite being subject to the same cryomilling procedure at the same commercial facility, the LLDPE material – which was the most challenging to micronise (Section 2.2) – is characterised by highly irregular particles where larger particles exhibit non-uniform filaments or tendrils extending from the main particle body and smaller particles vary between filamentous and more rounded forms (Fig. 2j,k). The particles appear to have undergone stretching during cryomilling to produce these morphologies, which is likely related to the ductility of the LLDPE films (e.g. the high elongation at break; Section 3.2.1) which potentially also underpinned the grinding challenges encountered (as described in Section 2.2).

The addition of dry ice to facilitate laboratory grinding of the thicker LDPE film (M-PEDE-45-black-A0) yielded a large number of very small particles. Fig. 2a,b reveals a high density of fine particles adhering to larger film fragments, and this is reinforced by the particle size distribution presented in Fig. 3. In fact, laboratory grinding of both LDPE and starch-PBAT blend films (M-PEDE-45-black-A0, M-BIOIT-15-black-A0) yielded a greater proportion of finer particles (<200 μm) than the commercial grinding of films or recycled film pellets (M-BIOEL-15-black-A0, M-rBIO-black-A0, M-rPE-black-A0). This can also be seen in
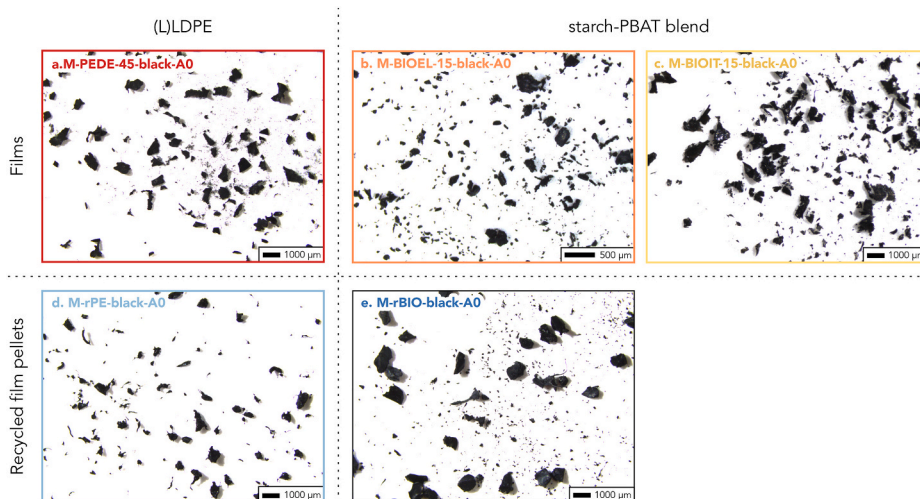

