Agcumulate: A study on the accumulation of microplastics in soils and terrestrial ecosystems

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Declaration

We, the undersigned declare that this thesis entitled ***Agcumulate*: A study on the accumulation of microplastics in soils and terrestrial ecosystems** is entirely the author's own work and has not been taken from the work of others, except as cited and acknowledged within the text.

The thesis has been prepared according to the regulations of Dundalk Institute of Technology and has not been submitted in whole or in part for an award in this or any other institution.

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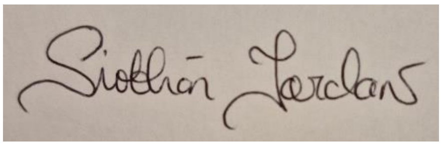
Supervisor Signature 

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List of publications and conference presentations

Referred journal paper

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Conferences/presentations

International Conference on Microplastics, Nanoplastics and Human Health, 2024, University City Dublin (UCD). Poster and two-minute oral presentation: The abundance, characteristics and potential sources of microplastic in Irish agricultural soils across different farming land-use types.

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Environ 2021, University College Cork (UCC). Oral presentation: Farmers' attitudes towards agricultural plastics – management and disposal, awareness and perceptions of the environmental impacts: preliminary results.

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List of Abbreviations

ANOVA	Analysis of Variance
ABS	Acrylonitrile butadiene styrene
BPA	Bisphenyl A
BS	Biosolids
CEC	Cation Exchange Capacity
CF	Chemical Fertiliser
CO	Carbon Monoxide
CO_2	Carbon Dioxide
CS	Cattle Slurry
CSO	Central Statistics Office
CW	Constructed wetland
DEHP	Bis(2-ethylhexyl) phthalate
DOC	Dissolved Organic Carbon
DRS	Deposit Return Scheme
EPA	Environmental Protection Agency
EU	European Union
EVA	Ethylene-vinyl acetate
FYM	Farmyard Manure
HCl	Hydrochloric Acid
HDPE	High Density Polyethylene
H_2O_2	Hydrogen Peroxide
H_2S	Hydrogen Sulfide
ICW	Integrated Constructed Wetland
IFFPG	Irish Farm Film Producers Group
IUCN	International Union for Conservation of Nature
LDPE	Low Density Polyethylene
LLDPE	Linear Low-Density Polyethylene
LOI	Loss-on-Ignition

MP(s)	Microplastic(s)
MPPs	Microplastic particles
MUB	Modified Universal Buffer
NaBr	Sodium Bromide
NaCl	Sodium Chloride
NaI	Sodium Iodide
NaOH	Sodium Hydroxide
NBR	Nitrile butadiene rubber
$NH4^+$	Ammonium
NOAA	National Oceanic and Atmospheric Administration
OM	Organic Matter
PA	Polyamide
PAEs	Phthalic acid esters
PAH	Polycyclic Aromatic Hydrocarbons
PBA	Poly (Butyl Acrylate)
PBAT	Polybutylene Adipate Terephthalate
PBSA	Polybutylene Succinate Adipate
PC	Polycarbonate
PCDF	Polychlorinated Dibenzofurans
PCL	Polycaprolatone
PCPs	Personal Care Products
PDAP	Polydiallyl Phthalate
PDBE	Polybrominated diphenyl ethers
PE	Polyethylene
PES	Polyester
PET	Polyethylene Terephthalate
PFOS	Perflourooctanesulfonic acid
PFAS	Per and polyfluoroalkyl substances
PHA	Polyhydroxylalkanoates

PLA	Polylactic acid
PLM	Plastic Mulch
POP	Persistent organic pollutants
PMMA	Polymethyl methacrylate
PNP	Para-nitrophenol
PP	Polypropylene
PPS	Polyphenylene sulfide
PS	Polystyrene
PTFE	Polytetrafluoroethylene
PU	Polyurethane
PVC	Polyvinyl Chloride
PVDF	Polyvinylidene Fluoride
PVF	Polyvinyl Flouride
RoI	Republic of Ireland
SD	Standard Deviation
SDGs	Sustainable Development Goals
SE	Standard Error
SO_2	Sulphur Dioxide
SOC	Soil Organic Carbon
SOM	Soil Organic Matter
SUPs	Single-Use Plastics
UK	United Kingdom
UN	United Nations
US	United States
UV	Ultraviolet
W.W.	Wet weight
WWTP	Wastewater treatment plant
$ZnCl_2$	Zinc Chloride

Agcumulate: A study on the accumulation of microplastics in soils and terrestrial ecosystems

CLODAGH KING

ABSTRACT

Microplastics are tiny plastic particles (≤ 5 mm) that are raising much concern due to their potential effects on living organisms, since plastics can leach chemical constituents that can be harmful, some known to possess endocrine disrupting or carcinogenic properties. Microplastics also have the capacity to absorb and act as vectors for organic pollutants and heavy metals in the environment. Microplastics have been found across various ecosystems including terrestrial environments, freshwater environments, marine, air and recently in the human body. Microplastics are well studied in aquatic environments, however; research on microplastic pollution in terrestrial environments including agricultural soils is limited. This research is the first in Ireland to take a holistic approach to addressing microplastic pollution in agricultural soils. A social science study examining the attitudes and behaviours of Irish farmers toward agricultural plastics revealed that while most farmers actively recycle agricultural plastic waste and recognise the importance of agricultural plastics in farming operations, they also expressed significant concerns about the environmental impact of agricultural plastics. Through field studies, this research shows that microplastics are prevalent in agricultural soils across multiple farming land-uses in Ireland with soils applied with biosolids and plastic mulch films containing significantly higher concentrations of microplastics in comparison to soils without these amendments. Pot trial experiments were conducted to investigate the potential effects of microplastics on the growth of two grassland species, and on soil chemical and biological properties. The results were variable, with certain types of microplastics having positive and negative effects or no impact. In the final study, microplastics were abundant in domestic wastewaters from a rural community in Ireland, as were the pond sediments in the wastewater treatment facility. One of the main issues that stems from this is that the sludges produced in these systems are often applied to agricultural land as fertiliser, which introduces microplastics into agricultural soils and the wider environment. This research highlights the pervasive presence of microplastics in terrestrial ecosystems and identifies numerous areas for future related research to build on. Addressing the issue of microplastic pollution in terrestrial environments will require a collaborative approach involving researchers, innovators, industry leaders, agricultural stakeholders, the general public, and policymakers.

Chapter 1: General Introduction

1.1. Introduction

Plastics have become integral to modern society due to their durability, versatility and cost-effectiveness effectiveness (Harrison and Hester, 2018). Plastics enter almost every facet of modern life, serving a wide range of applications in sectors such as packaging, construction, medical, automotive, agricultural, and more (Sahoo et al., 2023). Plastic is not just a single material; rather, it is a family of materials with diverse origins and properties. Plastics can be manufactured and shaped into an unlimited variety of textures and forms, offering a range of strength, rigidity and flexibility (Dennis, 2024). Since the 1980s, the developments in plastics have been transformative, leading us to what is now termed the “Age of Plastics” (Xu et al., 2022). Global plastic use is expected to triple from 2019 to 2060, rising from 430 million tonnes to 1,312 million tonnes (OECD, 2022). One of the most significant consequences of plastic consumption is the mismanagement of plastic waste. Plastic debris has been detected worldwide and the release and accumulation of plastic waste in the environment is presenting serious ecological risks (Kwon et al., 2023).

A major pressing concern is the proliferation of microplastics, which are tiny plastic particles that are generated through the breakdown of larger plastics that often contain hazardous chemicals (Andrady, 2017; Aurisano et al., 2021; Lasker and Kumar, 2019). In some cases, microplastics are deliberately produced and incorporated into specific products, eventually migrating into natural environments (European Parliament, 2018). There are approximately over 16, 000 known chemicals in plastics and of these only 6 % are currently subject to international regulation (PlastChem, 2024). More than 4,200 of the chemicals contained in plastics are classified as concerning because they are persistent, mobile, bioaccumulative and/or toxic (Wagner et al., 2024). Ten groups of chemicals are identified as being of major concern and include specific flame retardants, “forever chemicals” such as per and polyfluoroalkyl substances (PFASs), phthalates, bisphenols, biocides, certain metals, polycyclic aromatic hydrocarbons (PAHs) and other non-intentionally added substances (UNEP, 2023).

To date, considerable research has focused on the impact of microplastics in aquatic ecosystems, where their presence is now ubiquitous. The United Nations (UN) has provided international bodies with data on how plastics are adversely affecting marine life, and has recognised marine plastics and microplastics under 13 of the 17 Sustainable Development Goals (SDGs) (Usman et al., 2022). Research on microplastics in marine and freshwater ecosystems has taken precedence; however, recent years have seen an increasing awareness of the comparative lack of studies on microplastics in terrestrial ecosystems. In agriculture, plastics are used extensively as they provide benefits in terms of crop protection, water efficiency and productivity (Lakhiar et al., 2024). As a result, agricultural soils may become contaminated with microplastics through various practices, including plastic mulching, the use biosolids (a by-product of the wastewater treatment process) as fertiliser, and general degradation of plastic materials used in field operations such as bale wraps and tyres, amongst others (Huang et al., 2020; Radford et al., 2023). While research on microplastics in terrestrial ecosystems is growing it remains in its early stages. There is a significant knowledge regardings the occurrence and impacts of microplastics on soils, despite their critical importance to agriculture, food security, and overall ecosystem health.

Soils do not receive the same level of legal protection in the European Union (EU) as air and water, and it is now estimated that up to 60 to 70 % of soils are currently deemed in an unhealthy state and are depleting (European Commission, 2020). The degradation of soil is multifaceted, driven by both natural and human-induced factors. To name but a few, these include natural soil erosion and loss of soil organic matter through excessive tillage and overuse of agrochemicals, deforestation and land-use changes, climate change, and inadequate soil protection policies (Jie, et al., 2002; Panagos et al., 2016). To combat this, the EU have announced a Soil Strategy for 2030, and proposals for an EU soil health law, including a directive on Soil Monitoring and Resilience (European Commission, 2023). In both European and Irish contexts, knowledge on the accumulation and effects of microplastics in agricultural soils, as well as their sources, remains limited. Current legislation on soil health is insufficient for the sustainable management of soils, and especially in addressing the specific potential threats by microplastics. To bridge these knowledge gaps, research on microplastics in agricultural soils and their respective sources must be undertaken.

1.2. Aims and Objectives

The overall aim of this research was to investigate the abundance, sources, and potential impact of microplastics on selected agricultural production systems and agricultural communities in Ireland.

More specifically, the objectives of this study are to,

1. Conduct a survey to assess the behaviours and attitudes of Irish farmers towards the usage and disposal of agricultural plastics and to evaluate farmers' awareness of microplastics and their perceptions of the overall impacts of plastics on the environment.

2. Quantify and compare the abundance and characteristics of microplastics in Irish agricultural soils across multiple land-use types, including permanent grassland soils, tillage sector soils and enterprises that utilise plastic mulch films and biosolids.

3. Perform two mesocosm experiments to examine the effects of microplastics commonly found in agricultural soils on the growth of two widespread Irish grassland species, *Lolium perenne* and *Trifolium repens*. Additionally, evaluate the impact of these microplastics on a sandy loam soil representative of grassland systems in parts of Ireland.

4. To quantify the abundance, characteristics and removal efficiency of microplastics from an Integrated Constructed Wetland (ICW) system that receives domestic wastewater from a rural community in Ireland, focusing on microplastics in raw wastewater, treated water and sediments.

1.3. Outline of thesis

This thesis consists of four main studies that relate to plastics and microplastics in terrestrial ecosystems.

Chapter 1 briefly outlines the context of this research by introducing the field of plastics and microplastics in the environment, including the rationale for this study, based on which the research objectives are developed.

Chapter 2 presents a literature review on the history, development and fate of plastics in the environment, the types of plastics used in agriculture, the management of agricultural waste, and recycling schemes available in Ireland. Moreover, the review provides the status of microplastic pollution in agricultural soils worldwide including potential sources and effects on soil, plants and biota. The next section of the review focuses on microplastics in wastewater, and microplastics in constructed wetland (CW) systems. The final section provides an overview of national, European and international policies and initiatives to tackle plastic and microplastic pollution in the environment.

Chapter 3 is the first experimental study entitled "**Farmers' attitudes towards agricultural plastics – management and disposal, awareness and perceptions of the environmental impacts**". A social science-based study that evaluates the behaviours and attitudes of Irish farmers towards farm-plastics, their awareness of microplastics and their perceptions of the potential threats that plastics pose to elements in the aquatic and terrestrial environment. This is the first published study worldwide to examine farmers' attitudes toward microplastic and plastic pollution.

Chapter 4 is the second experimental study entitled "**The abundance, characteristics and potential sources of microplastic in Irish agricultural soils across different land-use types**". This study involved an assessment of twenty-four agricultural fields, each grouped into four different land-use types, including permanent grassland soils, tillage soils, and soils where plastic mulch films and biosolids have been applied. This is the first study carried out on microplastic contamination in Irish agricultural soils across multiple land-use types.

Chapter 5 is the third experimental study entitled "**Microplastic-induced changes in soil chemistry, enzymatic activity and biomass in grassland mesocosms**". This study involved spiking a sandy loam soil planted with *Lolium perenne* and *Trifolium repens*, in both monoculture and mixed sward, with three common types of microplastics found in agricultural soils, applied at three different concentrations. The aim was assess their effects on plant growth, soil pH, and the activity of three soil enzymes. To date, this is the first study to explore the impact of microplastics on *L. perenne* and *T. repens* in a mixed sward under a temperate climate conditions.

Chapter 6 is the final experimental study entitled "**The abundance, characteristics and removal efficiency of microplastics from an Integrated Constructed Wetland (ICW) system in Glaslough, Co. Monaghan, Ireland**". This study analysed raw wastewater, treated water and sediment samples to identify the characteristics and potential sources of microplastics originating from domestic sources, given that biosolids or sludges commonly applied to agricultural land are typically derived from domestic wastewater. This is the first study to investigate microplastics within an ICW system in Ireland.

Chapter 7 provides a general discussion summarising the key findings from the research, in addition to the main conclusions and recommendations for future research.

Chapter 2: Literature Review

2.1. The history and development of plastics

John Wesley Hyatt invented the first semi-synthetic polymer in 1869. He treated cellulose derived from cotton fibre with camphor and the material produced could be transformed to imitate shapes and objects to replace natural ivory, the hard white material obtained from the tusks and teeth of elephants (Pathak et al., 2014) Later in 1909, synthetic plastics were first commercialised by Belgian-American chemist Dr. Leo Baekeland. *'Bakelite',* a thermosetting plastic was formed through a condensation reaction with phenol and formaldehyde (Chandrasekaran, 2017). The result was a stiff, strong, lightweight and easily molded product, which at the time revolutionized product design. Widespread, large-scale plastic production began in the 1930-40s (Bläsing and Amelung, 2018). The Second World War played a significant role in the development of plastics. During that time, plastic production in the United States (US) increased by 300 %, with the equivalent of 371 million kg of plastic produced for military use alone (Meikle, 1995). In 1949, for the first time plastic polymers were competing with glass materials for food and beverage packaging. By the 1960s, the majority of common commodity plastics that are still in use today had been synthesised and manufactured (Crawford and Quinn, 2017). In the decades after, the surge in plastic production continued to rise, however; society started to become concerned about plastic waste and realised that despite the fact that many plastic products are made disposable; they last forever in the environment (Ryan, 2015). Nevertheless, modern plastic production continued to rise exponentially, which continues to this day. In the 1980s, plastic production companies were the first to offer recycling as a solution and helped develop the plastic waste and recycling industry (Crawford and Quinn, 2017).

2.2. The fate of plastics

In 1950, the global production of plastics amounted to 1.5 million tonnes (Wright et al., 2013). Since then, the production of plastics is believed to have increased by up to 9 % per year (Hirai et al. 2011), despite a drop in production during the 1973 oil crisis and the 2007 financial crisis (Crawford and Quinn, 2017). In 2009, the global production of plastics reached 250 million tonnes (Nuelle et al., 2014), which by 2014 had further increased to 311 million tonnes per year (Plastics Europe, 2015). This represented an annual increase in production of approximately 25 % in just five years. By 2050, it is anticipated that up to more than 33 billion tonnes of plastic will be produced (Ryan, 2013). Between 1950 when the mass production of plastics began, up until 2015, there had been roughly 8,300 million tonnes of plastics produced globally (Geyer et al., 2017). From the cumulative generation of plastics produced since the 1950s, it is estimated that around 30 % are still currently in use. However, as of 2015, roughly 6,300 million tonnes of plastic waste have been generated, and of this, only 9 % has been recycled, 12 % incinerated, leaving 79 % accumulating in landfills or the natural environment (Geyer et al., 2017). In 2024, it was estimated that up to 220 million tonnes of plastic waste was generated and of this waste, up to one-third or approximately 69.5 million tonnes was mismanaged, ending up in the natural environment (SAFE, 2024). Plastic wastes are susceptible to physical disintegration (fragmentation) resulting in the generation of smaller sized plastic particles, also known as microplastics (Zhang et al., 2021a).

2.3. Agricultural plastics

Plastics have created huge opportunities in modern agriculture as farmers heavily depend on plastic materials for mulching crops, wrapping silage, storing feed and fertiliser, greenhouses, polytunnels, and piping (Plastics Europe, 2020), and for many farmers, there are no alternative materials available. Plastics are also used to store and transport agricultural products (Razza and Cerutti 2017), and medicinal and artificial insemination injection products made from plastic are used to treat farm animals (Bas et al. 2011; Rethorst 2015). The most commonly used plastic polymer in agriculture is polyethylene (PE) and its derivatives low density polyethylene (LDPE), linear low-density polyethylene (LLDPE), and high-density polyethylene (HDPE). LDPE polymers are present in films for mulches, greenhouses, tunnels, irrigation tapes and silage/fertiliser bags (American Plastics Council, 1996). Polypropylene (PP) is the second most used plastic resin in agriculture found in fibres and filaments used to produce netting and twine to tie around bales (Scarascia-Mugnozza et al. 2012). Polyvinyl chloride (PVC) was once a widely used base polymer in agriculture for mulching (Brown 2004), but has been banned in the last twenty years due to its toxicity and carcinogenic properties (Steinmetz et al., 2016). Ethylenvinylacetate (EVA), a co-polymer of ethylene and vinyl acetate (Scarascia-Mugnozza et al., 2012), is mainly used for greenhouses and piping because they are highly durable, long lasting and inexpensive, insensitive to temperature and resistant to corrosion (Brown 2004). Other plastics such as polymethylmethacrylate

(PMMA) and polycarbonate (PC) are less frequently used in farming (Plastics Europe, 2020), but are also available as alternative materials to glass in the fabrication of greenhouses (Ali et al., 2015).

Table 2.1: Characteristics of the most common agricultural plastics, and their recyclability under the Irish Farm Film Producers Group Scheme.