Fig. 1. Micrograph images of the microplastic particles in the five batches of test materials produced. The images are divided by polymer type (a, d and b, c, e) and source material type (a-c and d, e) to facilitate a visual comparison of the particle morphologies generated. Note that the scale bar varies between images.
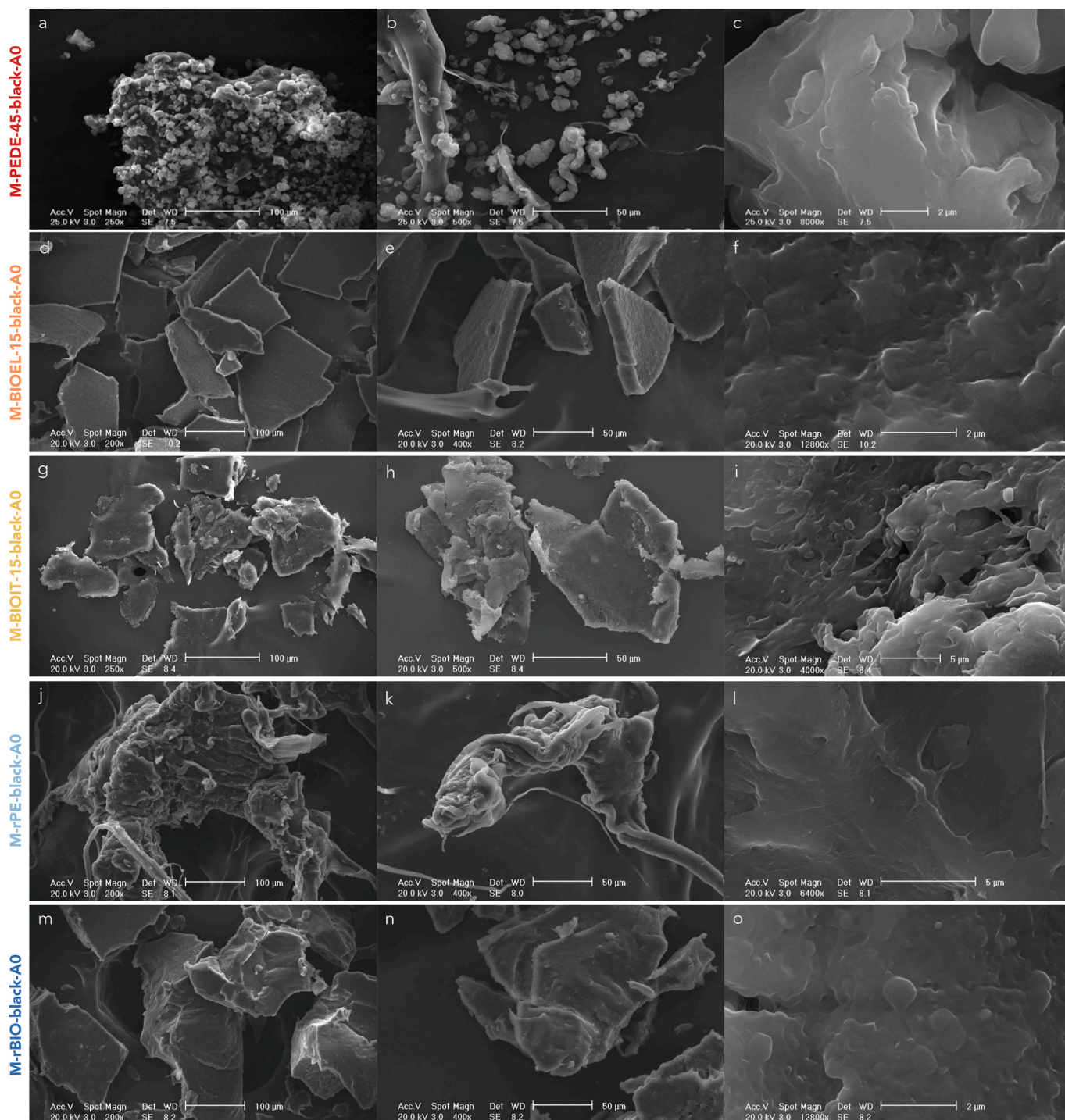
Fig. 2. Scanning electron microscopy images of the five batches of microplastic test materials. Three images with three different magnifications are included per batch (a-c, d-f, etc.) to demonstrate differences in particle morphology and surface texture. Note individual scale bars at the base of each image.

the similar particle size distributions observed for these laboratory-ground films (Fig. 3). M-BIOEL-15-black-A0 – the commercially ground starch-PBAT blend film – had comparatively fewer very small particles (i.e. <100 μm) but all particles were below 500 μm as a result of the use of 300 μm sieve that returned larger particles to the equipment to regrind (Section 2.2). The LLDPE recycled film pellets yielded the coarsest size distribution (Fig. 3). The factors which introduced challenges in grinding LLDPE films likely play a role in limiting the efficacy of micronisation of the recycled film pellets.