Common plastics used in agriculture	Polymer[1] (most common)	Colour	Recyclable in Ireland under the Irish Farm Film Producers Group scheme (Y/N)
Silage wrap and sheeting	PE	Mainly black and white but also come in other various colours (green, pink, etc.)	Yes
Plastic mulch films	Conventional mulch (now banned for sale in Ireland): PE Oxodegradable: can be either HDPE, LDPE, PP, PS, PET or PVC in combination with additives (metal salts) to facilitate breakdown in the presence of UV-light and O_2 Biodegradable: Mainly poly lactic acid (PLA), starch, cellulose, polyhydroxyalkanoates	Mainly clear or transparent (may come in other colours such as white, black, green, yellow, red, grey (depends on purpose/crop)	No
Netting	PP	Mainly black and white but also come in other colours (e.g., green)	Yes
Twine	PP	Mainly blue, red and green but also come in other colours such as black, white, etc.	Yes
Fertiliser and feedbags	Liner (LDPE) Outer material (PP)	Mainly white with multi-coloured print	Yes
Drums	PE	Mainly blue (may come in other colours)	Yes

Containers	HDPE	Mainly clear or transparent (may come in other colours)	Yes
Pipes	ABS PE PVC	Various colours (e.g., black, white, orange, blue, etc.)	No
Tapes	LDPE	Various colours (e.g., black, white, red, blue, etc.)	No
Greenhouse/polytunnel films/sheets	LDPE PC	Mainly clear or transparent (may come in other colours)	No

[1] (Polymer type) ABS: Acrylonitrile butadiene styrene, HDPE: High-density polyethylene, LDPE: Low-density polyethylene, PET: Polyethylene terephthalate, PLA: poly lactic acid, PS: Polystyrene, PS: Polystyrene, PVC: Polyvinyl chloride

2.3.1. Agricultural plastic waste management

Globally, the amount of plastic waste coming from agricultural practices is estimated within the range of 2 to 6.5 million tonnes per annum (Brodhagen et al., 2017). Most agricultural plastics are low-value, single use materials that may have encountered a high degree of contamination, therefore they are challenging to collect, recycle and reuse and so, recyclers typically ignore them in many jurisdictions (Muise et al., 2016). The cost of transportation, recycling and landfilling fees can lead to illegal dumping, burial or burning of wastes on-site, which subsequently has knock-on effects to the environment through the emission of harmful substances into soil, water and air (Muise et al. 2016). Plastics that are burnt on-site emit pollutants such as carbon dioxide (CO_2), carbon monoxide (CO), hydrogen sulphide (H_2S), sulphur dioxide (SO_2) and dioxins (Scarascia-Mugnozza et al., 2012). Emissions of particulate matter and dioxins are respectively up to 40 and 20 times greater from open-air burning than in controlled incinerator plants (Levitan and Barros, 2003).

The release of pollutants from open emissions can seriously affect local air quality and pose significant risks to human health, for example, disruption of the endocrine system; increased risk of heart disease and stroke (Humblet et al., 2008); endometriosis (Simsa et al., 2010); and carcinogenic effects (Levitan and Barros, 2003). Moreover, the release of toxic substances from plastics burnt on farms may affect the safety of food products produced in nearby fields (Vox et al., 2016). As described by Briassoulis et al., (2013), plastic wastes are often dumped along watercourses, which have been shown to threaten aquatic life, and wastes may be buried in soils, which may cause significant reductions in soil quality and crop yields. Plastics released into the environment can act as carriers of organic contaminants into the soil environment and other constituents of plastics such as additives like bisphenol A and phthalates leach into the environment and have been shown to exert negative biological effects on terrestrial and aquatic biota (Teuten et al., 2009).

In Ireland, the burning or burial of farm plastics are prohibited under the Air Pollution Act, 1987 and the Waste Management Act, 1996. The Environmental Protection Agency (EPA) of Ireland and Local County Councils are responsible for the enforcement of these regulations. In addition to these, the Waste Management (Farm Plastics) Amendment Regulations 2017, have provided the regulatory framework regarding the recycling of agricultural plastics in Ireland. These regulations promote the collection and recovery of farm plastic wastes.

2.3.2. Waste Management (Farm Plastics) Amendment Regulations, 2017 (S.I. No. 396/2017)

In Ireland, the recovery and recycling of farm plastics is underpinned by the waste management (farm plastics) regulations, with the aim of increasing the recycling of farm plastic waste. The regulations were amended in 2017, providing a new definition for the term "farm plastics". Farm plastics are defined as *"sheeting, netting, bale twine, bale wrap or bale bags composed mainly of polyolefin, including polyethylene, polypropylene or polyvinyl chloride, which is or are suitable for use for the holding, storage or conservation of fodder."*. The regulations impose obligations on the manufacturers and suppliers in relation to the collection and recovery of farm plastics placed on the Irish market. A producer/supplier of farm plastic materials thereby has the choice of either complying directly with the regulations or as part of an approved scheme. Since 2001, The Irish Farm Film Producers Group (IFFPG) is Ireland's only approved farm plastic recycling compliance scheme, whereby producers apply a levy on the sale of farm plastics, which is transferred to the IFFPG for the funding of the collection and recovery of materials.

2.3.3. Recovery and recycling schemes

The IFFPG operates Bring Centres across the country that are collection points where farm plastic waste can be dropped off by farmers, or waste can be collected from the farmyard at a higher price. The producers and suppliers of farm plastics must be members of the IFFPG and have to pay a recycling levy of €240 per tonne of plastic they put on the market (IFFPG, 2023). The scheme also receives extra funding through an additional weight-based collection service, which is charged to farmers. Farmers who return plastics can use codes received from their supplier in order to avail of discounted levies for recycling plastics. Without the codes, the farmer is charged a higher levy for each half tonne of each type of farm plastic they return.

Prices for farmers to recycle plastics have continuously increased in the last recent years. For silage plastics, the cost to farmers is €50 per half tonne when dropped off at Bring Centres, and €100 per half tonne including a minimum of €100 for the call out charge. As for non-silage plastics, fertilizer, and feedbags can be recycled for up to €10 per half tonne and netting and twine are also €10 per bag. The IFFPG report that they recycled up to 40,000 tonnes of farm plastics in 2023, which is the equivalent to the plastic of 20 million silage bales. Since the beginning of its establishment, the IFFPG report they have collected and recycled approximately 450, 000 tonnes of farm plastics. End-uses of recycled plastic wastes include piping, damp proofing products; refuse sacks, and garden furniture (IFFPG, 2023).

Bring Centres are farm plastic waste collection points that run around 235 times a year, and depending on the county, there can be from 1-2 to 10-20 days per year, usually in the same time period where plastics can be recycled under the scheme. These events typically take place in marts, co-operatives, GAA clubs and other local county council facilities. Not all farm plastics are recyclable, and accepted farm plastics include silage wrap and sheeting, netting and twine, large and small fertiliser and feedbags, and drums. Plastic must be segregated and stored in accordance with the guidelines provided by the IFFPG. Non-silage plastic wastes such as netting and twine should be stored in large bulk bags, with the linings removed. Large and small fertiliser and feedbags should be stored separately in large bulk bags with the linings removed, and drums (triple rinsed) must be stored in bulk bags. Farmers are advised to keep their plastics clean and dry because this enables easier recycling, but also because it will lower the weight of the waste resulting in lower recycling charges for the farmer. Farm plastic waste suspected of contamination will not be accepted for recycling.

As there is a minimum weight requirement of half a tonne for each type of farm plastic waste, it can be challenging for some farmers, particularly on smaller farms, to generate enough plastic waste for collection or drop-off, meaning that clean, dry, storage space must be available for more than 1-2 years. This can also be an issue on bigger farms, where a lot of waste is constantly generated in certain seasons and there is not adequate space to facilitate the storage of it. Other agricultural plastic waste such as mulching films, greenhouse and polytunnel sheeting, drinkers and feeders, medical equipment such as animal injection and spray bottles are not accepted for recycling by the IFFPG.

2.4. Microplastics

Small pieces of plastic were first discovered in the aquatic environment in the 1970s. In 1972, vast quantities of small plastic particles were found floating on the surface of the Sargossa Sea (Carpenter et al., 1972). At the time they were referred to as plastic particles, and it was not until 2004 when the modern term 'microplastic' was introduced by Thompson et al., (2004) to describe the small plastic particles accumulating in the sediments and floating on surface waters in Plymouth beaches in the United Kingdom (UK). Thereafter, the scientific community adopted the new term and it was later defined by the steering committee of the National Oceanic and Atmospheric Administration (NOAA) Marine Debris Programme as particles with their longest dimension less than 5 mm in size (Lippiatt et al., 2013) However, while there was an upper size limit established, there were no lower size limits classified at that point. The lower and upper size limits of microplastics are still up for debate and have been considered as 'non-standardised' and ambiguous which can make comparing and reporting results challenging. Crawford and Quinn, (2017) introduced a standardised microplastic size categorisation of pieces of plastic which is depicted in Figure 2.1. Moreover, Frias and Nash, (2019) defined microplastics as *"any synthetic solid particle or polymeric matrix, with regular or irregular shape and with size ranging from 1 um to 5 mm, of either primary or secondary manufacturing origin, which are insoluble in water".* There is now a greater consensus among microplastic researchers that the lower size limit is 1 µm and the upper is 5 mm.

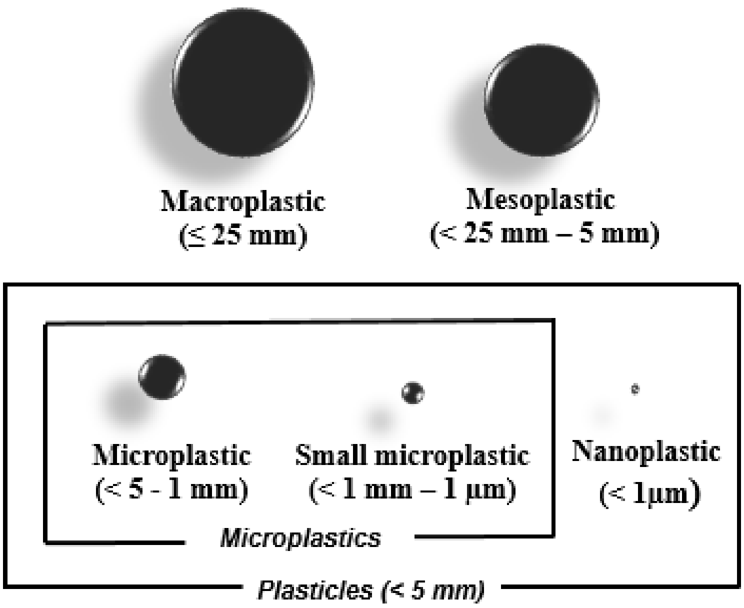

Figure 2.1: Description and illustration of plastic particle size classifications, image adapted by Crawford and Quinn, (2017).

Microplastic pollution has been documented globally across many systems; including freshwaters (Free et al., 2014), oceans (Shim and Thompson, 2015), and terrestrial (de

Souza Machado et al., 2018). As mentioned previously, microplastics are indefinitely defined throughout the literature, however; most authors describe microplastics as small plastic particles < 5 mm in size (Barnes et al., 2009; Lee et al., 2013). Other classifications include 2 - 6 mm (Derraik 2002), < 2 mm (Ryan et al., 2009) and < 1 mm (Claessens et al., 2011). Microplastics persist and accumulate in the environment by direct release or through fragmentation which divides them into two categories; namely primary or secondary microplastics (Da Costa et al., 2019).

2.4.1. Primary microplastics

Primary microplastics are manufactured as microbeads, fibres, pellets or capsules for a specific purpose and are directly released into the environment through various pathways from several sources. Primary microplastics are added to industrial, cosmetic and detergent products for multiple functions including exfoliation, emulsification, suspension, filling, binding, film forming and surface coating (Crawford and Quinn, 2017). They are added to pharmaceutical products to control the release of ingredients present in some drugs (European Commission, 2017), can originate from the erosion of tyres while driving (Reifferscheid et al., 2016) and through the abrasion of synthetic textiles in clothing items during washing (Dris et al., 2016). Synthetic fibres are intentionally manufactured for the production of garments and are typically the most abundant category of microplastics recovered in the environment (Crawford and Quinn, 2017). According to Browne et al., (2011) a single item of synthetic clothing can potentially release up to 1900 plastic fibres in one wash cycle alone and another study found that recycled polyester clothing can potentially release more microplastic fibres than virgin polyester under the same washing conditions (Akyildiz et al., 2024). The International Union for Conservation of Nature (IUCN) estimate that up to 15-31% of all microplastics found in oceans come from primary sources and of those, the majority (98%) are derived from land-based activities (Boucher and Friot 2017).

2.4.2. Secondary microplastics

Secondary microplastics come from various everyday plastic items, originating from the degradation of larger plastic objects such as bags, films, bottles and fishing gear (European Parliament, 2018). Secondary microplastics are generated through the degradation of larger plastic debris into smaller plastic particles, mainly driven by processes involving UV radiation, heat and mechanical stress (Kalogerakis et al., 2017).

Larger plastics become brittle and eventually disintegrate into fragments. Due to the extent of plastic pollution in the environment, studies report that most microplastics belong to this category. Secondary microplastics are indefinitely shaped as a result of the fragmentation process (Efimova et al., 2018). The lifespan of conventional plastics is estimated to be between hundreds to thousands of years depending on the properties of the polymers and surrounding environmental conditions (Zhang et al., 2021). The environmental degradation mechanisms that facilitate the breakdown of plastics are due to biological and/or abiotic processes. Abiotic degradation is a result of a change in the chemical or physical properties of polymers due to light, temperature, water or mechanical forces (Andrady, 2015). Photo-oxidation, which involves the degradation of polymers due to combined action of light and oxygen, is one of the most significant pathways initiating plastic degradation (Zhang et al., 2021), causing the polymer chains to break (chain-scission) or become very brittle resulting in the generation of lower molecular weight compounds (Zhang et al., 2021). Thermal degradation is another mechanism by which plastics breakdown due to elevated temperatures, typically after photo-oxidative processes (Andrady et al., 2022). Certain polymers can absorb sufficient heat, which can result in the breakdown of long-polymer chains and the generation of free radicals (Pirsaheb et al., 2020). Mechanical degradation of plastics occurs due to the action of external forces, which depend on the mechanical properties of plastics. Polymers with lower elongation break values will tend to fragment under external forces and continued stress can lead to chain-scission of molecules (Zhang et al., 2021) . Biotic degradation of polymers refers to the deterioration of plastics, induced by organisms. Larger fauna can physically breakdown macroplastics by biting or chewing, and through physical digestive fragmentation or biochemical processes (Mateos-Cárdenas et al., 2020; Cau et al., 2020). Some microbes also have plastic biodegradation capabilities which have been reported in numerous publications (Giacomucci et al., 2020; Jeon and Kim, 2015; Kyaw et al., 2012). Biodegradation of polymers involves processes such as biodeterioration, fragmentation, assimilation and mineralisation of molecules and the extent to which these occur ultimately depends on polymer characteristics and environmental conditions (Amobonye et al., 2021). Plastic-degrading microbes can transform carbon from long-polymer chains into CO_2 or incorporate the carbon into biomolecules (Alshehrei, 2017).

2.5. Microplastics in agricultural soils

Plastic litter arrives to the soil as meso- (5 mm-5 cm) and macroplastics (> 5 mm). Over time, these plastics are slowly broken down into smaller sized particles, known as micro- (< 5 mm) and nanoplastics (< 0.1 µm). The decrease in size allows for easier integration into soils and increases their surface area (Filella, 2015), bioavailability (de Souza Machado et al., 2020), and ubiquity in the environment. There is mounting evidence of the ubiquitous presence of microplastic pollution in aquatic environments (Eerkes-Medrano et al., 2015; Koelmans et al., 2019), particularly marine systems (Ryan et al., 2009; Andrady, 2011; Wright et al., 2013; Boucher and Friot, 2017; Guo and Wang, 2019), with soil microplastic research receiving far less attention compared to their aquatic counterparts (de Souza Machado et al., 2018). The first publication on microplastic contamination in soils was documented just over a decade ago (Rillig, 2012). Since then, an increased amount of research has been conducted on microplastics found in soils (Corradini et al., 2019; Liu et al., 2018; Zhou et al., 2018; Ding et al., 2020; Corradini et al., 2020) and their effects on soil biota (Huerta Lwanga et al., 2016; Maaß et al., 2017; Boots et al., 2019; Büks and Kaupenjohann., 2020).

One of the main reasons why microplastic research in aquatic ecosystems started much earlier than microplastics on land and in terrestrial ecosystems may be because plastic items and microplastics are more easily noticeable in water. Whereas soil is a much more complex matrix consisting of minerals, organic matter formed over-time by the weathering of rocks and decomposition of plants and animals. Moreover, the lack of standardised methods and the challenges associated with microplastic extraction from soils may also contribute to the delayed onset of microplastic characterisation in soils (Zhang et al., 2019). To date, many different types of plastic polymers have been detected in a range of agricultural soils, in concentrations as little as one microplastic per kg of soil to concentrations as high as 50,000 microplastics per kg of soil. Research on microplastics in agricultural soils has been continuously increasing sine 2017, and a summary of some the main findings from the literature is provided in Table 2.2.

Table 2.2: Evidence and characteristics of microplastic pollution in agricultural soils (summary of the main findings from the literature).

Location	Abundance	Polymer type[1]	Shape	Size	Reference
China (Shanghai)	78.00 ± 12.91 MP items kg^{-1} (shallow soil) 62.50 ± 12.97 MP items kg^{-1} (deep soil)	PP, PE, PS	Fibres, fragments, films, pellets	0.03-5 mm	(Liu et al., 2018)
Germany (Middle Franconia)	0.34 ± 0.36 MP items kg^{-1} (cropland: no sludge or plastic mulch)	PE, PS, PP,	Films, fragments, fibres	2-5 mm	(Piehl et al. 2018)
China	7100-49,960 MP items kg^{-1} (cropland: with sewage sludge and greenhouse film)	-	Fibres, fragments, films	95% of MPs 0.05-1 mm	(Zhang and Liu, 2018)
Chile	0.6-10.4 MP items g^{-1} (with sewage sludge)	-	Fibres, films	Median length = 0.97 mm	(Corradini et al., 2019)
China (Wuhan)	4.3×10^4–6.2×10^5 MP items kg^{-1} (vegetable farmlands)	PE, PP, PS, PA, PVC, Nylon	Fragments, fibres, bead, ball, foam, film	< 500 µm	(Zhou et al., 2019)
China (Baoding City)	-	PP, PA6, PET, PVC	Pellets, fragments	0-35 µm	(Du et al., 2020)
Spain (Valencia)	1015 ± 655 MP items kg^1 (without sewage sludge application) 3660 ± 1790 MP items kg^{-1} (with sewage sludge application)	-	Fragments, fibres, films	50-5000 µm	(van den Berg et al., 2020)
China (Hangzhou Bay)	571 MP items kg^{-1} (with mulching) 263 MP items kg^{-1} (without mulching)	PE, PP, Nylon, PET, Acrylic, PA	Fibres, fragments, films	1-3 mm	(Zhou et al., 2020)

China (Shihezi)	0.1-324.5 kg/ha (cropland with plastic mulch)	PE	-	-	(Huang et al., 2020)
China (Wuhan)	320-12560 MP items kg^{-1} (vegetable farmland)	PA, PP, PS, PE, PVC	Fibres, fragments, foams, beads	70% of MPs <0.2 mm	(Chen et al., 2020)
Chile (Metropolitana region)	540 ± 320 MP items kg^{-1} (cropland) 420 ± 240 MP items kg^{-1} (pasture)	Acrylates, PU, Varnish, PE, PP, NBR, PS, PET, EVA, PA, PLA	Fibres, films, fragments, pellets	Median length = 1.6 mm (Fibres)	(Corradini et al. 2020)
Korea (Yong-In Province)	10-265 MP items kg^{-1} (plastic mulch) 215-3315 MP items kg^{-1} (greenhouse film)	PE, PP, PET, PS, PVC, PVA, PU, PTFE, Acrylic, Epoxy resin	Fragments, fibres, sheets, spherules	0.1-2.51 mm	(Kim et al., 2021)
China (Jiangxi Province)	43.8 ± 16.2 MP items kg^{-1} (with pig manure) 16.4 ± 2.7 MP items kg^{-1} (without pig manure)	PES, PP, PE, Rayon	Fibres, Fragments, films	0.02-5 mm	(Yang et al., 2021)
Northeast Germany	0-217.8 MP items kg^{-1}	PE, PP, nylon, PA, PVDF, PDAP, PMMA, PET, PVF, PVA, PVS	Fibres, foils, platelet, fragments	1-5 mm	(Harms et al. 2021)
Korea	241 ± 52 MP items kg^{-1} (tilled with mulching) 195 ± 37 MP items kg^{-1} (uncultivated land)	PE, PP, PET, PS, PVC	NA	< 2 mm	(Park and Kim, 2022)

	306 ± 56 MP items kg^{-1} (greenhouses)				
Poland	4050 ± 2831 MP items kg^{-1} (sewage treated croplands)	NA	Fibres, Fragments, films, microbeads, pellets	NA	(Medyńska-Juraszek and Szczepańska, 2023)
Hungary	300 ± 93 MP items kg^{-1} (shallow 0-20 cm) 150 ± 93 MP items kg^{-1} (deeper: 2-40 cm) (greenhouses)	PE, PVC, PET, PP	Fibres, films, foams, fragments	0.5–5 mm	(Sa'adu and Farsang, 2022)
Ireland	0–2103 MP items kg^{-1} (biosolids)	Nylon, PES, PET, PP, PMMA, ABS, PC, PS,PU, PVC	Fibre, film, fragment	NA	(Heerey et al., 2023)
Sweden	53,700 ± 5900 MP items kg^{-1} (sewage sludge)	PE, PP, PVC, PES, PA, PLA, PU	Fragments, fibres,	10–500 µm	(Heinze et al., 2024)

[1] (Polymer type) EVA: Ethylene-vinyl acetate, NBR: Nitrile rubber, PA: Polyamide, PA6: Nylon 6, PE: Polyethylene, PES: Polyester, PDAP: Polydiallyl phthalate, PET: Polyethylene terephthalate, PMMA: Polymethyl methacrylate, PP: Polypropylene, PPS: Polyphenylene sulphide, PS, Polystyrene, PTFE: Polytetrafluoroethylene, PUR: Polyurethane, PVA: Polyvinyl alcohol, PVC: Polyvinyl chloride, PVF: Polyvinyl fluoride, PVS: Polyvinyl siloxane

2.6. Sources and pathways

Microplastics can enter agricultural soils through the degradation of farm plastic materials such as silage wrap and plastic mulch films (Huang et al., 2020); but also, through the incorporation of organic fertilisers such as composts (Vithanage et al., 2021), manures (Sheriff et al., 2023), biosolids (Christian and Köper, 2023) and inorganic chemical fertilisers that are coated in synthetic polymers (Lian et al., 2021). Other sources include flooding (Wang et al., 2020), littering and road run-off (Zhang et al., 2020), and atmospheric deposition (He et al., 2018).