The occurrence of very small particles associated with M-PEDE-45-

black-A0 may also indicate the production of nanoplastic particles as part of the grinding process. In this study, the particle size characterisation had a lower limit of 10 μm. As such, we cannot confirm the presence of particles in the nano-range; however, many of the particles analysed in the size analysis were close to the limit of detection. Further characterisation is needed to investigate the occurrence of nanoplastic particles potentially produced for all batches. The highest magnification SEM images do not immediately reveal the presence of sub-micron particles but other techniques could confirm this with greater accuracy and representativity. Other studies have investigated the use of
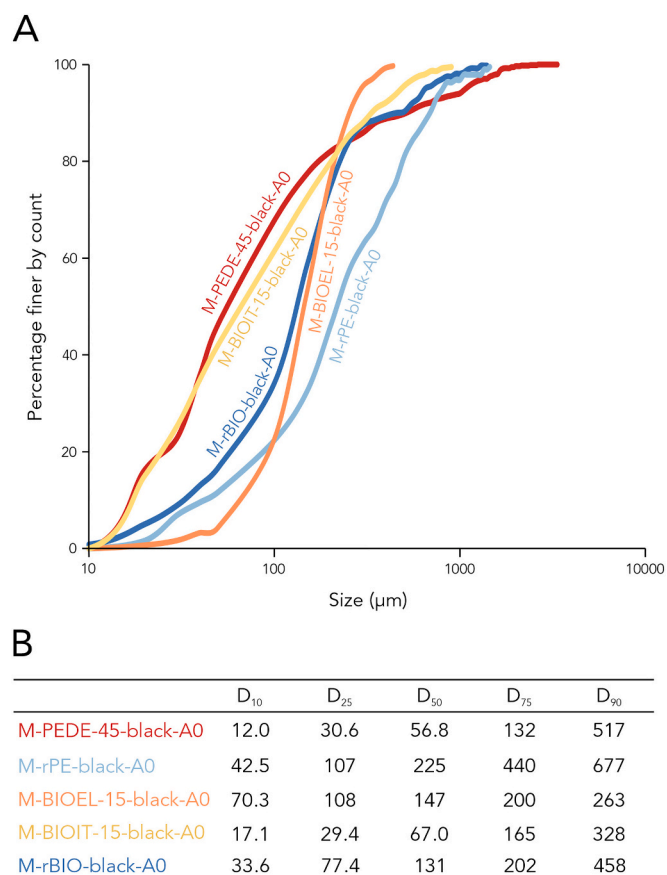
A

B

	D_{10}	D_{25}	D_{50}	D_{75}	D_{90}
M-PEDE-45-black-A0	12.0	30.6	56.8	132	517
M-rPE-black-A0	42.5	107	225	440	677
M-BIOEL-15-black-A0	70.3	108	147	200	263
M-BIOIT-15-black-A0	17.1	29.4	67.0	165	328
M-rBIO-black-A0	33.6	77.4	131	202	458

Fig. 3. Particle size distributions of the five test material batches shown as plots showing the cumulative proportion finer than by count (A) and a summary of percentiles (B). Note the log scale used in A.

grinding to produce nanoplastic (e.g., McColley et al., 2023) and found that longer griding times are capable of producing nanoplastics for certain investigated polymer types. Thus far, only a single pioneering study has investigated the presence of nanoplastic in agricultural soil environments (Wahl et al., 2021). No study has yet identified nanoplastic in fields subject to mulching film use. Further research is needed to both assess the occurrence of nanoplastics from agricultural plastic in soils and to further develop techniques to manufacture environmentally relevant particles in the nano-range.

3.2.3. Chemical characterisation

Fig. 4 presents ATR-FTIR spectra for the five batches of test materials. Chemical characterisation of the material performed by IPCB-CNR and UCT combining results from pyr-GC/MS, TD-GC/MS, UHPLC-MS/MS, and ICP-MS are listed for each batch in Supplementary Tables S3-S7.

The main characteristic FTIR peaks for PE are 2940 and 2865 cm^{-1} (C—H stretching), 1460 cm^{-1} (CH$_2$ bending and asymmetric CH$_3$ bending), 1370 cm^{-1} (symmetric CH$_3$ bending), and 720 cm^{-1} (CH$_2$ rocking in the long chain) (Noda et al., 2007). These are present in the spectra for both M-PEDE-45-black-A0 and M-rPE-black-A0, the two PE-based batches (Fig. 4a,b). It is difficult to separate LLDPE and LDPE using FTIR analysis, particularly when other constituents of the material are also present; however, upon inspection of 1377 cm^{-1} relative to 1368 cm^{-1}, the spectra are confirmed to be either LLDPE or LDPE, as opposed to other higher density forms of PE (Jung et al., 2018). Pyr-GC/MS analysis of M-rPE-black-A0 identified alkanes, alcohols, and alkenes that are typical degradation productions from the pyrolysis of PE (Table S6). Additional peaks at 3300 and 1600 cm^{-1} observed in the FTIR spectrum for M-rPE-black-A0 represent carbonyl and hydroxyl

groups (Campanale et al., 2023), indicating that the artificial recycling process indeed induced simulated ageing of the material. Similar peaks are also observed for the biodegradable test materials, further indicating degradation of the materials during storage (Section 3.2.1).