2.6.1. Biosolids as a source

Biosolids are sewage sludge that is treated to remove pathogens and heavy metal contamination (Badzmierowski et al., 2021). Biosolids are a nutrient-rich material applied by farmers to fertilise the land, however; the use of biosolids on agricultural land is estimated as one of the largest sources of microplastics into the environment (Hurley and Nizzetto, 2018). During the wastewater treatment process, microplastics are concentrated in the sludge produced. The efficiency of microplastics removal during wastewater treatment depends on the extent of treatment. Wastewater treatment plants (WTTPs) that provide both secondary and tertiary treatment are believed to contribute minimally to the transport of microplastics to oceans and freshwater environments (Carr et al., 2016). However, the sludge produced retains the synthetic fibres released during washing and microplastics from other sources such as cosmetic and medicinal products (Ziajahromi et al., 2017). These microplastics are subsequently spread on land and either accumulate in soils or end up in freshwater and marine ecosystems via surface run-off. In parts of Europe and the US, up to 50 % of the sewage sludge produced is spread on agricultural land and in Ireland, up to 98 % of sludges (biosolids) are reused in agriculture as fertiliser to soil (EPA, 2015; Uisce Éireann, 2023). Mahon et al., (2017) studied the impacts of wastewater treatments on the abundance and characteristics of microplastics and detected concentrations ranging from 4,196 to 15,385 particles kg^{-1}, which were similar to the results collected by (Zubris and Richards, 2005), who reported between 3000 to 4000 particles kg of sludge. Nizzetto et al., (2016) were the first to estimate the amount of microplastics entering European and North American agricultural soils. Their findings suggest that sludge produced and used for agriculture in Europe and the US could contain between 63,000 to 430,000 and 44,000 to 300,000 tonnes of microplastics, respectively, with an estimated 125 to 850 tonnes of microplastics per million inhabitants entering European agricultural soils. In accordance with Directive 86/278/EEC on the protection of the environment, and in particular of the soil, when sewage sludge is used in agriculture, there are specific limit values for heavy metals, pathogens and organic compounds contained in sludge intended for use in agriculture. However, there are no criteria in place to address microplastic contamination in sewage sludge (European Commission, 1986).

2.6.2. Plastic mulch as a source

Plastic mulching is a common agricultural cropping technique that involves covering sowed soil around the base of plants with plastic films to provide favourable conditions for crop growth and yields (Lamont, 2017) The influence of the film on the physical properties of the soil can accelerate plant growth and productivity, improve crop and fruit quality, and promote earlier harvests (Steinmetz et al., 2016). In addition, the use of plastic mulch in crop cultivation has been shown to suppress weed growth, and deter pathogens and pests (Summers et al., 2003). Therefore, it has become a globally applied practice due to its agronomic and economic benefits. Despite the benefits provided by the films during the growing season, in many parts of the world it seems plastic mulching may become the main source of plastic pollution in agricultural soils (Chae and An, 2018). Conventional plastic mulch films, composed largely of PE must be removed from the field. The removal of these films is so labor-intensive and costly, thus plastic films are often tilled back into the soil leaving the remnants of both macro- and microplastic particles in the ground long-term (Brodhagen et al., 2017). To combat the undesirable negative environmental effects of conventional plastic mulch films, several types of films claiming biodegradability were designed and put on the market to provide an appealing alternative method, with farmers and other stakeholders showing great interest in the products (Goldberger et al., 2015). However, photo- and oxo-degradable plastic mulch films also rely on non-renewable petroleum-based plastics for their production (Kasirajan and Ngouajio, 2012). Small concentrations of metal salts are added to these polymers, which aid their degradability in the presence of air and sunlight. This becomes an issue when films are not exposed to sufficient light and oxygen as the degradation of these products can only commence through oxidative and cell-mediated phenomena under specific conditions. Oxo-fragmentable plastics are another term used for these products as the additives present have merely shown to facilitate the fragmentation of films, without fully breaking them down and smaller fragments or films can persist in the environment (Briassoulis and Dejean, 2013). Studies have shown that plastic mulch applications do contribute to microplastics in soil. For example, Huang et al., (2020) investigated the presence of microplastics in agricultural fields in China that were considered as plastic mulch 'hotspots', where plastic mulches were used for over 30 years. They found that the abundance of microplastics present in soils increased over time, with concentrations of 80 ± 49, 308 ± 138, and 1075 ± 346 MP items kg^{-1}, in fields applied with plastic mulch films for 5, 15 and 24 years, respectively.

2.6.3. Other potential sources

Additional potential sources of microplastics into agricultural soils include alternative organic and inorganic fertilisers. The presence of microplastics in different types of manure have been recently determined by Sheriff et al., (2023), and there were $9.02 \times 10^2 \pm 1.29 \times 10^3$ MP items kg^{-1} in pig, $7.40 \times 10^1 \pm 1.29 \times 10^2$ MP items kg^{-1} in cow, 0 to 5000 MP items kg^{-1} in sheep, and 129.8 ± 82.3 MP items kg^{-1} in chicken manure. Moreover, another study done on microplastic concentrations in livestock manure found over 9000 MPs kg^{-1} (Tan et al., 2022). Composts are also reported as a source of microplastics in agricultural soils. Composts contain organic wastes, but inert materials such as plastic and glass make their way into composts from municipal sources and can therefore potentially make their way into the soils amended composts (Watteau et al., 2018). It is speculated that chemical fertilisers (such as polymer-coated fertilisers) may release microplastics into agricultural soils. Several studies claim that polymer coated fertilisers potentially release microplastics into agricultural soils (Bian et al., 2022; Katsumi et al., 2021) . Microplastics can potentially enter soils through other sources such as littering and road run-off, however, there is currently no data available on the extent of microplastic pollution in agricultural soils or other, from littering or illegal dumping as quantifying this is complex (Bläsing and Amelung, 2018). The improper management and disposal of farm plastic materials may contribute towards microplastics in agricultural soils. In particular, regions where there are no farm plastic recycling facilities, or in countries where recycling schemes have been established, but the service may be inaccessible to farmers due to lack of infrastructure or cost constraints. This could potentially lead to a build-up of agricultural plastic waste (e.g., silage wraps, feed and fertiliser bags, netting and twine) dumped on-site and over-time; weathering can cause this litter to fragment into microplastics that transport into soils. The abrasion of automotive tyres can introduce microplastics into roadsides via dust or wash-off from rain (Wagner and Lambert, 2018). The microplastics shed from road-paints, discarded plastic items in the environment, or vehicle tyres may either run directly into soils through overland flow or make their way into sewer systems, eventually ending up in WWTPs, where they are retained in sludge and later spread on land as a fertiliser (Verschoor et al., 2016).

Microplastics may also enter agricultural soils through atmospheric deposition. Microplastics have been measured in atmospheric deposition in the cities of Paris, France (Dris et al., 2016), Dungguan, China (Cai et al., 2017), Breman, Germany (Bergmann et al., 2019), London, England (Wright et al., 2020), but also in remote locations such as the French Pyrenees Mountains (Allen et al., 2019) and the Arctic Swiss Alps (Bergmann et al., 2019). Although most of the research has been carried out in urban environments and remote locations, it can be said that agricultural soils may be directly affected by this source of microplastic pollution and further studies should be done to address this.

2.7. Effects on soils, plants and biota

Several concerns have been raised about the effects of microplastics on the physical and chemical properties of soil. Studies suggest that microplastics can potentially change soil structure, porosity, water retention, and nutrient cycling, which subsequently may have knock-on effects to microbial communities and plant growth (Wang et al., 2022). As soils serve as the foundation for food production and ecosystem health understanding these effects is critical (McBratney et al., 2014). The physical structure of soil is largely determined by the aggregation of soil particles, which influences porosity, aeration and the movement of fluids, solutes and colloids in soil. Soil structure and formation of aggregates have a major influence on root growth and plant productivity (Nimmo, 2013). Depending on the specific characteristics of microplastics such as shape and size, microplastics have been shown to disrupt soil aggregation. For example, De Souza MacHado et al., (2018) found that polyester (PES) fibres had concentration dependent effects on soil structure including bulk density and water holding capacity, which affected the relationship between microbial activity and water stable aggregates. On the other hand, Lozano and Rillig, (2020) found that soils with microplastic fibres increased the shoot and root mass of a mix of grass and herb species, which was linked with reduced soil bulk density, improved aeration and thus, better penetration of roots in the soil. Another study found that PES microplastic fibres decreased macroaggregation and increased microaggregation of soil colloids (Zhang et al., 2019). Lozano et al., (2021) showed that fibres can hold water for longer in soils, film microplastics can decrease bulk density and foams and fragments can potentially increase soil aeration and microporosity, which in turn promoted better plant performance indicators. The variability in results indicate that microplastic effects on soil depend on microplastic characteristics, and in some cases soil type; however, most of these studies were performed using sandy-loam soils. Most of the findings in the literature demonstrate that the effect of microplastics on soil structural properties depends on both soil type and the specific characteristics of the microplastics such as shape, size and polymer type.

Microplastics in soil has been reported to change various soil chemical properties such as Soil Organic Matter (SOM), and soil pH (Shafea et al., 2022). Microplastics can absorb hydrophobic compounds in soil including Persistent Organic Pollutants (POPs) and bind heavy metals due to their large surface area relative to their volume (Cao et al., 2021; Yu et al., 2021). As a result, additives contained in and bound to microplastics can potentially interact with soil chemistry including Dissolved Organic Carbon (DOC), Olsen-P, Cation Exchange Capacity (CEC) and soil pH. As a consequence, microplastics can potentially cause shifts in microbial communities and disrupt soil nutrient cycling (Salam et al., 2023). Moreover, microplastics can provide surfaces for microbial colonisation and have been shown to alter the microbial community composition of soil (Zhang et al., 2021). In a recent study, soils containing microplastics exhibited reduced microbial diversity and changes in nitrogen cycling, possibly due to the release of additives or pollutants from the plastic particles (Zhang et al., 2023). Changes in microbial communities can affect nutrient availability for plants, as microbes play a crucial role in breaking down organic matter and the release of nutrients. Soil pH is a major factor in determining the binding capacity of minerals and SOC, and therefore plays a huge role in the bioavailability and adsorption of nutrients, and microbial community compositions and activity (Kuśmierz et al., 2023). Publications have shown that microplastics facilitate changes in soil pH, depending on the polymer type (Wang et al., 2022). For example, Boots et al., (2019), reported that soils exposed to microplastics became slightly more acidic over time, which could potentially limit the availability of nutrients like calcium (Ca) and potassium (K) in soils (Laqua, 2015). Alterations in pH can affect plants directly, but mainly influence microbial communities that are sensitive to pH changes. A shift in microbial composition can have broader implications for soil fertility and plant growth, as diverse microbial communities are critical for nutrient cycling and organic matter decomposition. Changes in soil pH can threaten soil microbes by inhibiting enzyme activity and cell metabolism, or indirectly, by limiting nutrient availability (Aralappanavar et al., 2024).

Given the potential of microplastics to alter soil structure and soil chemistry, it is unsurprising that microplastics have been reported to impact plant growth and productivity. Microplastics can potentially affect plant growth by altering the physical properties of soil and by changing root structure, which in turn can influence water and nutrient uptake. Hasan and Jho, (2023) demonstrated that lettuce (*Lactuca sativa* L.) exposed to LDPE, had a dose-dependent effect on the length and weight of shoots and roots, with higher concentrations reducing the growth of shoots and roots. Such changes in root structure can limit access to deeper soil layers, which affect nutrient absorption, ultimately affecting plant growth (Wang et al., 2020). Bosker et al., (2019) found that microplastics significantly reduced the germination rate of cress seeds (*Lepidium sativum*) with a greater effect observed as plastic size increased. Moreover, significant reductions in root growth were observed after 24 hours of exposure, but not after 48 hours or 72 hours. Depending on the type of microplastic, and the dosage/concentration, some studies have shown to negatively affect plant growth by accelerating plant growth or in some cases, no effect is observed (Dong et al., 2022),. For example, a study found that PES fibres (5 mm) at 0.2 % concentration increased the total biomass of *Allium fistulosum*, whereby the dry biomass of bulbs doubled in comparison to the control (De Souza Machado et al., 2018). Judy et al., (2018) investigated the effects of microplastics on wheat seedling emergence and wheat biomass production but found no significant negative effects on plant growth.

Numerous publications have highlighted the risks that microplastics in soil may pose to soil biota. One of the main indicators of soil health is the activity of soil microbes that catalyse biogeochemical transformations (Shafea et al., 2022). Studies have shown that microplastics can alter microbial communities. Bacteria and fungi may interact with microplastics through surface colonisation (Zhang et al., 2021). Zhang et al., (2019) observed soils with high microplastic content showed significant alterations in microbial diversity and activity, leading to reductions in enzyme activities essential for decomposing SOM. The disruption of microbial function can impair nutrient availability for plants, particularly nitrogen (N) and phosphorus (P), essential for growth. Moreover, larger soil dwelling organisms such as earthworms and nematodes may also be affected by the presence of microplastics in agricultural soils. One of the primary pathways for microplastics to affect soil organisms is through ingestion. Most studies carried out on the effects of microplastics on soil biota focus on earthworms. Huerta Lwanga et al., (2016) found that *Lumbricus terrestris* exposed to microplastic-contaminated soil exhibited a reduction in growth and reproduction. Microplastics can accumulate in the digestive system of the animal and cause blockages, which reduces nutrient absorption and can limit energy for growth and reproduction. Cao et al., (2017) found that microplastics were ingested by earthworm species (*Eisena fetisa*), and at higher concentrations (1 and 2 %) significantly reduced growth and induced mortality. Although most research on the effects of microplastics on soil biota have been conducted in laboratory-controlled settings, these findings indicate that microplastics could impair the ability of soil organisms to aerate the soil effectively and contribute to SOM decomposition. More research is required to determine the extent of microplastic accumulation in soil biota on a field-scale level.

2.8. Microplastics in wastewater

A huge quantity of microplastics enter the environment due to mismanaged waste systems, including effluents from commercial establishments and industrial plants, urban surface run-off and domestic wastewater (Barkmann-Metaj et al., 2023; Wang et al., 2020). WWTPs are not designed to remove plastic particles and the number of microplastics entering and exiting domestic WWTPs depends on a wide range of factors such as the infrastructure and treatment technologies, population densities, lifestyle regimes, consumption of MP-based products and specific microplastic characteristics (Ho, 2022; Lv et al., 2019; Schmidt et al., 2020). Microplastics in domestic wastewater come from a variety of sources including the washing of synthetic textiles and apparel, discarded synthetic wet wipes, sanitary products, personal care products (PCPs) such as exfoliants, scrubs and household items like plastic lunch boxes that shed microplastics into dishwasher effluents (Talukdar et al., 2024). Studies have reported that concentrations of microplastics in wastewater influent can vary from 1 to over 100,000 microplastics per litre of wastewater and effluent concentrations of 0 to 447 microplastics per litre of effluent water discharged into freshwater and marine ecosystems (Sun et al., 2019). Depending on the extent of wastewater treatment, some WTTPs are highly effective in capturing the microplastics present in domestic wastewater and preventing their direct entry into aquatic ecosystems. Studies show that WWTPs can remove the majority of microplastics from influent waters to prevent release, and that removal efficacies of up to 95 % have been reported (Talvitie et al., 2017). However, one of the main drawbacks to this is that vast majority of these microplastics are retained in the sludges produced at WTTPs which are subsequently applied directly on-land as a fertiliser, thereby releasing microplastics into the soil environment and eventually to the aquatic environment.

2.9. Microplastics in constructed wetlands

Constructed wetlands (CWs) are engineered systems that mimic natural wetlands to treat wastewater through physical, chemical, and biological processes. CWs are recognised for their cost-effectiveness and environmental benefits in removing pollutants from various wastewater sources including domestic wastewaters, stormwater runoff, industrial, food and agricultural wastewaters, acid mine drainage and landfill leachate (Kadlec et al., 2020). They utilise natural resources such as wetland vegetation, soils and associated microbial assemblages to remove nutrients, suspended solids, organic compounds, heavy metals and pathogenic organisms, protecting downstream waters. The mechanisms by which they remediate contaminants from water include the uptake, immobilisation and transformation of soluble organic materials, nutrients and metals by plants and microbes. In addition to filtration, adsorption and precipitation of soluble chemicals by substrates and plants, and the sedimentation of suspended solids and pathogens and biological degradation of organic pollutants, which occur naturally in CWs. The increasing prevalence of microplastics in wastewater poses new challenges for CWs. Microplastics enter CWs primarily through influent wastewater, and studies have documented the presence of microplastics in CWs, highlighting their ability to retain these particles to varying degrees. Lu et al., (2022) investigated the distribution and retention of microplastics in CWs and found that CWs could effectively capture microplastics, with removal efficiencies influenced by factors such as particle size, shape, and density. Moreover, other research shows that microplastics tend to accumulate more in the sediment and root zones than in the water column. This suggests that the physical structure of CWs, including vegetation and substrate composition, play significant roles in retaining microplastics. Rozman et al. (2023) conducted a study on a horizontal sub-surface flow laboratory CW and observed that microplastics were predominantly retained in the sediment layer, with minimal presence in the effluent, emphasising the effective retention of microplastic within the system. Several factors influence the retention of microplastics in CWs, such as hydraulic conditions, including flow rate and hydraulic retention time, vegetation, substrate type and microbial communities present in the water and solid phase of the CW. The retention of microplastics in CWs may potentially pose ecological risks to the biodiversity that reside in the CW systems.

2.10. Legislation on plastics and microplastics

The EU has implemented some legislation to address plastic pollution, with a focus on both macroplastics and microplastics. As part of the European Green Deal, the Zero Pollution Action Plan aims to reduce pollution in soil, water and to acceptable levels that are no longer considered harmful to health and natural ecosystems. By 2030, the plan has a specific target to reduce the release of microplastics by 30 % (European Commission, 2024). In 2019, the EU directive (2019/904) was set out to combat the reduction of the impact of certain plastic products on the environment. The Single-Use Plastics Directive (SUPD) came into effect in July 2021, banning certain Single-Use Plastics (SUPs) including plastic plates and cutlery, straws, balloon sticks and cotton buds. Moreover, the directive prohibits the manufacturing and usage of all products made of oxodegradable plastic (European Commission, 2019). The Packaging and Packaging Waste Directive (94/62/EC) sets targets to reduce packaging waste and mandates that a significant portion (70 %) of all packaging will be recyclable or reusable by 2030 (European Commission, 2022). Under the REACH regulation, the European commission placed a restriction on intentionally added microplastics, which was adopted in September 2023 to restrict microplastics intentionally, added to products such as PCPs and has been effective since October 2023. It prohibits the sale of microplastics as such and in products that release them during use. Again, in October 2023, the European Commission proposed a regulation to prevent plastic pellet losses from industrial raw materials that are used to make plastic products (European Commission, 2024). These legislation measures highlight the commitment of the European Union to reduce plastic pollution as a whole and make the transition towards a circular economy.

Ireland has implemented several legislative measures to address plastic and microplastic pollution, that for the most part align with EU directives. However, some national initiatives have been introduced. Back in 2002, Ireland's plastic bag levy imposed a charge on SUP bags at the point of sale. This initiative led to a significant reduction in plastic bag usage and has been prominent in changing the behaviour of consumers towards reusable alternatives (S.I. No 605/2001 – Waste Management (Environmental Levy) (Plastic Bag) Regulations, 2001). In 2019, The Microbeads (Prohibition) Act was implemented to ban the manufacture, sale, and import of rinse-off cosmetic and cleaning products containing plastic microbeads in order to prevent microplastic pollution in marine and freshwater environments (EPA, 2019). In July 2021, Ireland enacted the EU Directive on SUPs and transposed it into national law, whereby the sale of specific SUP items, including cotton bud sticks, cutlery, plates, stirrers, balloon sticks, straws, expanded PS cups and food containers were banned. In addition, the sale of oxodegradable plastics were banned which included plastic mulch films made from these materials (EPA, 2024). In 2024, Ireland introduced a Deposit Return Scheme (DRS) to incentivise consumers to the return plastic bottles and aluminium cans. They did this by placing a refundable deposit of €0.15 for containers between 150 ml and 500 ml, and €0.25 for containers over 500 ml up to 3 L. Consumers can return eligible containers to participating retailers or reverse vending machines to reclaim their deposit, with the aim of promoting recycling and reducing discarding beverage packaging in the environment (Citizens Information, 2024).

Outside of the EU and Ireland, on a global scale, numerous countries have enacted legislation to combat plastic and microplastic pollution. Like in the EU and Ireland, most of the focus has been on reducing SUPs, banning microbeads but also through promoting sustainable alternatives. In 2015, the US passed the Microbead-Free Waters Act of 2015, which is a federal law, prohibiting the manufacture and sale of rinse-off cosmetics containing plastic microbeads, which became effective in 2017 for manufacturers and 2018 for the sale of these products (FDA, 2018). Moreover, in the US some states have implemented bans on SUPs. For example, California has enacted legislation to phase out SUPs (including bags and straws) (Governor, 2024). In 2022, Canada announced a ban on the manufacturing and importation of SUPs, which again included bags, straws and take-away cutlery (Environment and Climate Change Canada, 2023). In China, a Plastic Reduction Plan (2021-2025) was put in place in 2020 to cut 30 % of plastic waste within a timeframe of five years. Again, including a ban on SUPs such as plastic bags and straws across major cities (Fürst and Feng, 2022). In Australia, the National Plastics Plan (2021) was introduced to phase out SUPs including cutlery, straws and PS packaging materials by 2025 (Department of Climate Change, Energy, the Environment and Water 2022). In

2021, New Zealand also announced their plans to ban SUPs that are difficult to recycle such as cutlery, food takeaway containers by 2025 (Ministry for the Environment, 2022). Since 2021, other countries such as Japan have already passed bills aiming to reduce microplastic pollution by focusing on PCPs containing microbeads and in South Africa, after the detection of microplastics found in tap water sources, a ban on microbeads has been proposed.

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Chapter 3: Farmers' attitudes towards agricultural plastics - Management and disposal, awareness and perceptions of the environmental impacts

3.1. ABSTRACT

The amount of plastic waste resulting from agricultural practices is increasing and this trend is expected to continue. Although plastics are essential for certain farming tasks, their impact on the environment is becoming a major issue of concern. Mismanaged larger plastics can disintegrate into microplastics and make their way into soils, surface and groundwater sources. Microplastics are extremely persistent and have the potential to facilitate the transfer of contaminants through the environment, potentially affecting terrestrial and aquatic wildlife. A descriptive survey was conducted on a sample of farmers (N = 430) in Ireland to assess their attitudes on agricultural plastic waste management and their awareness and perceptions of the impacts of microplastics and plastics on the environment. This study found that most farmers (88.2%) are concerned about the amount of plastic waste generated by farming activities. Agricultural plastic disposal methods vary, and recycling rates mostly depend on the type of plastic, the cost of recycling and access to facilities. Most farmers view agricultural plastics negatively due to their impact on the environment but also because of the monetary and logistical burdens associated with them. Farmers were relatively aware of microplastics (57.5%), but overall; more farmers felt they knew more about plastic pollution than microplastic pollution and these issues in aquatic systems. This was also evident when it came to their perception of the risk's plastics pose on the environment with more farmers believing that aquatic environments are at greater risk than the terrestrial environments. Future research efforts must focus on plastic and microplastic pollutions in soils to inform policymakers and to create greater public awareness. In addition to this, several developments are needed which should be done in a collective effort by the government, policymakers and other stakeholders to reduce the plastic and microplastic problem in agricultural soils.

3.2. Aims and Objectives

The main aims and objectives of this study were to:

1. Assess the behaviours and attitudes of Irish farmers towards the usage and disposal of agricultural plastics.

2. Evaluate farmers' awareness of microplastics and their perceptions of the overall impacts of plastics on the environment.

3.3. Methodology

A descriptive mixed-method survey design was chosen using a self-report structured questionnaire to achieve the main research goals. Descriptive mixed-method designs are widely used in survey research design to integrate quantitative and qualitative findings to strengthen the outcomes of the study. The classic definition of mixed methods research by Greene et al., (1989), is “those that include at least one quantitative method (designed to collect numbers) and one qualitative method (designed to collect words)”. Mixed methods involve using quantitative data and qualitative data to gather information that can be used in parallel to complement each other (Zohrabi, 2013; Shorten and Smith, 2017). Descriptive studies using questionnaires can help define the opinions, attitudes and behaviours of data subjects on a given topic (Best, 2003). Prior to the administration of the final survey, a pilot-study (N = 18). There is very little information available in social science literature recommending the appropriate sample size for pilot studies. Hill and Hamilton, (1998); Bell, (1982); and Johanson and Brooks, (2010) suggest sample sizes between 10 and 30 for pilots in survey research. Pilot studies are done to increase research quality (Gudmundsdottir and Brock-Utne, 2010) by identifying potential weaknesses in the survey so they can be rectified prior to the implementation of the full study (Malmqvist et al., 2019). The pilot consisted of a questionnaire containing twenty-four questions, which was developed between March and May 2020 and administered through an online platform (Microsoft® Forms® 2016). only farmers took part in the pilot study. For the pilot study, the researcher sought participants through social media platforms and personal networks. After respondent data and feedback was analysed, appropriate amendments were made to the survey. These included the addition of four questions: one on consent, one on the level of agricultural training and education and two on plastic disposal methods. Other minor adjustments were made on the multiple-choice options listed and the layout of some questions. The new amended survey was refined through several rounds of prototyping assisted by the supervisory research team. It contained 28 questions, using Likert-type, single and multiple-choice questions in order to collect quantitative data and open-ended questions for qualitative data.

3.3.1. Participation group

The study group (farmers) was selected with the assumption that they have practical experience with the use of plastics in agriculture because many farming tasks, especially in Ireland, rely heavily on agricultural plastics, no matter what type of enterprise they hold. For this reason, a purposive sampling strategy was adopted. This type of sampling is employed to look for information-rich participants who share certain characteristic(s) related to the research topic (Barglowski, 2018). In order to meet the inclusion criteria of participation, all participants of the study owned or worked full/part-time on a farm in the Republic of Ireland. In addition, all participants had to be over the age of 18 and capable of giving informed consent to take part in the study. In contrast, respondents who did not identify as farm owners or workers and those who were not farming in the Republic of Ireland were excluded from the study. Farmers under the age of 18 or incapable of giving informed consent were also excluded. No groups of farmers from specific farming enterprises alone (i.e., dairy farmers or tillage farmers) or farmers from specific locations of the country were targeted as the aim of the survey was to get a broad representation of the Irish farming community as a whole.

3.3.2. Ethics approval

Before any data collection commenced, the study underwent a thorough institutional ethical review following the procedures of the Declaration of Helsinki by the DkIT ethics committee board. This process involved completing an ethics approval application form outlining a description of the study, main rsearch objectives, the location and duration of the research, participant and sample details. Moreover, justification for the proposed sample size were given and reasons for selecting the study group. Risks posed to participants, data collection distribution modes and any safeguard mechanisms put into place that related to confidentiality were included. Active informed consent was obtained from respondents who participated in the survey and full anonymity to all participants of the study was ensured. All precautions and safeguard mechanisms of data security and storage were/are taken by the researchers.

3.3.3. Survey structure

The survey was designed in five sections. The first section included questions collecting quantitative data on the socio-demographics of participants (age, gender, position on the farm, full-time/part-time, number of years farming, educational level), and questions related to their agricultural production systems (farm type, farm size, farm location). The second section contained questions on the usage and disposal of agricultural plastics, including questions relevant to assessing the current state of agricultural plastic usage in Ireland and what the attitudes of participants were towards these types of plastics. In this section, to collect quantitative data, farmers were asked, “During your time as a farmer, have you noticed an increase in the use of plastics in agriculture?”, where they were given the option to select either “Yes”, “No” or “I don’t know”. This was followed by a Likert-type question “How concerned are you about the amount of plastic used in farming activities?” with “Extremely concerned”, “Concerned”, “Somewhat concerned” and “Not at all concerned” available for choice to participants. To further assess farmers’ attitudes towards the usage of agricultural plastics, the following open-ended question was used for the collection of qualitative data “Please list below two words/phrases you associate with ‘farm plastics’ (either positive or negative)”. Respondents were asked to answer multiple-choice questions (to collect quantitative data) around the disposal of several different agricultural plastics. The first question on the disposal of agricultural plastics was phrased “How do you dispose of the following plastics (silage wrap and sheeting, netting, twine, fertiliser and feed bags) used on the farm?” with the following options presented to participants “Pay for disposal”, “Pay for recycle”, “On-farm disposal”, “Reuse on farm” or “Other”. Following this, respondents were asked to elaborate further on their mode of agricultural plastic waste disposal in which qualitative data was collected using open-ended questions, “If other, please specify in the space below” and “If plastics are disposed of on the farm, which method is used?”. A series of questions were included to capture the reasons behind farmers not recycling their agricultural plastic waste. Again, both quantitative and qualitative data were collected here. A multiple-choice question to collect the former, “In relation to the plastics that you do not recycle, what are your reasons for not recycling these?” The following choices were available to respondents “Too expensive”, “Don’t know how to”, “Lack of facilities”, “Contamination (not accepted)”, “Not enough generated to recycle” and “Other”. Respondents were given the option to provide additional information qualitatively “If other, please specify in the space below”. Following this, respondents were asked if they “Agree”, “Disagree” or are “Unsure” with the statement “Recycling farm plastics is convenient”. Next, to test the knowledge that farmers have on the recyclability of agricultural plastics, the following statement was presented “All farm plastics are recyclable” to which they had the option to respond to it with “Agree”, “Disagree” or “Unsure”. The final question in this section was a similar style to the previous. Respondents were asked if they “Agree”, “Disagree” or are “Unsure” about the statement “The disposal of farm plastics is a big environmental problem”. The third section of the survey consisted of questions on biodegradable agricultural plastic use in

Ireland, however, the questions were later omitted and not written up for the final study. The questions included in the fourth section aimed to verify the farmers' awareness of microplastics and plastic pollution in both aquatic and terrestrial systems. This was measured quantitatively using a multiple-choice question "Prior to this survey, had you heard of the term 'microplastic'" with the options "Yes", "No" or "I don't know" available to participants. A Likert-type question was presented to participants "Please indicate to what extent you are aware of the following: "Plastic pollution in the oceans", "Plastic pollution on land (incl. farmlands)", "Microplastic pollution in the oceans" and "Microplastic pollution on land (incl. farmlands)". Respondents had a choice to select either "Very aware", "Aware", "Somewhat aware" or "Not aware" for each of the categories. The final section of the survey sought to determine the perceptions of farmers on the impact of plastics on the environment. Respondents were asked "How serious of a threat do you think plastic pollution poses for each of the following: "Oceans", "Freshwaters (e.g., lakes and rivers)", "Land (incl. farmlands)", "Soils (incl. agricultural soils)", "Marine and freshwater wildlife (e.g., fish, seabirds, etc.,)", "Farm animals", "Soil animals (e.g. earthworms etc.,)", "Wild plants", "Crops" and "Humans". Again, a Likert-type question was included to collect quantitative data, which presented the following choices available to participants: "Very serious", "Serious", "Somewhat serious" and "Not at all serious".

3.3.4. Sample size

The minimum sample size of participation for the final administered questionnaire was 384 observations based on Cochran's sample size formula for categorical data, which was previously employed by Bartlett et al., (2001). This considers a 95 % confidence level and a standard level of precision at 0.5. According to figures provided by the Central Statistics Office of Ireland, the sample population (Irish farmers) was 278,600 during the time the survey was developed (CSO, 2022)

3.3.5. Data collection

The collection of survey responses was carried out between July and October 2020. The survey was disseminated through an online platform Microsoft® Forms® 2016. The link to the questionnaire was distributed via email through a variety of agricultural networks. In addition to this, a link to the survey was shared on social media platforms and with farming print media in Ireland. Hard copies of the survey were made available in order to cater for farmers who were inexperienced with digital platforms or/and those with limited or no online access.

3.3.6. Data analyses

Data was cleansed using Microsoft® Excel® 2016. Initially, data were cleansed to remove any replicate responses or responses received from outside of the Republic of Ireland. All unintelligible open-ended answers were removed from the data set to ensure data quality. Post-data cleansing, 430 surveys remained for analysis. All quantitative analysis was done using Minitab® 20.3. Open-ended answers were analysed qualitatively using inductive content analysis (Vaismoradi et al., 2013) using Microsoft® Excel® 2016. This was done to investigate the patterns of words and phrases in order to formulate concepts and themes to answer the main research goals of the study. A wordcloud was generated on Wordart.com. Wordclouds are used as a way to display text data in a graphical form and typically visually represent word frequency (Atenstaedt, 2012; DePaolo and Wilkinson, 2014). Words listed by at least two respondents were only included for this analysis (Yeganeh et al., 2020). A top-down approach was used to group words into one of the four categories (negative connotations; positive connotations; neutral; or ambiguous) (Dilkes-Hoffman et al., 2019a). The word cloud shows the negative words associated with agricultural plastics in red, positive words in blue and neutral/ambiguous words in green. The size of the words reflects the frequency of occurrence in the data. The positive and negative words collected were later analysed using a bottom-up approach to identify additional groupings. Pearson's chi-squared tests for independence were used to determine whether there were statistically significant differences between the expected and the observed frequencies in categorical variables.

3.4. Results and Discussion

In total, 430 survey responses were taken into account for the final analyses of the study. However, although the majority of respondents completed the full survey, some did not answer all questions but were included in most of the analyses presented here.

3.4.1. Respondent demographics and agricultural production systems characteristics

The majority of respondents were male (82.9%) and aged between 25-39 years of age (33.6%) (Table 3.1). The high percentage of male to females involved in the farm labour workforce are in line with what was expected. Eurostat figures on the breakdown of farmers in the EU showed that on average, women accounted for 35.1% of the agricultural workforce (Eurostat, 2017). The Census of Agriculture 2020 reports that 26.9% of the Irish agriculture labour work force are female (CSO, 2022a). A higher number of respondents who took the survey were under 40 years of age, however, most (55.3%) of the farmers in Ireland are 55 years of age or more and only 5.3% represent the under 35 years category (CSO, 2018).

The majority of responses came from the Border (33.7%) region of Ireland, followed by Midland (21.7%), Mid-East (12.9%), Western (10.8%), Mid-West (7.3%), South-East (6.8%) and South-West (6.6%). Responses were received from all the 26 counties of the Republic of Ireland. Most respondents identified as the farm owner/manager (80.94%), with the rest of respondents identifying as farm workers (19.06%). The number of respondents who reported as working either full-time or part-time was 44.82% and 44.18% respectively, with the highest proportion reporting that they have been working in farming for more than 20 years (46.1%). The top three main farming enterprises were beef production (39.5%) followed by dairy (24.5%) and mixed grazing livestock (14.2%). However, in Ireland, beef production systems represent 56.4% of the farm types, followed by dairy and sheep (CSO, 2022b). Respondents reported they owned or worked on farms between 26-50 ha in size the most (29.5%), followed by farms of 51-75 ha (23.1%). The average farm size in Ireland is approximately 32.4 ha (CSO, 2022b). The majority of respondents reported they had completed at most a level 5/6 (agricultural cert/green cert) (38.35%), which followed by level 8+ (honours degree or higher) (21.41%), and 16.94% received no formal agricultural training.

Table 3.1: Respondent demographics and characteristics of their agricultural production systems.