The FTIR results also confirm that the fragments produced from the starch-PBAT blend material indeed contain a PBAT component. Characteristics peaks at 3000–2800 cm^{-1} (C—H stretching in the aliphatic and aromatic portions), 1710 cm^{-1} (carbonyl groups in the ester), 1267 cm^{-1} (C—O in the ester linkage), and 720 cm^{-1} (four or more adjacent methylene groups) (Kijchavengkul et al., 2008) are present in the spectra for M-BIOEL-15-black-A0, M-BIOIT-15-black-A0, and M-rBIO-black-A0 (Fig. 4c-e). Pyrolysis products derived from the degradation of PBAT were also identified for all starch-PBAT blend based test materials (Tables S4, S5, S7). The large peak between 1200 and 900 cm^{-1} in the FTIR spectra indicates the presence of starch, and therefore confirms that the material is a PBAT-based starch blend (Fig. 4c-e). 1,6-anhydro-beta-D-glucopyranose – a pyrolysis product derived from the breakdown of cellulose was identified in the pyr-GC/MS analysis of M-BIOEL-15-black-A0, M-BIOIT-15-black-A0, and M-rBIO-black-A0, further indicating the presence of starch (Tables S4, S5, S7). In the FTIR data, there are slight spectral differences between the batches produced from virgin films (M-BIOEL-15-black-A0, M-BIOIT-15-black-A0) and the one generated from the recycled film pellets (M-rBIO-black-A0), in the regions 1680–1570 cm^{-1} and 3390 and 3180 cm^{-1}. This indicates that the artificial recycling process does introduce some changes in the chemical composition, potentially introduced by the mechanical and thermal stress of reprocessing. However, no differences were observed in the organic additives detected in M-rBIO-black-A0 and M-BIOEL-black-A0 (Tables S4 and S7), which both derive from the same original material (Table S1), so this stress did not appear to dramatically alter the additive content.

All the FTIR spectra, with the exception of M-PEDE-45-black-A0, present a non-linear baseline with an increase in slope with decreasing wavenumber (Fig. 4b-e). This is indicative of the presence of carbon black (Datta et al., 2017), which is a common additive in black agricultural films (Briassoulis, 2006; Tocchetto et al., 2001). The lack of a baseline shift seen for M-PEDE-45-black-A0 suggests a lower carbon black content. The presence of carbon black at a lower concentration in the material is, however, indicated by the identification of aluminium, cobalt, and manganese through ICP-MS analysis, all of which are typical contaminants deriving from carbon black (Table S3; Wang et al., 2012).

In addition to the main constituents of the materials, several additives were identified (Tables S3-S7). This includes a series of light stabilisers and antioxidants to slow the degradation of the material, anti-slip agents used for the production of films, and plasticisers to deliver the properties of the mulching film materials (Hahladakis et al., 2018). These results represent a qualitative assessment of the presence of additive compounds, and so the relative quantities of additives in the material is not known. Yet, this characterisation is important to help explain potential observed effects in studies that utilise microplastic test materials to assess potential hazards.