Item	No.	Percentage (%)
Gender (N = 387)		
Male	321	82.95
Female	66	17.05
Age range (N =360)		
18-24	72	20
25-39	121	33.61
40-49	71	19.72
50-59	56	15.56
60-69	33	9.17
70-79	7	1.94
80+	0	0
Region of Ireland (N = 424)		
Border	142	33.71
Midland	92	21.41
Western	46	10.85
Mid-East	55	7.31
Mid-West	31	6.86
South-East	29	6.85
South-West	28	6.61
Position on farm (N = 404)	327	80.94
Farm owner/manager	77	19.06
Farm worker		
Working (N = 415)		
Full-time	186	44.82
Part-time	229	55.18

Duration farming (N = 424)		
0-5 years	40	9.46
5-10 years	75	17.73
10-15 years	64	15.53
15-20 years	59	11.58
20+ years	195	46.1

Main farming enterprise (N = 427)		
Beef production		
Dairy	169	39.58
Mixed crops livestock	105	24.59
Mixed grazing livestock	22	5.15
Sheep	61	14.29
Tillage	44	10.3
Other	20	4.68
	6	1.41

3.4.2. Attitudes towards plastic usage and disposal methods of agricultural plastics in the Irish agricultural sector

3.4.2.1. Agricultural plastic use is increasing and most farmers are concerned

Respondents were asked if, during their time as a farmer, had they noticed an increase in the use of farm plastics, to which 79.6% reported 'Yes' they have, 14.3% answered 'No', leaving 6.1% who did not know (Figure 3.1a). When considering that more than half of the respondents have been farming for over twenty years, it is no surprise that almost 80% of respondents reported to seeing an increase in the use of plastics in agriculture. Plastics have been used for agricultural practices since the 1950s, and over-time agriculture has become increasingly intensive, resulting in agricultural plastics becoming more available to farmers (Robinson and Sutherland, 2002). While there is no regional data available on the coverage or quantity of agricultural plastic in Ireland, in the European Union around 1.7 million tonnes of agricultural plastics were used in 2018, which is between 3 and 4% of the total converter demand of European plastic usage. Global forecasts predict that plastics used for greenhouses, mulches and silage films are set to increase by 50% from 6.1 million tonnes in 2018 to 9.5 million tonnes in 2030 (FAO, 2021). In this study, the results showed that 88.3% of respondents expressed some level of concern about the amount of plastics used for farming activities, with 15.6% extremely concerned and 11.8% who are not at all concerned (Figure 3.1b). A decline in concern is evident among younger cohorts, with only 9.8% of farmers aged under 40 stating they are extremely concerned, in comparison to 25% of farmers over the age of 50 years. This result ties in well with a recent study carried out by the Irish Environmental Protection Agency (EPA) on the attitudes and behaviours towards single-use plastics in Ireland. Over half of the sample population stated they were 'very concerned', and only 5% stated that they are not at all concerned with the amount of plastic used as a society. Their results also show that age may impact the attitudes of society in relation to amount of plastic used, with more adults over the age of 65+ feeling more concerned about it (EPA, 2022). In this study, the region ($p = 0.035$) and size ($p = 0.006$) of the farm respondents own or work on was seen to make a statistically significant difference in how concerned they were about the amount of plastics used for farming activities. For example, farmers owning or working on smaller sized farms were more concerned about the amount of plastics used in farming activities, compared to those on bigger farms. In relation to the region, the farmers were located; farmers in the Border and Western regions of Ireland were more concerned about the amount of plastics used in agricultural activities. This makes sense because typically farms on the West of the country are smaller than farms in the Midland and Eastern regions.

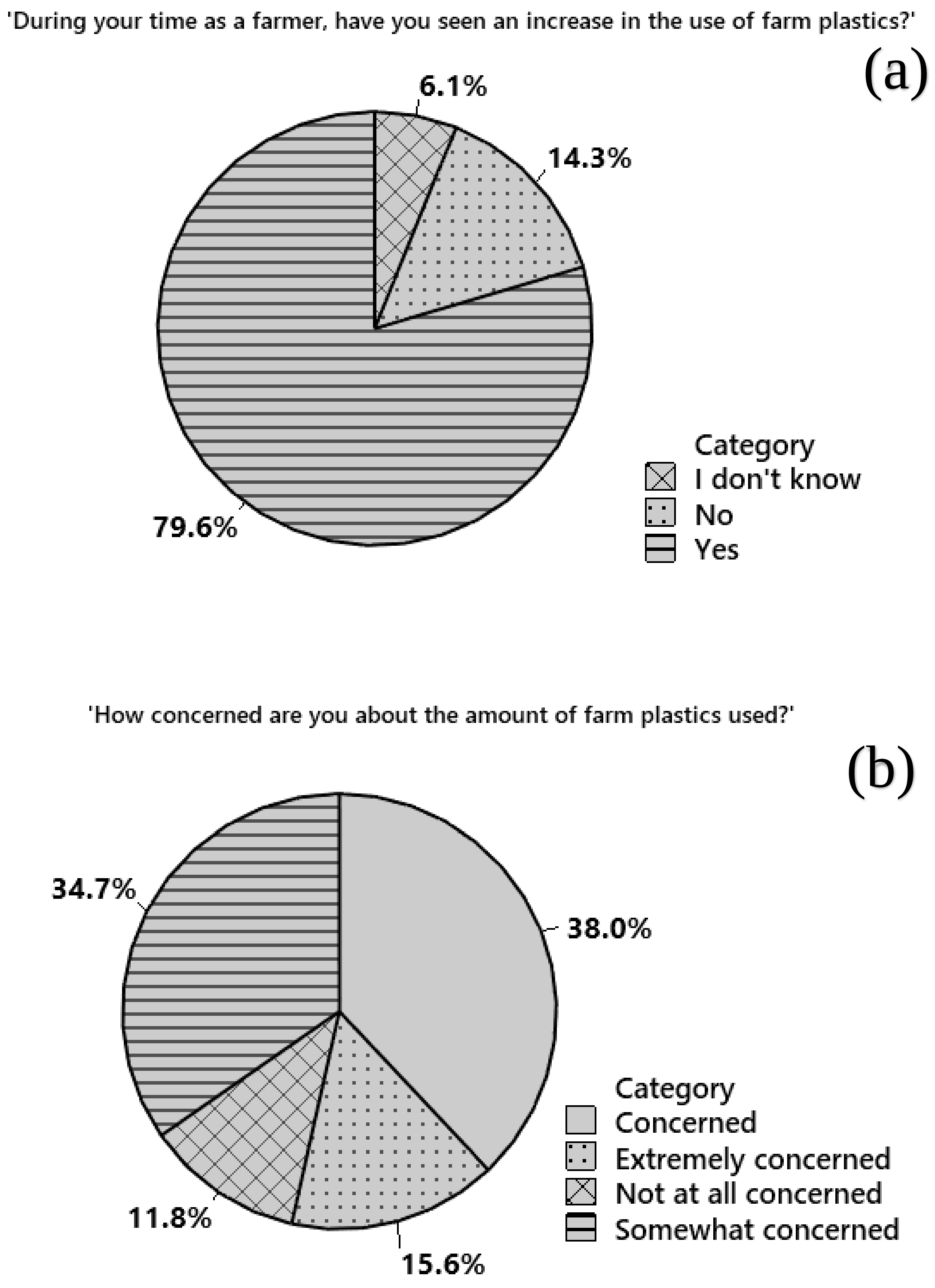


Figure 3.1: The total percentage of farmers noticing an increase in the use of farm plastics since they began farming (a). Respondents concern about the amount of plastics used in agriculture (b).

3.4.2.2. Agricultural plastics are necessary for farm tasks, but are bad for the environment and have monetary and labour costs

Respondents were asked to 'Please list two words/phrases you associate with the words: farm plastics (either positive or negative)'. In total, 349 responses, which amounted to the collection of 500 words, were counted, analysed and presented as a Wordcloud (Figure 3.2). Responses mostly fell into the negative connotations category (65.8%), which is followed with 23.4% of responses falling into the positive connotations category. The words considered ambiguous and neutral consisted of 7% and 3.8% of the responses respectively (Table 3.2). Additional inductive content analysis was carried out to identify concepts and themes within the negative and positive connotations categories. In relation to the negative words collected, responses were mostly related to the environment (147), followed by general negative associations (95), words related to time and labour constraints (36), cost (32) and accessibility (19) (Table 3.3). In the positive connotations category, most positive words provided were related to the material properties and function of agricultural plastics (52), followed by general positive associations (44), words related to the environment (17) and accessibility (4) (Table 3.4). The majority (67.6%) of respondents listed only negative words, followed by 15.3% who chose only positive words, and 14.4% who included both one positive and one negative word in their answers. The remainder of respondents chose only neutral or ambiguous words for their response (Table 3.5). Perhaps, as expected, some respondents (14.8%) only negatively associated agricultural plastics with the cost factors, including monetary, time and labour constraints. Some participants stated 'Costly. Difficult to manage when it piles up in springtime, dirty, messy wet.' (P 187), 'The cost of disposal and the amount of room it takes up after being used' (P 214), and 'Expensive to buy and expensive to dispose of' (P 235). However, more farmers (29.4%) only associated agricultural plastics with the negative impact they have on the environment. Some participants included more general comments such as 'Bad for the environment' (P 19), 'Environmental disaster' (P 125) and 'Long life in the environment' (P 118), but some were more specific. One participant responded with 'Eyesore and environmental issue as it blows across fields and see it on the roads' (P 61), another with 'Blowing everywhere' (P 178), and 'Blowing in the wind' (P 176). Others stated 'Harmful to environment when not disposed of in the correct manner, ending up in seabeds etc.' (P 72) and 'Visible in every ditch in the country (and) will be an issue in the food chain in years to come' (P 124). Moreover, other results show there is a high level of agreement among respondents on the disposal of agricultural plastics presenting a big environmental problem, with 82% of respondents agreeing with this statement. Age ($p = 0.041$) was seen to have a statistically significant effect on farmers' perception of agricultural plastics presenting a big environmental problem, with younger farmers more in agreement than older farmers do. While an overwhelming number of respondents solely focused on their negative associations with agricultural plastics, many respondents expressed conflicted attitudes. For example, 14.4% of farmers responded with both one positive and one negative association. In some of these cases, many farmers believe that agricultural plastics are negative due to the cost factors associated with them and their impacts on the environment. Yet they also acknowledge that plastics are positive and a necessity due to their functionality on the farm. Seven participants commented that agricultural plastics are a 'Necessary evil'. One participant responded with '(A) necessary evil, (but) what else would I cover the silage pit with?' (P 430) and another with 'Good for ensiling grass or maize, however in some cases is not disposed of in an environmentally friendly manner, leading to pollution' (P 107).

Figure 3.2: Most commonly listed words respondents associated with the words 'Farm plastics'. The font size of the words reflects the frequency of responses.

Table 3.2: Responses to 'Please list two words/phrases you associate with the words farm plastic'.

Main category	**Count**	**Percentage (%)**
Negative connotations	329	65.8
Positive connotations	117	23.4
Neutral connotations	19	3.8
Ambiguous connotations	35	7
Total	500	100

Table 3.3: Content relating to the negative connotations listed.

Content (Negative connotations)	**Count**
Related to the environment (e.g., pollution, waste, non-recyclable)	147
Related to cost (e.g., expensive, extra-cost)	32
Related to time/labour constraints (e.g., extra-work, time-consuming)	36
Related to accessibility (e.g., excessive, no-alternative)	19
General negative association (e.g., dangerous, evil, difficult)	95
Total	329

Table 3.4: Content related to the positive connotations listed.

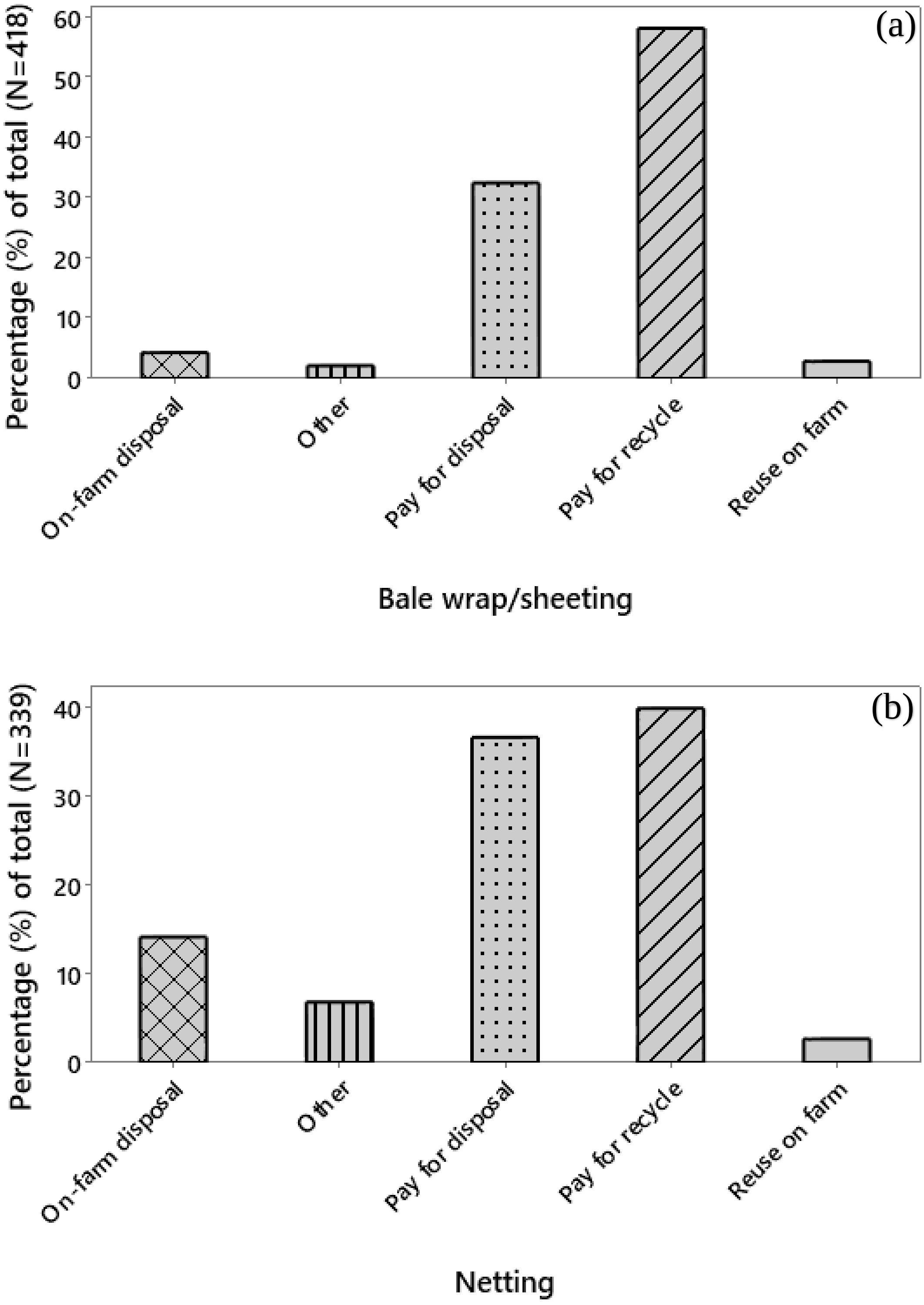
Additional category (Positive connotations)	**Count**
Related to the environment (e.g., recyclable, reusable)	17
Related to material properties and function (e.g., clean, durable)	52
Related to accessibility (e.g., available)	4
General positive associations (e.g., convenient, important)	44
Total	117

Table 3.5: The breakdown of the number of times only negative or positive words were listed; both one negative and one positive word were listed together; and neutral/ambiguous listings.

Type of response	Count	Percentage (%)
Only negative word(s) listed	237	67.6
Only positive word(s) listed	53	15.3
Both one negative and one positive word listed	50	14.4
Neutral/ambiguous word(s)	9	2.7

3.4.2.3. Agricultural plastic disposal methods depend on the type of plastic, cost and access to facilities

Disposal methods vary by the type and composition of the plastic with some being easier to dispose of. Of all the types of agricultural plastics, bale wrap is mostly recycled by participants (58.1%) (Figure 3.3a). Education ($p = 0.038$) was seen to make a statistically significant effect on how farmers dispose of bale wrap. A higher level of education tends to lead to a higher level of recycling and reusing. Other agricultural plastics widely sent for recycling include netting (Figure 3.3b) (41.4%) and fertiliser and feed bags (38.5%) (Figure 3.3d). The data shows that twine may be considered the most difficult to recycle, with only 20.1% recycling twine, however, more farmers (41.8%) find alternative uses for twine on the farm (Figure 3.3c). Some expressed that they find their own means of disposal on the farm, or they use ‘other’ methods, which was mostly the case for netting (20.9%) and twine (16.2%). This may be because there are minimum weight requirements for acceptance of agricultural plastics at the national farm plastics recycling compliance scheme. The farmer is charged per half tonne for each type of agricultural plastic they choose to recycle, and as netting and twine are typically lightweight plastics, generating enough waste may be challenging on different farms.


(a)
Percentage (%) of total (N=418)
0
10
20
30
40
50
60
On-farm disposal
Other
Pay for disposal
Pay for recycle
Reuse on farm
Bale wrap/sheeting
(b)
Percentage (%) of total (N=339)
0
10
20
30
40
On-farm disposal
Other
Pay for disposal
Pay for recycle
Reuse on farm
Netting

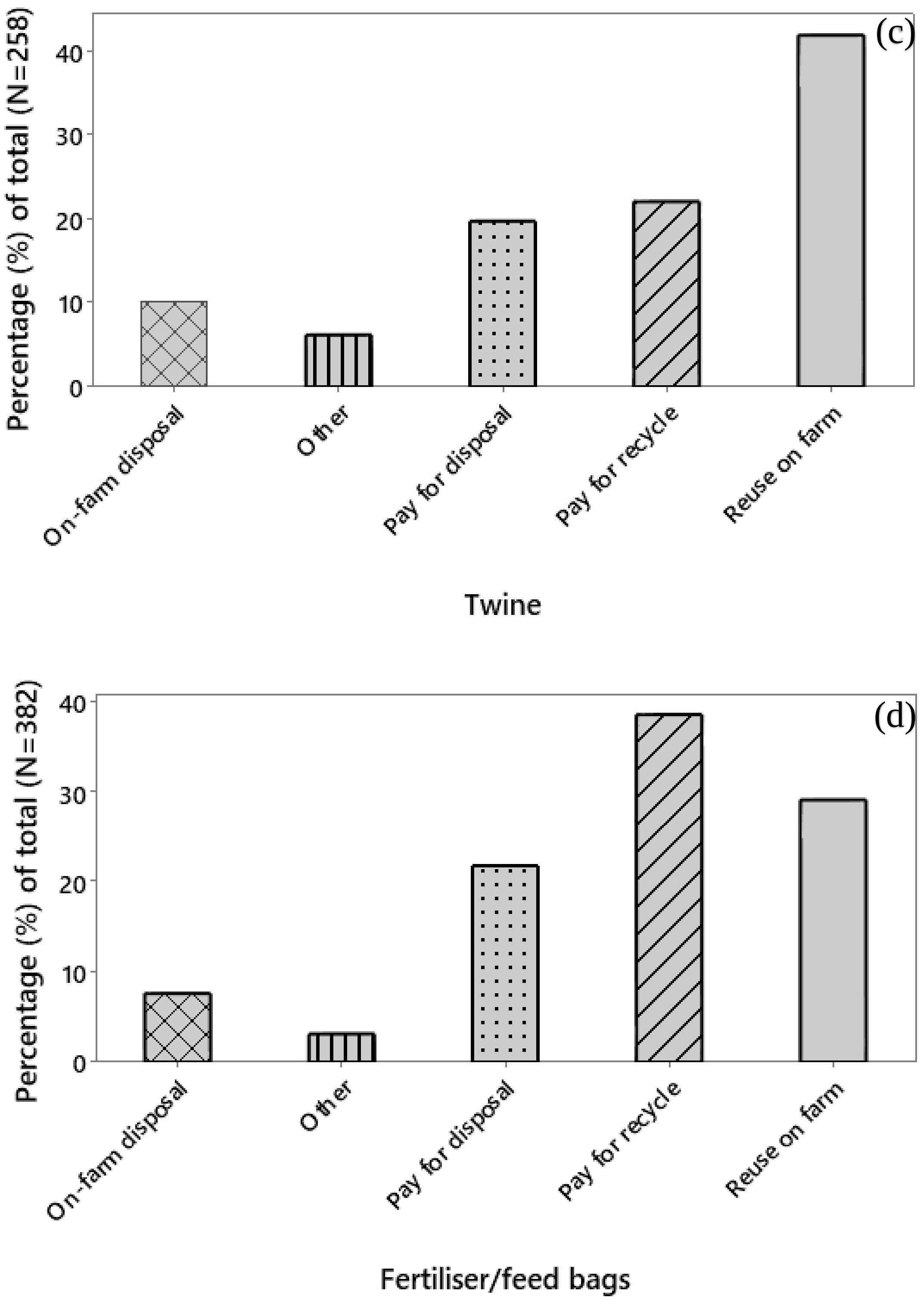


Figure 3.3: Farm plastic disposal methods for each type of plastic: (a) bale wrap/sheeting, (b) netting, (c) twine and (d) fertiliser/feed bags.

Under current Irish legislation, (S.I. No. 396/2017 - Waste Management (Farm Plastics) (Amendment) Regulations 2017) farmers have an obligation to recycle the farm plastic material waste they generate. However, when respondents were asked to elaborate on what types of 'other' or 'on-farm' methods were used, 14% (N = 63) (Figure 3.4) volunteered information stating they either burn or bury certain agricultural plastic waste. One respondent stated they 'Recycle wraps and sheeting, (but) pit burn twine and net' (P 307), and another said, 'Netting is the only thing disposed of on the farm, it is burned' (P 43). Burning plastic in open field is illegal under the Waste Management (Prohibition of Waste Disposal by Burning) (Regulations 2009), as it releases toxic gases into the environment, including substances such as dioxins, and furans. Other by-products of burnt plastic (soot and ash) can cause health and environmental impacts through the release of volatile organic compounds, particulate matter, particulate bond heavy metals, and PAHs, which travel depending on atmospheric conditions, settling on crops in neighbouring fields and entering waterways, potentially making their way into the food we eat (Verma et al., 2016). Moreover, while some farmers understand that doing this is 'wrong', disposal methods need to be convenient and cost-effective in order to motivate farmers to manage agricultural plastic waste effectively in an environmentally sound manner.

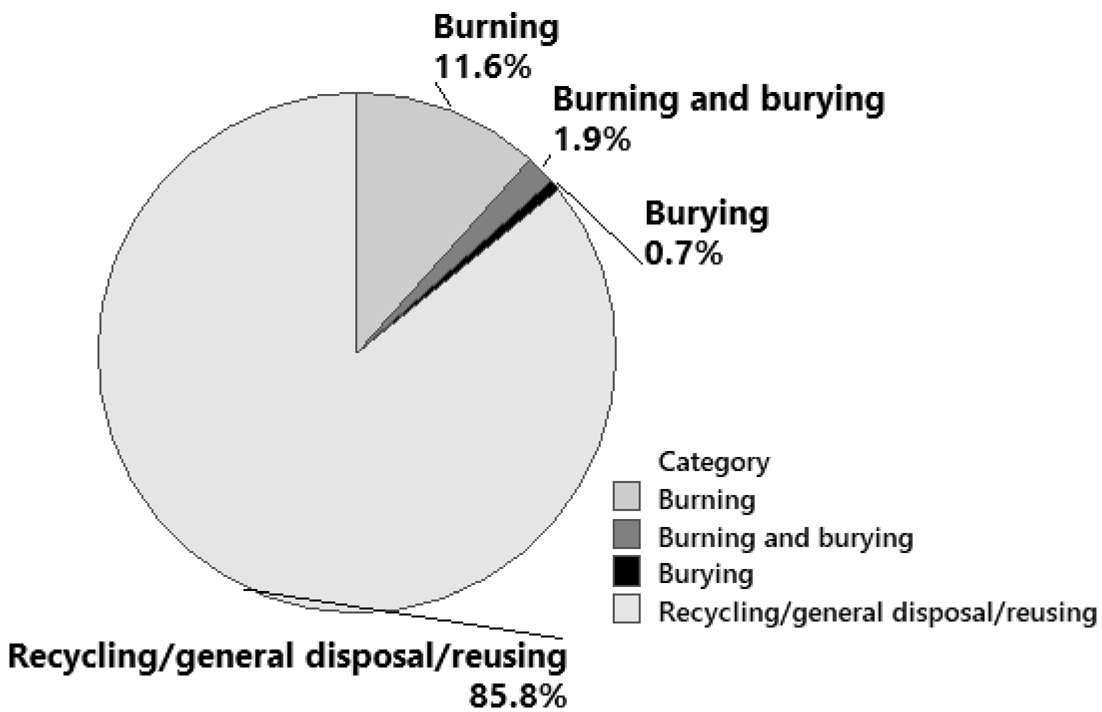
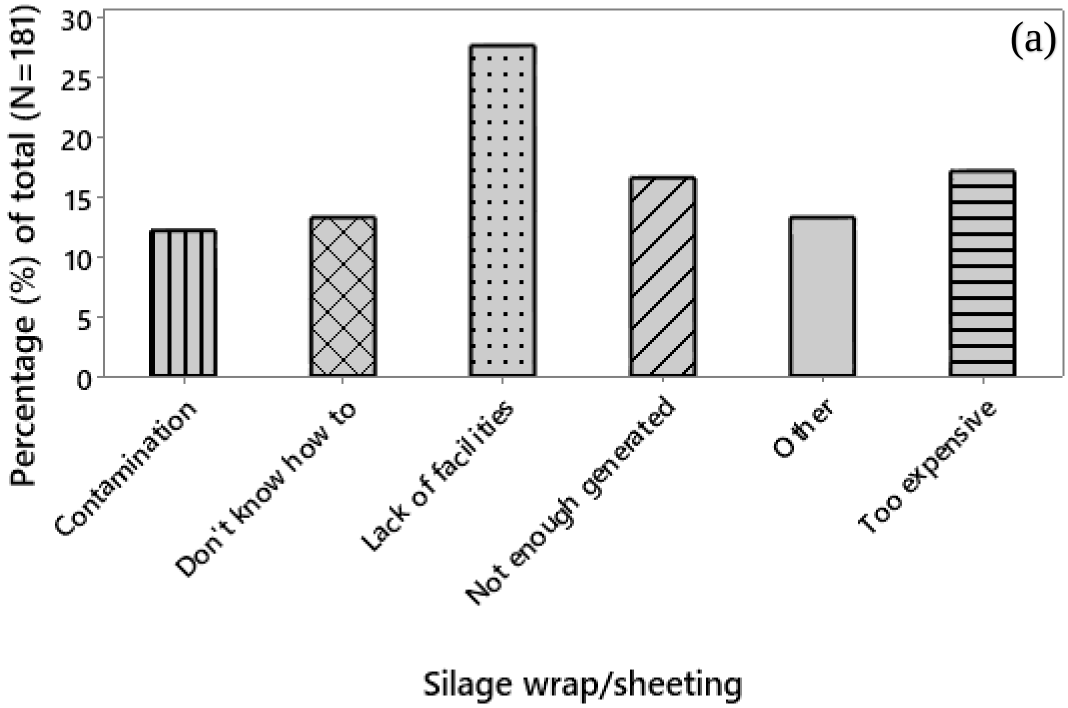


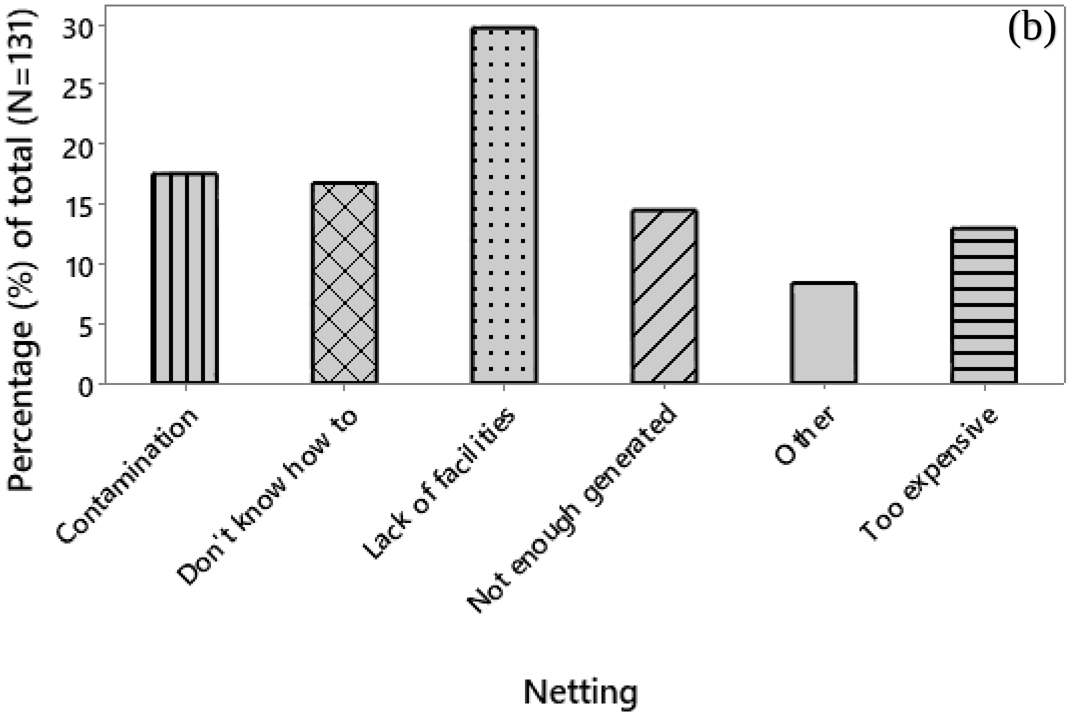
Figure 3.4: The percentage of farmers burning and burying certain farm plastic types.

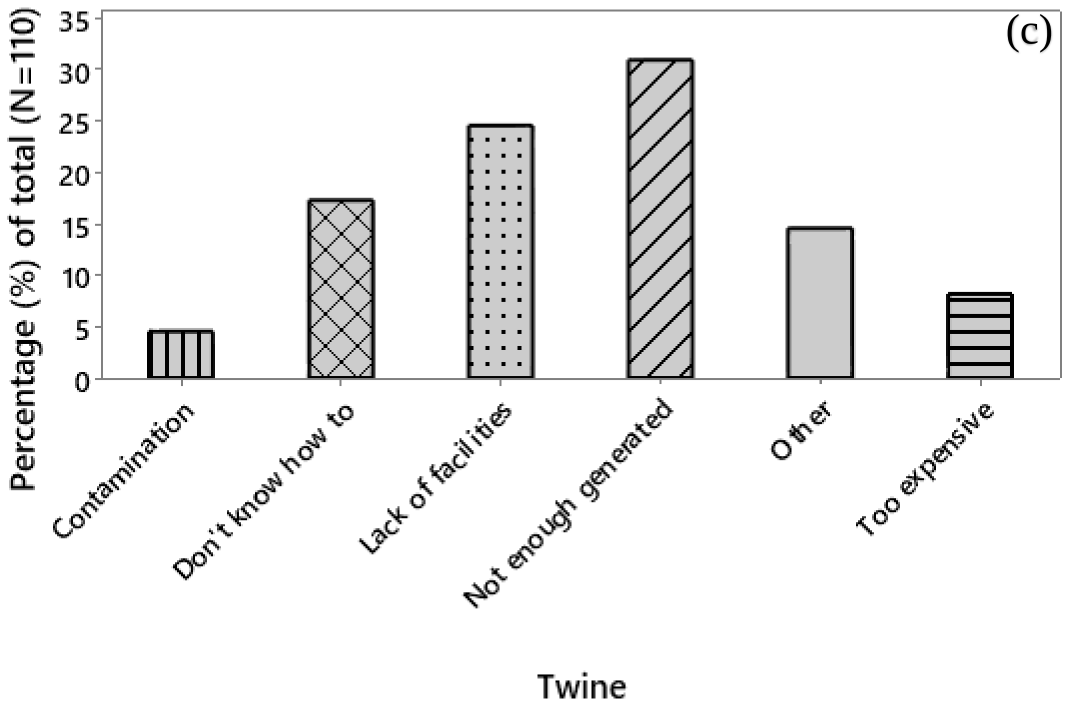
There seems to be some misconception among some farmers about the recyclability of certain agricultural plastics. One participant responded that they 'Burn or bury netting and fertiliser bags as they can't be recycled' (P 376). The national agricultural plastics recycling compliance scheme allows for the recycling of silage bale wrap and sheeting, netting, twine, fertiliser and feed bags, drums and containers. Other plastics used on the farm such as plastic mulch films and piping are not accepted for recycling. In a separate question, results showed that 41% of farmers agree that 'All farm plastics are recyclable', but 22% disagree and 37% are unsure whether they are. Age ($p = 0.040$) made a statistically significant difference on whether farmers think that all farm plastics are recyclable. For example, more farmers older than fifty years of age think that all agricultural plastics are recyclable in comparison to farmers under the age of 40. However, a higher number of younger farmers are unsure if they are all recyclable. These results provide evidence that there are gaps in the level of knowledge some farmers have around the recyclability of agricultural plastics in Ireland, which present several implications. First, if farmers are unaware on how to recycle agricultural plastics, it may discourage them trying which may ultimately lead to them choosing a different method of disposal, for example burning or burying on the farm. Second, not all agricultural plastics are recycled in the same way. There is a different method for recycling silage plastics in comparison to recycling fertiliser and feed bags. Prior to the recycling of agricultural plastics, there are certain separation criteria that must be adhered to and if farmers are unaware of this, they may be storing and separating incorrectly, which can cause problems down the line for the collectors and recyclers of agricultural plastics. For example, if a person brought all their agricultural plastic wastes mixed together to the recycling depot, this would slow down the process and/or those plastics may potentially become unfit for recycling and thus not accepted. Farmers incorrectly storing and separating plastics may be charged extra at recycling depots, which may discourage them from recycling agricultural plastics thereafter. Due to the hydroscopic nature of certain agricultural plastics, they can retain water, but also, the plastics may be covered in bits of dust, grit and soil, which will result in a heavier weight. To ensure extra charges are not applied, farmers must be aware of the consequences of poor storage conditions.

It may be of benefit if the media (such as national/local farming newspapers and radio stations) publish or announce notices regularly on how to store and separate agricultural plastics correctly for recycling all throughout the year. In addition to this, it may be of benefit if notices are displayed in local marts, co-ops, and shops to encourage and educate farmers on how to recycle agricultural plastic waste. It is important to consider that certain groups of farmers may use different sources of information. Läpple, (2013) found that organic farmers used advisory services more often than conventional farmers, but no differences were found between the two groups in terms of the use of media information. Another strategy to help improve farmers' knowledge and confidence on the recycling of agricultural plastics may be through education and training surrounding agricultural plastic waste stream management and recycling. It may be of benefit if agricultural courses include content on how to manage and recycle agricultural plastics. Moreover, content on the implications of poor agricultural plastic management, such as plastic and microplastic pollution and their potential impacts in agro ecosystems should also be added to these courses to help improve the current state of knowledge on these topics.

A follow up section was included to try to understand why agricultural plastic waste is not being recycled by some farmers. The most commonly stated primary reasons farmers do not recycle agricultural plastics are due to the following factors: (1) a perception of a lack of facilities available to them, (2) they feel they do not generate enough agricultural plastic waste to recycle at the standard cost, and (3) they do not know how to recycle agricultural plastics. As expected, many farmers (30.9%) reported twine as the most difficult type of agricultural plastic to generate an adequate amount of waste for acceptance at recycling facilities. Moreover, to a lesser extent, contamination issues were also among the reasons reported as to why farmers do not recycle agricultural plastics, with bale wrap and netting considered the most difficult to keep free from contamination (Figure 3.5: a, b). Age ($p = 0.030$) has a statistically significant effect on why farmers do not recycle agricultural plastics, with older farmers believing there are a lack of recycling facilities available to them, however, younger farmers perceive the cost of recycling to be the main barrier. Typically, in Ireland, in any given jurisdiction of the country, which would serve up to thousands of farmers, there are only a set number of days available each year where farmers can recycle their agricultural plastic waste. Due to transportation and monetary costs, farmers may be less inclined to travel to the recycling depots to avail of the services. Forty one percent of farmers reported that recycling agricultural plastics is inconvenient for them, while 46% think it is convenient and 13% were unsure. The size of the farm ($p = 0.011$) and the region ($p = 0.021$) farmers worked had a statistically significant difference on their attitude towards the convenience of recycling agricultural plastics. Farmers occupying larger farms find it more convenient to recycle agricultural plastics than those who own or work on smaller sized farms. Again, this may be due to the minimum weight cap placed on the amount of plastic that is accepted at recycling centres. Larger sized farms equal more production, therefore generating enough plastic waste to recycle on bigger farms may not be an issue in comparison to smaller farmers that generate less plastic waste. With regards to location, farmers in the Western regions of the country stated that they find recycling agricultural plastics less convenient than farmers in the Midlands and Eastern regions. The size of farms in the Midlands and in the East of Ireland are on a much bigger scale than farms on the West which supports the idea that it is more difficult for farmers working on smaller holdings to recycle agricultural plastics. Another factor, which may affect the convenience of recycling agricultural plastics in the West, is potentially due to poorer roads and public infrastructure in this region of Ireland.


(a)
Percentage (%) of total (N=181)
30
25
20
15
10
5
0
Contamination
Don't know how to
Lack of facilities
Not enough generated
Other
Too expensive
Silage wrap/sheeting


(b)
Percentage (%) of total (N=131)
30
25
20
15
10
5
0
Contamination
Don't know how to
Lack of facilities
Not enough generated
Other
Too expensive
Netting



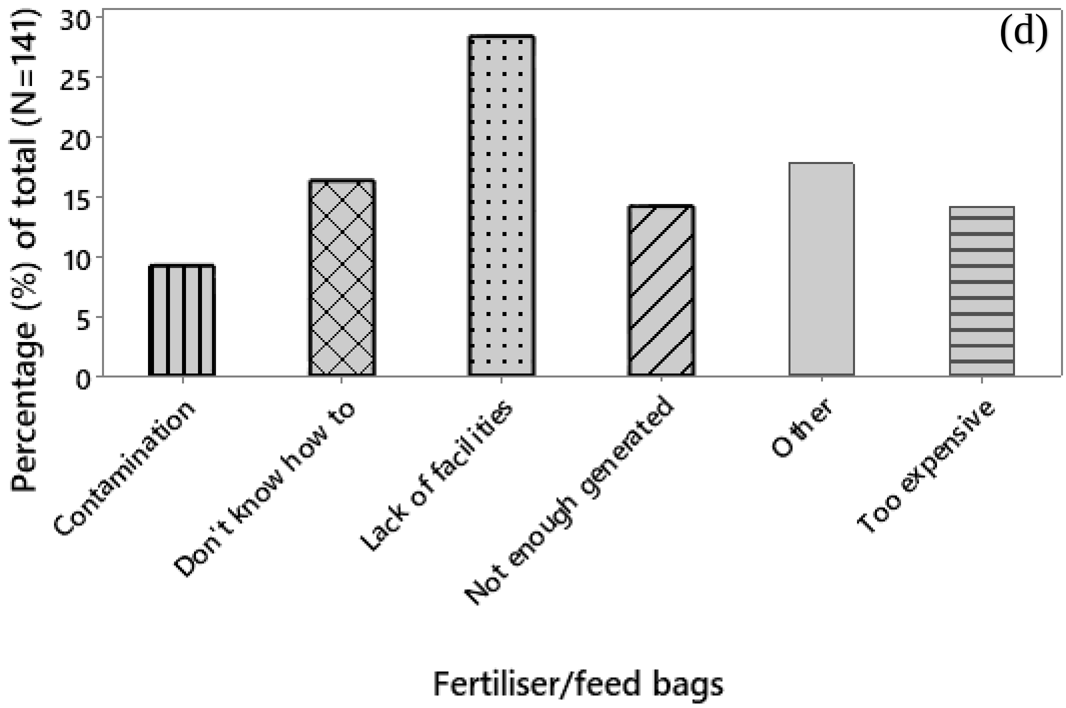


Figure 3.5: Reasons farmers are not recycling farm plastics: (a) bale wrap/sheeting, (b) netting, (c) twine and (d) fertiliser/feed bags.