In this study, this characterisation also reveals the potential variability between similar products. Two of the batches of microplastic test materials (M-BIOEL-15-black-A0 and M-BIOIT-15-black-A0) were produced from two different products that, on the market, appear to have several similarities: same main material composition, colour, and thickness. However, the additive characterisation reveals a different profiles of detected organic additives (Tables S4 and S5). In the case where effects studies link observed results to the role of additives, it is important to note that these additives can vary even in similar products. This also highlights the need for thorough characterisation of all test material batches produces, to reveal these potentially important differences.
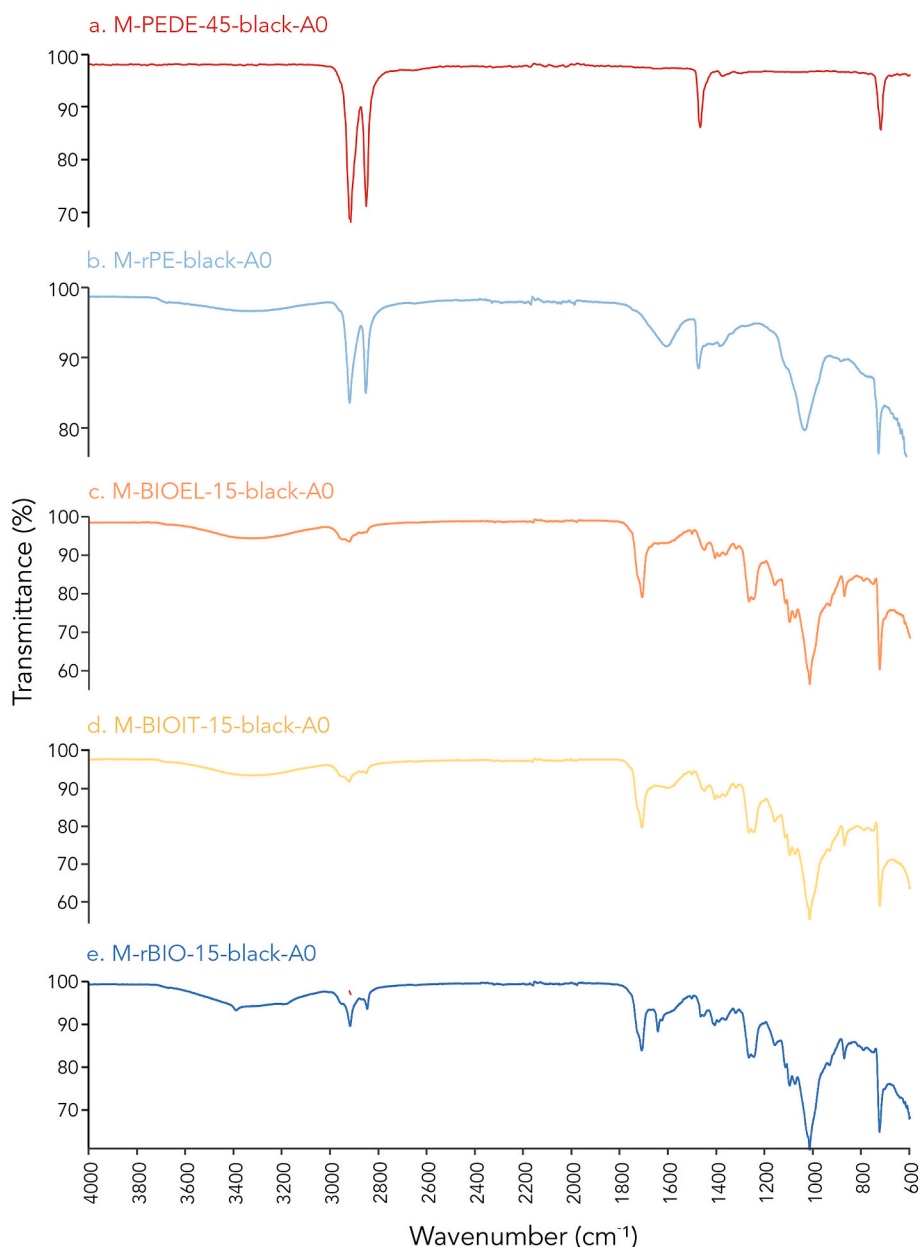
Fig. 4. Fourier transform infrared (FTIR) spectra of the five batches of test materials (a-e). Note that y-axes vary between plots due to differences in absorption/transmittance.

4. Advances towards the production and characterisation of environmentally relevant microplastic test material

Several batches of microplastic test materials were successfully produced from different mulching film products. Yet, several challenges emerged during the generation of these materials which are relevant for the further development of the field.

For example, several challenges were encountered when attempting to micronise LLDPE-based mulching films (M-PEIT and M-PEEL; Table S1). It is necessary to adapt micronisation approaches to facilitate the production of microplastic particles from these materials: using thicker films or reprocessing films into pellets, adding dry ice as an additional mechanical stress in grinding, and opting for smaller scale grinding over larger scale facilities with a poorer control over cooling all appeared to present valuable opportunities to encourage improved micronisation. Longer grinding times have been identified in the literature as effective in achieving micronisation (McColley et al., 2023);

however, it appears that the material composition (polymer type, the processing conditions, and – potentially – the additives used) plays an important role on the efficacy of grinding. The composition of the LLDPE films is such that the material is reasonably resistant to mechanical and chemical stress, particularly in a virgin state. Fragmentation of PE-based agricultural films has been noted in fields (e.g. Ramos et al., 2015; Zhou et al., 2019); however, this is following deployment for one or more seasons where weathering due to environmental exposure may also be further enhanced by mechanical stress during removal from the field and mishandling after use (Bhattacharjee et al., 2023; Liang et al., 2024). At this point, the material may have degraded sufficiently, so as to facilitate the release of microplastic particles – thus explaining the occurrence of debris in soils.