3.4.3. Farmers' awareness of plastic and microplastic pollution and their perceptions of the environmental impacts

Many farmers (57.7%) reported they had previously heard of the term 'microplastic' prior to taking the survey. As expected, education had a statistically significant difference on the awareness of farmers towards microplastics ($p = 0.016$). In addition, while most respondents are, at some level aware of plastic and microplastic pollution in the oceans and on land, there is greater awareness of plastic pollution and less awareness of microplastic pollution (Figure 3.6.). This is no surprise because macroplastics are visible and microplastics are mostly 'invisible'. Furthermore, research articles and media reports on plastic pollution started earlier than those on microplastics. Interestingly, respondents reported that they are more aware of plastic and microplastic pollution issues in the ocean than on land. Although farmers are out working on the land every day, they may be unaware of the issues on land because again, most research articles and media attention has been mainly focused on the aquatic environments (Jenkins et al., 2022). Increased emphasis must be placed on the occurrence and impacts of microplastics on-land and in soils by the media in order to strengthen the publicity and education of relevant knowledge of microplastics in these terrestrial systems; however, this is only possible if adequate scientific research has been done to inform the media. It can thus be suggested that there is a need to increase the number of research studies on the impacts of microplastics and plastic pollution on terrestrial systems.

As farmers are more aware of plastic and microplastic pollution in aquatic environments, this seems to influence their risk perception. Overall, farmers think that plastic pollution threatens aquatic environments more than the terrestrial systems. For example, more respondents perceive plastic pollution as a bigger threat to the oceans, freshwaters, and marine and freshwater animals in comparison to soil, soil animals, crops and wild plants. The majority of respondents (over 80% for every category) do think that the threats plastics pose is to some extent serious in all of the ten environmental compartments presented (Figure 3.7.**)**. However, over half of the respondents perceive these risks are *very serious* in oceans and freshwaters, compared to less than a fifth who interpret the same level of risk towards components of the terrestrial systems such as soil, wild plants and crops. These results relate to findings by Deng et al., (2020) who found that 43.8% of respondents believed that microplastics mostly accumulate in the oceans, followed by animals and plants (14.2%), air (13.5%), rivers and lakes (6.3%) and soil (4%). Filho et al., (2021) also found that most (60%) of their survey respondents considered the problems associated with plastic in the ocean as *extremely serious*, and less than 40% of respondents reported the effects of plastics on soils as *extremely serious*, and even less (20%) consider the effects of plastics on air as *serious*. Another interesting finding to our study is that over 70% of respondents feel that farm animals are at serious risk from plastic pollution. Some respondents reported that agricultural plastics are 'Dangerous towards livestock' (P 155) and that 'Animals (are) eating the plastics' (P355). Rumen impaction due to foreign bodies such as plastic materials can cause many different problems for animals. The ingestion of these materials can hinder physiological and chemical processes such as fermentation, which can lead to indigestion and microflora disruption. In addition to this, plastic ingestion over-time may lead to a build-up of toxins in the animal, which in turn could affect meat and milk quality intended for human consumption (Akraiem and Abd Al-Galil., 2016). Despite findings showing that annual plastic waste released into the environment by land-based sources is estimated as 4 to 23 times higher than from marine based sources, our results show that the perception of most farmers is that plastic pollution is more damaging to marine and freshwater environments. This reiterates the fact that knowledge of plastic and microplastic abundance and effects in terrestrial systems is still extremely limited (Horton et al., 2017).

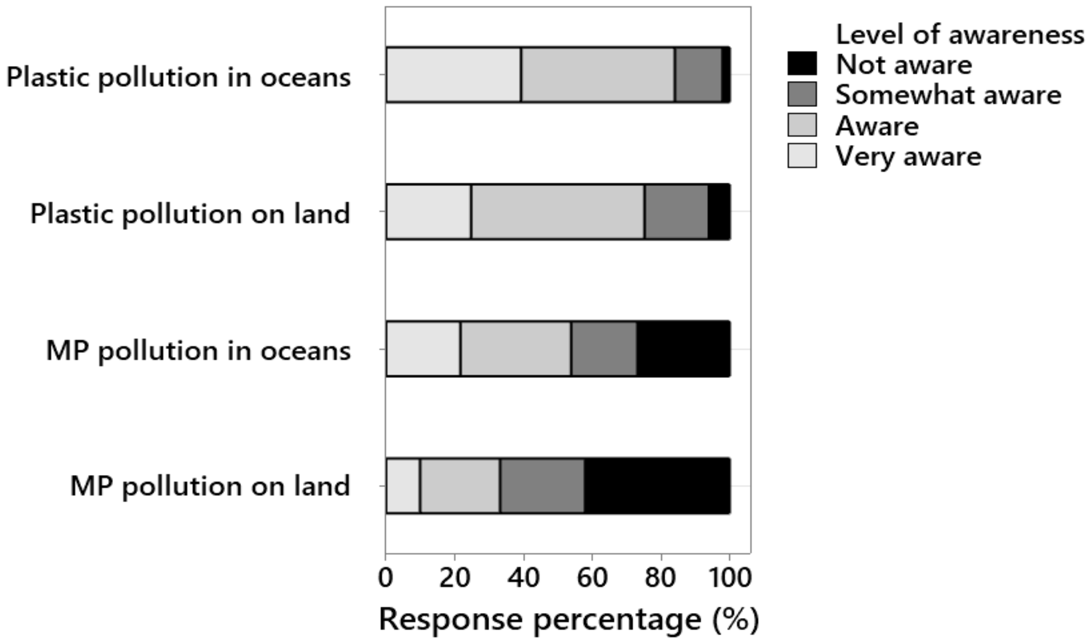


Figure 3.6: Respondents' level of awareness of plastic and microplastic pollution in the ocean and on land.

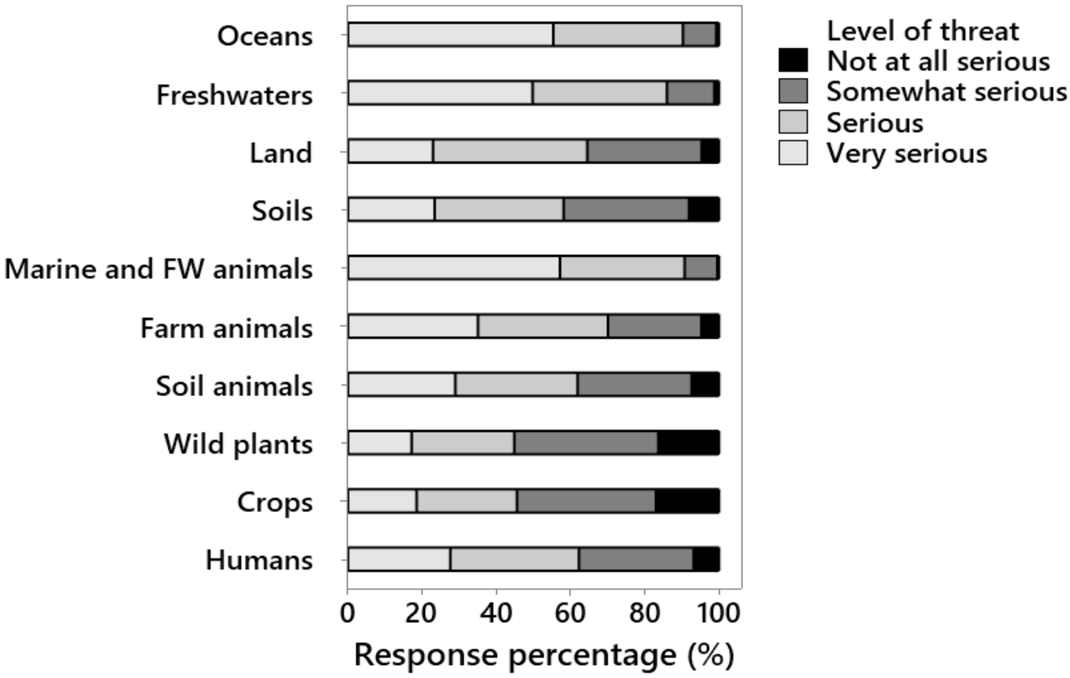


Figure 3.7: Respondents' perception of how serious of a threat plastic pollution poses on different compartments of the environment.

3.4.4. Strategies to combat plastic and microplastic pollution in agricultural soils

At a global level, in the coming decades, decisive changes regarding the implications of plastic pollution urgently need to be undertaken. Currently, there are a lack of specific legislation at a European level on the use of plastics in agriculture. There are also no criteria for sustainable soil management with reference to microplastics contamination in agriculture soils. There are policy developments in the EU coming out next year that focus on a new Soil Health Law with the vision that all soils will be 'healthy' by 2050. Two of the eight objectives of the EU Horizon Soil Mission include to "Reduce soil pollution and enhance restoration" and "Improve soil literacy in society". Some of the main findings from our study show that farmers believe agricultural plastics are a source of pollution; however, they perceive plastics pose a bigger risk to aquatic environments in comparison to the terrestrial, including soils. This indicates that an emphasis on the importance of soils and the pollution of soils by plastics must be delivered to society. Therefore, it is necessary for these new policy developments to include monitoring programmes to assess plastic and microplastic contamination levels in soils and initiatives to promote communication and citizen engagement.

Many farmers expressed that they use conventional agricultural plastics because there are no alternatives available. There is currently no single EU law in place on biobased, biodegradable or compostable plastics in a comprehensive manner. These materials may offer alternative solutions to the plastic pollution problem, but they also present many challenges. The certifications for these products are limited and unstandardised. Therefore, it is recommended that the new policy frameworks for biobased, biodegradable and compostable plastics should cover the economic and practical viability of these materials for use in agriculture.

3.4.5. Limitations of the study

Data for this study was collected both online and through hard copies. Collecting survey data online can be advantageous because it is relatively easy to conduct and can be administered via free platforms (Wu et al., 2022). However, there are also several limitations associated with collecting survey data online. The ability to reach certain types of participants can be challenging, and in our case, it was difficult to collect responses from potential participants who were inexperienced with digital platforms and/or those who have limited or no internet access due to living in remote areas. Thus, to capture the attitudes of these individuals, hard copies were made available to retrieve responses. However, the reliability of survey data using multiple data collection methods can depend on certain factors The primary concern is “Are responses to online surveys identical, similar or different to paper copy surveys?”. Online survey data collection may result in response bias due to the nature of data collection (Boyer et al., 2001).

Another limitation to the study is the socio-demographic profile of respondents. The number of participants under the age of fifty years represented the majority (73.3%) of respondents, in comparison to the number of respondents over the age of fifty (26.7%). However, according to Central Statistics Office (CSO) data, 53.3% of farm holders in Ireland are >55 years old (CSO, 2018). Whilst acknowledging that the population of the study were younger than the average named farmer, the younger generation of farmers will still have a significant impact on the sector for the coming decades and therefore the relevance of the sample population remains high. In addition to this, it is believed that the CSO data is collected based on the person who owns the farm, which is something that is not necessarily the same as the average age of farmers, and this subsequently makes it difficult to quantify the age-dynamic in Irish family farm structures.

3.5. Conclusions

The main conclusions of this study are:

1. Most farmers are recycling agricultural plastic waste. However, the rate of recycling depends on a wide range of factors including the type of agricultural plastic, cost, accessibility to recycling facilities and their knowledge on what can be recycled and how to recycle it. Initiatives should be put in place to educate farmers on how to recycle farm plastics correctly to help mitigate plastic and microplastic pollution in soils.

2. Farmers acknowledge that they need agricultural plastics to perform tasks on the farm, and that realistically no other material with suffice. However, many farmers view agricultural plastics as a burden due to the logistical and monetary factors associated with them. Farmers are also concerned about the negative impacts that the disposal of agricultural plastics present to the environment. Despite this, awareness and concern towards the environment does not always correspond into positive action. Some farmers openly admitted to the burning and burial of plastic waste on-site, which is not only damaging to the environment, but also is illegal.

3. Farmers are relatively aware of microplastics but are more aware of plastic pollution than microplastic pollution. In addition to this, farmers feel that aquatic environments are under greater threat than the terrestrial environments are. This demonstrates that farmers understand and care more about the impacts plastics and microplastics have on waterbodies and their entities (e.g., freshwater biota), which might because most of the research efforts have focused on these ecosystems to date. Further research on the abundance and potential effects of microplastics on soils is needed.

4. Combined efforts by the government, policy makers, and other stakeholders must be undertaken in order to reduce the plastic and microplastic problem. Developments should be made in relation to the policies regarding soil health and this includes the contamination of soil via plastics and the potential impacts plastics have on soil stability and structure. Moreover, the government should set out initiatives to promote citizen engagement to help improve the functionality of agro ecosystems. Furthermore, new research and innovation into the economic and practical viability of biobased and

biodegradable plastics should be addressed to investigate the potential of these materials as alternatives to conventional plastics.

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Chapter 4: The abundance, characteristics and potential sources of microplastic in Irish agricultural soils across different land-use types

4.1. ABSTRACT

Modern-day intensive agricultural production systems, such as high grass and crop yielding systems in temperate regions rely heavily on plastic materials. Consequently, these materials may act as entry routes for microplastics into agricultural soils and the wider environment. Prevention of microplastics from entering the human food chain is critical. To tackle this, the prevention and control of microplastics in agricultural food production systems is needed, which requires an evaluation of microplastics in the core resources of primary production such as soil. In this study, soils from 24 Irish agricultural fields, categorised into different land-uses (permanent grasslands, tillage soils with biosolids applied, tillage soils with plastic mulch films (PMFs) applied and tillage soils without biosolids or PMFs) based on their plastic pollution potential, were analysed. Soils from every field were found to be contaminated with microplastics, in concentrations ranging from 200 to 4899 MP items kg^{-1}. Significantly higher concentrations ($p \leq 0.001$) were found in soils treated with biosolids and PMFs, than in those without these treatments, indicating that these amendments are contributors of microplastic pollution in Irish agricultural soils. This study highlights potential risks from microplastic contamination in agricultural soils and the need for collective action by policymakers, regulators, scientists, and farmers to address them. It is recommended that systematic monitoring of microplastics in agricultural soils take place and that existing regulations on biosolids and PMFs consider microplastic as a factor. Further research is also warranted to examine the implications of microplastic on soil health and crop productivity, as well as the potential long-term impacts associated with PMFs accumulating in agricultural soils.

4.2. Aims and Objectives

The main aims and objectives of this study were to:

1. Quantify the abundance and characterisitcs of microplastics in agricultural soils across different farming land-use types in Ireland.

2. Identify the potential sources of microplastics found in Irish agricultural soils.

4.3. Methodology

4.3.1. Experimental design and study sites

Composite soil samples were collected from 24 agricultural fields between November 2020 and March 2021 located in the Midlands (Longford), North-East (Cavan and Monaghan), East (Louth, Meath, Kildare) and South-East (Wexford) of Ireland. Each field was grouped into one of the following four land-use categories: 1) tillage soils with no known history of PMF or biosolids application, 2) grassland (pasture) soils with no known history of PMF or biosolids application, 3) soils with a history of PMF application and 4) soils with a history of biosolids application. Six fields were assessed for each category, which is reported as farming land-use type. A pre-fieldwork questionnaire was completed on-site with the landowner to collect information of sample site and land-management history for each field sampled. Sample site information is shown in Table 4:1. Soil types were classified using Teagasc Soil Maps (Teagasc, 2017a). Three main soil types were recorded across the fields sampled (Surface Water Gleys, Brown Earths and Luvisols).

4.3.2. Soil sampling and preparation

As there is currently no established standardised method for sampling microplastics in soils (Möller et al., 2020), the method employed in this study was based on a similar sampling design used for soil nutrient and soil fertility testing (Thomas et al., 2014; Scrimgeour, 2008; Teagasc, 2017b). At all sites, a systematic "Inverted W" sampling design was modified and implemented (Figure 4:1). In total, 11 soil cores were collected in each field using a soil corer made with a steel extension bar steel auger (50 mm diameter) and a wooden handle. Soil samples were retrieved from the first 20 cm to capture microplastics in shallow topsoil (0 - 10 cm) and deeper topsoil (10 - 20 cm) and formed into one composite sample per field. Around 2 - 3 kg of wet soil was collected in each composite sample. Soils were sieved using a 5 mm mesh size stainless steel sieve (Endecotts™) and 200 g of soil from each composite sample was transferred to aluminium trays and dried in the oven at 50 °C over-night, or until a constant weight was recorded. Samples were dried at 50 °C as it is a temperature frequently adopted in microplastic studies, in order to preserve polymer integrity (Thomas et al., 2020). Any meso- and macroplastics found in soil cores were removed, washed down with Milli-Q water and dried overnight at room temperature; length measured and photographed using a Nikon camera (D3400). Mesoplastics were classified as plastics sized between 5 – 25 mm and macroplastics were classified as plastics sized > 25 mm (Jeyasanta et al., 2020; Li et al., 2022).

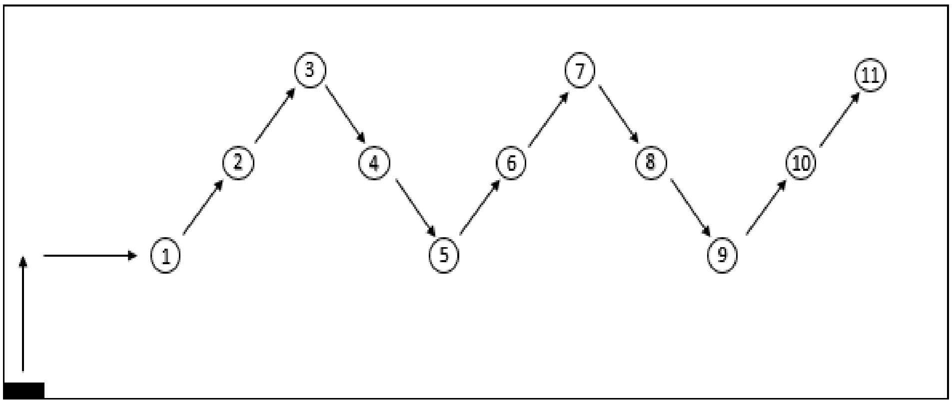

Figure 4.1: Schematic diagram of modified sampling system

Table 4.1: Farming land-use type, soil association, soil amendments (CS, CF, FYM, L, BS and PMF), crop rotation (2016-2020), ploughing depth (cm) and county.