Artificial ageing of the materials prior to grinding could therefore help by simulating and accelerating this weathering process and facilitate improved yields of LLDPE microplastic test materials in the future. Unfortunately, however, this opportunity faces new challenges. Namely,

the physical dimensions of ultraviolet (UV) radiation chambers used for artificial ageing and the duration of exposure required: the material must be laid flat to be sufficiently and homogeneously exposed to the UV radiation and a long exposure time is needed to achieve a 50 % reduction in tensile resistance of the material. Over 50 m^2 of film equates to approximately 1 kg of material (approximately, 50 m^2 for LLDPE and approx. 70 m^2 for starch-PBAT blend), which is far beyond the dimensions of UV chambers. Thus, only small amounts of films – in the order of grams – can be subject to artificial ageing at a given time, drastically increasing the time required to produce large batches of test materials. On this basis, it was not feasible to artificially age the material using UV to encourage micronisation in this case. Whilst the environmental relevance of unaged microplastic particles is already subject to discussion (Rozman and Kalčíková, 2022), this challenge of manufacturing large quantities of aged particles is a critical hindrance. There are, therefore, trade-offs to consider between what is ideal to produce and what is possible to produce. The artificially recycled film pellets used here may represent a useful compromise, where FTIR analysis indicated some ageing of the materials. However, further research and technological advancement is required to evaluate new techniques to rapidly and homogenously age larger volumes of mulching films or simulate ageing through other techniques. This could include, for example, different forms of artificial ageing such as inducing mechanical or thermal stress, or simulating biofilm formation (Astner et al., 2023; Binda et al., 2024).

The results of the physical characterisation of the microplastic particles produced in this study suggest that the grinding method can influence the particle size and shape of the microplastics generated. This finding could be further applied to specifically tailor the micronisation technique to the desired final particle properties. For example, commercial grinding of starch-PBAT blend films yielded particle morphologies that most closely resemble fragments of films that have retained their original thickness – albeit with fewer fine particles – whilst laboratory-scale grinding of a similar starch-PBAT blend film generated a larger proportion of small microplastics, with more spherical and subrounded morphologies. There is a need to better characterise the morphology of microplastics derived from mulching films that are found in real agricultural soils, to understand what particle morphologies are released through fragmentation of agricultural mulching films under field conditions. This includes a description of the particle size and shape- which is often not reported in detail in occurrence studies – and could be further improved with SEM imaging of real mulching film fragments produced in real agricultural soils. This is a critical step to identify environmentally relevant particle properties, where micronisation approaches can then be tailored to more accurately mimic genuine mulching film residues.

There is often a desire for smaller particle sizes for use in ecotoxicological and risk assessment studies, as these particles are more likely to be ingestible by different soil fauna (He et al., 2018b; Kim et al., 2024; Kokalj et al., 2024). Finer particles were more effectively produced through laboratory-scale grinding (Fig. 3), suggesting that this approach should be used to address this demand. However, this also produces the lowest yields (Table 1) and requires substantial labour, energy, and resource (e.g. liquid nitrogen) inputs. This introduces an important cost consideration. In addition, there is a competing need to improve harmonisation within the microplastic research field. This can be facilitated by producing sufficiently large batches of test materials, such that multiple studies can be conducted using the same materials. In this case, commercial grinding of films or recycled film pellets can satisfy this need, producing several tens of kilograms of microplastic particles. However, new trade-offs are subsequently introduced: using commercial grinding limits the reproducibility of test material production as companies do not reveal extensive details about the specific grinding method or apparatus as this represents proprietary information linked to market competitiveness. This in turn may hinder wider harmonisation in the research field.

The physical and chemical characterisation undertaken in this study reveal several differences between microplastic test materials generated from different parent materials. This includes two biodegradable mulching films (M-BIOEL and M-BIOT) that initially appear as similar products – both with the same primary composition, colour, and thickness – but in fact exhibit varying results in their mechanical and radiometric properties (Section 3.2.1) and differences in their additive content (Tables S4 and S5). This highlights the need for thorough chemical characterisation of all batches of microplastic test materials produced, to ensure that the specific properties are established. In addition, these differences *even* for similar products indicate the urgent need for further research on effective methods for producing test materials from a wider variety of plastic mulching materials (e.g., composed of other polymer types such as polylactic acid), agricultural plastics (such as greenhouse films, irrigation pipes, etc.), and other sources of microplastic to the environment, followed by thorough characterisation in how the microplastics generated may differ in important ways.

Mulching films are widely used globally, and the results presented here demonstrate that these products can vary in their physical and chemical properties. Based on this variability in composition and the lack of disclosure by manufacturers about material properties, several important considerations have been identified for the generation of microplastic test materials:

i) The choice of the parent agricultural plastic material (in this case, different types of mulching film) needs to be carefully framed around considerations of the scope and objective(s) of the study to be conducted, as well as how commonly they are used and the likelihood for releasing microplastic contamination (based on their characteristics and use). This is fundamental to make sure that the research conducted using the obtained test materials has environmental and societal relevance.

ii) The prioritisation described in point i should be based on a consideration of the physical and technical characteristics of the selected materials. Hence, a detailed characterisation of the technical, physical, and radiometric properties of the parent materials should be included in the broader information package accompanying the individual test materials.

iii) The selection of a micronisation method needs to balance costs and benefits, which are often dependent on the production volumes required. Whilst laboratory-scale manufacturing can potentially achieve better control over the desired properties of the generated particles, they are often insufficient with regard to producing larger batches, for example to feed mesocosm or field-scale experiments. Large batches are also crucial to create the stocks that enable reproducibility and repeatability of experiments, and represents an important step towards greater harmonisation in the research field.

iv) The morphological characterisation of the generated test materials should leverage on both light and scanning electronic microscopy, to reveal a more complete understanding of the three-dimensional structure and nanoscale surface features of the generated particles. Characterisation of test materials should also closely follow emerging research related to the size and morphological characterisation of real debris found in soils, including pioneering research on nanoplastics derived from agricultural plastics.

v) Analysis of the chemical composition should be as comprehensive as possible, leveraging on both target and non-target screening and a range of complementary extraction and detection techniques. This provides important detail that can potentially explain observed fate and effect results.

5. Summary

The present study systematically addressed the challenge of producing large batches of microplastic test materials representative of soil contamination originating from the degradation of agricultural mulching films. The availability of such materials is pivotal for enabling research and advancing knowledge in the field of fate, distribution, and impacts of this pollution. A catalogue of five distinct, large batches of microplastic test materials are presented, which can be used as a proxy for studying the behaviour and effects of microplastics derived from the use of common conventional and biodegradable (in soil) agricultural mulching films.

Several insights obtained during the production and characterisation of these test materials reveal important opportunities to advance techniques for generating microplastic particles from complex material types, generating particles in large quantities, and adapting production methods to obtain different priority properties. However, several trade-offs associated with producing large batches of environmentally relevant particles in a time- and cost-effective manner were also revealed. These trade-offs represent shortcomings in producing particles that mimic environmental particles with a very high degree of accuracy, and must be considered when evaluating reference or standard test materials as a proxy for environmental contamination.

CRediT authorship contribution statement

Rachel Hurley: Writing – original draft, Visualization, Project administration, Funding acquisition, Formal analysis, Conceptualization. **Gilberto Binda:** Writing – review & editing, Formal analysis. **Demetres Briassoulis:** Writing – review & editing, Funding acquisition, Formal analysis, Conceptualization. **Sabrina Carola Carroccio:** Writing – review & editing, Formal analysis. **Pierfrancesco Cerruti:** Writing – review & editing, Formal analysis. **Fabiana Convertino:** Writing – review & editing, Formal analysis. **Darina Dvořáková:** Writing – review & editing, Formal analysis. **Sarmite Kernchen:** Writing – review & editing, Formal analysis. **Christian Laforsch:** Writing – review & editing. **Martin G.L. Löder:** Writing – review & editing. **Jana Pulkrabova:** Writing – review & editing. **Evelia Schettini:** Writing – review & editing, Formal analysis. **Davide Spanu:** Writing – review & editing, Formal analysis. **Aristeidis S. Tsagkaris:** Writing – review & editing, Formal analysis. **Giuliano Vox:** Writing – review & editing, Formal analysis. **Luca Nizzetto:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Rachel Hurley reports financial support was provided by European Commission. Rachel Hurley reports financial support was provided by Research Council of Norway. Christian Laforsch reports financial support was provided by German Research Foundation. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2024.174325.

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