Farming land-use type	**Soil association**	**Soil amendments**						**Crop rotations (2016-2020)**	**Ploughing depth (cm) (all annual)**	**County**
		CS	**CF**	**FYM**	**L**	**BS**	**PMF (in 20 y)**			
Tillage (1)	Surface Water Gleys	annual	(-)	(-)	(-)	(-)	(-)	Wheat/peas, oats/peas, oats/barley, oats, oats	15	Monaghan
Tillage (2)	Surface Water Gleys	annual	(-)	(-)	(-)	(-)	(-)	Wheat/peas, oats, wheat/peas, oats, oats/peas	15	Monaghan
Tillage (3)	Brown Earths	(-)	annual	-	1 in 4 y	(-)	(-)	Beet, wheat, barley, beans, wheat	18	Meath
Tillage (4)	Surface Water Gleys	(-)	annual	annual	(-)	(-)	(-)	Oats, wheat, wheat, barley, oil seed rape	20-25	Meath
Tillage (5)	Surface Water Gleys	(-)	annual	annual	1 in 4 y	(-)	(-)	Oats, wheat, wheat, barley, oil seed rape	20-25	Meath
Tillage (6)	Surface Water Gleys	(-)	annual	annual	1 in 4 y	(-)	(-)	Oats, wheat, wheat, barley, oil seed rape	20-25	Meath
Grassland (1)	Brown Earths	annual	(-)	(-)	(-)	(-)	(-)	(-)	(-)	Louth

Grassland (2)	Luvisols	(-)	annual	annual	(-)	(-)	(-)	(-)	(-)	Monaghan
Grassland (3)	Surface Water Gleys	bi-annual	(-)	(-)	(-)	(-)	(-)	(-)	(-)	Monaghan
Grassland (4)	Luvisols	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	Kildare
Grassland (5)	Luvisols	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	Kildare
Grassland (6)	Surface Water Gleys	bi-annual	annual	(-)	1	(-)	(-)	(-)	15-17.5 (1/15 y)	Cavan
PMF (1)	Luvisols	(-)	annual	annual	1 in 4 y	(-)	10	Maize	20	Longford
PMF (2)	Luvisols	(-)	annual	annual	1 in 4 y	(-)	20	Maize	20	Longford
PMF (3)	Brown Earths	annual	annual	annual	(-)	(-)	10	Maize	15	Wexford
PMF (4)	Brown Earths	annual	annual	annual	(-)	(-)	10	Maize	15	Wexford
PMF (5)	Surface Water Gleys	annual	annual	annual	(-)	(-)	5	Maize	30	Meath
PMF (6)	Luvisols	annual	annual	(-)	(-)	(-)	1	Maize	15	Meath
Biosolids (1)	Brown Earths	annual	annual	annual	2 in 5 y	1 app (2020)	(-)	Potatoes, barley, oil-seed rape, wheat, barley	20-25	Meath
Biosolids (2)	Brown Earths	annual	annual	annual	2 in 5 y	1 app (2019)	(-)	Potatoes, barley, oil-seed rape, wheat, barley	20-25	Meath
Biosolids (3)	Brown Earths	annual	annual	(-)	(-)	1 app (2019)	(-)	Peas, perennial ryegrass, barley, perennial	20	Louth

								ryegrass, oil seed rape		
Biosolids (4)	Brown Earths	annual	(-)	(-)	(-)	1 app (2020)	(-)	Peas, wheat, barley, oil seed rape, perennial ryegrass	20	Louth
Biosolids (5)	Brown Earths	annual	annual	(-)	(-)	1 app (2019)	(-)	Peas, wheat, barley, oil seed rape, perennial ryegrass	20	Louth
Biosolids (6)	Surface Water Gleys	(-)	(-)	(-)	(-)	3 app (2019,2020, 2021)	(-)	Willow (*Sallix sp.*)	20-25	Meath

CS = cattle slurry, CF = chemical fertiliser, FYM = farmyard manure, L = lime, BS = biosolids, PMF = plastic mulch film
(-) = not applicable

4.3.3. Microplastic extraction using density separation techniques

The most common techniques used to isolate microplastics from soils tend to rely on density separation methods (Thomas et al., 2020). These methods involve agitating soil samples with aqueous salt solutions to exploit the buoyancy of microplastic particles in solutions of higher densities than that of plastics (Cashman et al., 2020). In principle, certain plastic particles will remain in suspension or float to the top of the solution after a certain amount of time, while the precipitate (containing the soil mineral fraction), will remain at the bottom (Hidalgo-Ruz et al., 2012). The supernatant (floatation media containing potential microplastic particles) is then extracted for further processing. As the density of plastic polymers range from 0.8 - 2.35 g cm^{-3}, lower density solutions such as de-ionized filtered water (ρ = 1.00 g cm^{-3}) and saturated NaCl solutions (ρ = 1.20 g cm^{-3}) are suitable to separate low-density polymers including PE (ρ = 0.917 - 0.965 g cm^{-3}), PP (ρ = 0.9 - 0.91 g cm^{-3}) and PS (ρ = 1.04 – 1.1 g cm^{-3}) (Thomas et al., 2020). However, using higher density salts such as zinc chloride ($ZnCl_2$) (ρ = 1.5 - 1.7 g cm^{-3}) and sodium bromide (NaBr) (ρ = 1.55 g cm^{-3}) can facilitate the separation of denser polymers like PVC (ρ = 1.16 - 1.58 g cm^{-3}) and PET (ρ = 1.37 - 1.45g cm^{-3}) (Möller et al., 2020). This is because $ZnCl_2$ and NaBr reach higher densities that enable the flotation of e.g. PVC and PET. Standard NaCl solutions cannot float these denser polymers, leading to their underrepresentation (Zhang et al., 2020). Notably, using higher density salt solutions can make the differentiation of plastics and other soil components (e.g., SOM) is difficult because the density of SOM (ρ = 1.6 g cm^{-3}) is similar to the densities of plastic polymers and therefore, some organic matter may remain in suspension or float to the top of solutions (Cerli et al., 2012; Hidalgo-Ruz et al., 2012).

4.3.4. Microplastic extraction method development

One of the main challenges in microplastic research is the lack of harmonised and standardised methods for identifying and quantifying microplastics in a given environmental matrix**.** Matrices include water, soil and sediments, and there are many different methods that can be applied depending on the composition of the matrix (Raj and Maiti, 2023). Since microplastic research in soils started later than microplastic studies in water and marine systems, there are some gaps and limitations in the development of robust methods for quantifying microplastics in soils. Moreover, because soils are made up of mineral particles and organic materials, their structure is far more complex than that of water and so; it can be more difficult to isolate microplastics (Möller et al., 2020; Rede et al., 2023). Thus, the reliability of microplastic extraction procedures in soil is affected by multiple factors related to their structure. However, in order to support monitoring programmes and research, it is important to continue developing and evaluating the comparability of analytical methodologies.

In the initial stages of this research, multiple methodologies were trialed to identify the most effective approach for accurately quantifying both the abundance and types of microplastics present in agricultural soils. Given the limited availability of established protocols specifically tailored to soil at the time, existing methods developments for sediment analysis were adapted and refined to suit the more complex matrix of soil in order to enhance microplastic recovery.

4.3.4.1. Method A

The first method was taken from a study by Maes et al., (2017); which had been previously carried out on sediment samples. In brief, 200 ml of 5 M sodium chloride (NaCl) (ρ = 1.2 g/cm^{-3}) was added to 25 g of dried soil in a large beaker and magnetically stirred for 2 min, left overnight to settle and then the supernatant was filtered using vacuum filtration. The second method was adapted from Liebezeit and Dubaish, (2012), who used this method to extract microplastics from sediment samples. In total, 10 g of dried soil was digested with 40 ml of 30 % hydrogen peroxide (H_2O_2), by leaving in an orbital shaker overnight at 60 °C at 200 rpm. After this, 40 ml of 5 M $ZnCl_2$ (ρ = 1.55 g/cm^{-3}) was added to the digested soil solution and left settle overnight before filtration was carried out. For both methods, post-filtration, the filter papers were overly clogged with soil, which made the microplastics uncountable and unidentifiable under the microscope. These methods were excluded going forward based on the following learning outcomes; the 'washing' step with saturated salt solutions should be repeated multiple times, using both salt solutions instead of one over the other. Moreover, the sample volume of soil used was considered too large.

4.3.4.2. Method B

With this in mind, later experiments were conducted by adding 200 ml of 5 M $ZnCl_2$ (ρ = 1.55 g/cm^{-3}) to 5 g of dried soil, and shaken in an orbital shaker for 2 hours at 200 rpm. The supernatant was extracted, collected and stored in a glass jar. Forty ml of 30 % H_2O_2 was added to the remaining soil and $ZnCl_2$ layer, which was incubated overnight at 50

°C, followed by supernatant filtration. Microplastics were present and identifiable on filter papers, despite some organic matter still remaining. This method was not selected due to the high volumes of chemicals used, given the number of field soil samples that would be required for analysis.

4.3.4.3. Method C

The effectiveness of using separation flasks to isolate microplastics from soils was explored during method development. Approximately 10 g of dried soil was pre-digested overnight with 40 ml of 30 % H_2O_2 in an orbital shaker at 60 °C, overnight at 200 rpm. After this, 200 ml of 5 M $ZnCl_2$ (ρ = 1.55 g/cm^{-3}) was added to the flask and left to settle overnight in the separation flask. In this method, two distinct phases formed within the separation funnel: an upper layer containing the supernatant, where potential microplastics were suspended, and a lower layer composed of soil organic matter and mineral particles. However, the separation process was hindered during the elution stage, as the two layers frequently mixed upon opening the funnel valve, compromising the clarity of phase separation. To address this, repeated settling periods and increased care during elution were demonstrated, though successful separation often required multiple attempts over the course of three days per sample. This method was not selected, due to the considerable time it took to process each sample. Moreover, as with previous methods, excessive volumes of $ZnCl_2$ were consumed, and given the large number of soil samples, was impractical and resource-intensive. It also raised concerns regarding the environmental and logistical sustainability of using such high quantities of a high-density solution.

4.3.4.4. Method D

Based on a method performed by Ding et al., (2020), a different extraction solution was used. Approximately 10 g of dried soil was added to 100 ml of 3 M calcium chloride ($CaCl_2$) (ρ = 1.55 g/cm^{-3}) and the mixture was magnetically stirred for 2 min and then left to stand for 24 hours. The same steps were repeated on a 5 g sample of dried soil. The supernatant was collected and transferred to clean beakers. Approximately, 30 ml of 30 % H_2O_2 was added to the beaker and left to digest for 12 hours in an orbital shaker at 60 °C at 80 rpm.

This method was not adopted due to several limitations. Firstly, the organic matter digestion was ineffective, as indicated by the persistent brown coloration of the solution.

This residual coloration interfered with the post-filtration analysis, making microplastics unidentifiable under the microscope. Additionally, the magnetic stirring step proved insufficient; the $CaCl_2$ was slow to dissolve, reducing the overall efficiency of the process. Furthermore, the protocol lacked an adequate number of washing steps, which further compromised the clarity of the final extract.

4.3.4.5. Method E

During this stage of the method development, two studies were published based on microplastic extraction protocols from agricultural soils. One of the methods was adapted from Van den Berg et al., (2020) and involved a two-step density flotation protocol using a 3 g soil sample, distilled water and sodium iodide (NaI) solution (8 M) (ρ = 1.55 g/cm^{-3}). It also included an orbital shaking step (120 rpm for 2 hours), centrifugation (3000 rpm for 10 min), followed by vacuum filtration. This method was not selected because the iodide, which appears colourless in solution, exposed to oxygen, oxidized to iodine (I_2), caused a localised orange-brown stain on filter paper, which hindered the visibility of the extracted microplastics.

4.3.4.6. Method F

Instead, the method developed by Corradini et al., (2019) was selected for further refinement and implementation. In order to optimise the extraction of microplastics from soil samples, several practical adjustments were made. Glass centrifuge tubes were used to minimise contamination, and smaller volumes of soil and extraction solutions were used. Additional washing and centrifugation steps were incorporated using three sequential density solutions to enhance microplastic recovery. The use of H_2O_2 for organic matter was excluded, as it did not reliably remove all organic material, and posed risks of bleaching or degrading microplastics, particularly under higher temperatures.

4.3.5. Microplastic extraction from soils

A modified density separation wet extraction technique was implemented based on previous studies by Corradini et al., (2019) and Corradini et al., (2020). Modifications included adjusting the centrifugation speed to 5000 rpm and adding an extra extraction/washing and filtration step for better separation. Each field sample was analysed in triplicate. Dried soil samples were weighed on a balance and 5 ± 0.01 g was placed into 50 ml glass centrifuge tubes. Note that only 3 g of dried soil was used from fields applied with biosolids as these samples were expected to have higher numbers of microplastics. Twenty ml of Milli-Q water ($\rho = 1.00$ g cm^{-3}) was added to each tube and the samples were centrifuged at 5000 rpm for 15 min. Post-centrifugation, samples were allowed settle for a further 15 min. The supernatants in each tube were slowly decanted into clean, labelled beakers, carefully avoiding disruption of the sediment precipitate on the bottom of the centrifuge tubes. In between steps, all glassware-containing soils were covered with aluminium foil to prevent airborne plastic contaminants entering samples. Twenty ml of saturated 5 M NaCl solution ($\rho = 1.20$ g cm^{-3}) was added to the remaining precipitate in the tube and centrifuged for a second time, left settle and supernatants were collected and combined. A higher concentrated salt solution was used for the following washing/extraction steps. Twenty ml of 5 M $ZnCl_2$ 5M ($\rho = 1.55$ g cm^{-3}) was added to the precipitate in each tube for two final extractions, using the same steps as above. All supernatants were collected and filtered using Whatman MN Filter GF-3 Glass Microfibre Circles (pore size: 0.47 µm) papers. Filters were dried at room temperature in a glass desiccator and stored in labelled LDPE clear plastic petri dishes until visual identification.

4.3.6. Visual identification of microplastics

Filter papers containing microplastics were examined under an Olympus SZXY microscope, counted and characterised using microplastic identification protocols reported by Hidalgo-Ruz et al., (2012). Microplastics were identified and classified based on their shape (fibre, film bead, or fragment), size (0 – 499µm, 500-1000 µm and 1000-5000 µm) and colour (transparent, blue, black, red, green, multicoloured, brown, yellow or other). Several measures were adopted to test if suspected microplastics were made from plastic. First, the hardness test was performed to assess the texture and stiffness of suspected particles using a steel needle. Particles that are easily broken with minimal force are likely to be derived from organic matter, and mineral particles will also generate a "crunching" when pressure is applied. However, plastics will tend to have a higher degree of integrity and will resist the pressure and should not physically change, although highly weathered plastics may also be brittle (Lusher et al., 2020). In the case of highly weathered microplastics additional identification measures were implemented, such as the hot needle test. Under the pressure of a hot needle, plastic materials will melt or curve, while non-plastic material will not (Battaglia et al., 2020; De Witte et al., 2014). Microplastics were counted and characterised based on their size, shape and colour. After this, a sub-sample (13.8%) of microplastics were selected at random from soils of each different land-use type for further characterisation and polymer identification using Raman Spectroscopy. Other studies have used polymer identification methods on up to 10 % and 20 % (Huang et al., 2020; Horton et al., 2017) of the visually identified microplastics, however another study only used 5 % of the total number of suspected microplastics (Jiang et al., 2020). The percentage examined typically depends on a variety of factors including the number of microplastics found in samples, the number of samples, and can sometimes also depend on time, logistics and money constraints.

4.3.7. Raman spectroscopy

Raman spectroscopy is one of the identification methods used to characterise the polymers of microplastics under study. Raman spectroscopy is an analytical technique used to provide detailed information about the chemical structure and molecular interactions in a sample. Monochromatic light from a laser interacts with a sample and most of the photons are elastically scattered (Rayleigh scattering). A small fraction of the light undergoes inelastic scattering whereby photons transfer energy to or from the sample, which results in a shift in wavelength (Raman effect) (Keresztury et al., 2006).

These energy shifts directly correspond to vibrational modes of the molecules in the sample, generating a spectrum that provides a unique "fingerprint" to the material (Käppler et al., 2016). The spectra were then compared to a spectral referenced library or database to find matches in order to determine whether the sample was plastic, inorganic, or other. Any suspected microplastic particles detected as non-plastic or not identified, were deducted from the total original suspected microplastic counts (Xu et al., 2020).

Microplastics were analysed further using Raman spectroscopy. Microplastic particles were picked with the aid of a thin-tipped tweezers (n = 107) from the original counts (n = 773), mounted to a glass slide using double sticky tape, and analysed using the Raman spectrophotometer (Horiba LabRAM II, Horiba Jobin-Yvon, France). The Raman Spectrometer had a 600-groove mm^{-1} diffraction grating, a confocal optical system, a Peltier-cooled CCD detector and an Olympus BX41 microscope (O'Briain et al., 2020; Loughlin et al., 2021). Spectra were obtained at a range of 100 - 3500 cm^{-1} using a 532 nm laser. All spectra were compared to a spectral reference library (KnowItAll, Bio-Rad) and an in-house extension library was used which contained known virgin polymer type spectra (purchased from CARAT GmbH, Bocholt, Germany) (Mendes et al., 2021). The websites 'Open Specy' (Cowger et al. 2021) (https://openanalysis.org/openspecy/) and 'PublicSpectra' (https://publicspectra.com/SpectralSearch) were also used to identify polymers via Raman. The Raman spectrophotometer was used in the Ryan Institute in the University of Galway under the authorisation of both Dr. Liam Morrison and Dr. Ana Marques Mendes.

4.3.8. Quality control measures

Thorough quality control measures were implemented at all stages of sample collection and analyses to minimise microplastic contamination (Hermsen et al., 2018). All work conducted on samples was carried out in a 'clean room' which was specifically used for microplastic work exclusively by a lone operator. The entrance to the room had a sticky mat in place to catch any dust or potential microplastics trapped on footwear and transferred into the room. Before commencing any work in the room, floors were hoovered, and all workspaces were cleaned. A © Dyson model hoover was used as it contained a High Efficiency Particulate Air (HEPA) Filter specifically designed to remove airborne particles such as microplastics. All materials used were made from glass or steel, and wearing synthetic clothing during field and lab-work was minimised. Only 100% cotton lab coats and latex gloves were used during laboratory analyses.

Before and between all steps, the equipment was triple rinsed using pre-filtered Milli-Q water (0.22 µm). All materials including glassware, samples, tweezers, etc., that were not in use were covered with aluminium foil to avoid airborne contamination in between steps.

Procedural blanks were also incorporated (n = 9) (See Appendix) (Munno et al., 2023) and all solutions used in this study for microplastic extractions were pre-filtered before use (Whatman MN Filter GF-3 Glass Microfibre Circles (pore size: 0.47 µm). To monitor airborne contamination, settling plates (the same type of filter papers) were left out (n = 75) on days of sample processing and analyses and examined for suspected microplastic contamination (Gwinnett and Miller, 2021). Airborne contamination was considered negligible when compared to the amount of microplastics recovered from each sample matrix thus no correction measurement was carried out (Aminah and Ikejima, 2022) (See Appendix). In total, 10 potential microplastic fibres were identified on the filter papers used to monitor airborne contamination. Half of these suspected microplastics were analysed using Raman Spectroscopy. One black fibre was determined as PET and four transparent fibres were detected as cotton (see Appendix).

Positive controls were adopted to validate the microplastic extraction method. Numerous authors have noted the importance and necessity to perform the spike-and-recovery method as a standardisation protocol in microplastic analysis (Mai et al., 2020; Miller et al., 2017; Wright et al., 2020). Each soil sample (n = 6) were spiked with prepared microplastics (n = 24) including white PP spheres (n = 6) (size: 1.55 ± 0.05 mm) red PE spheres (n = 6) (0.5 - 0.6 mm) purchased from Cospheric LLC ©. Transparent PC fragments were prepared by cutting smaller fragments (n = 6) (2.2 ± 0.4 mm) of plastic from a PC plastic petri dish using a scalpel blade, scissors and tweezers. Pink polyester fibres were removed from a 100 % polyester fleece and cut into smaller fibres (n = 6) (2.7 ± 0.9 mm) using a scissors. Over 90% recovery rate was achieved for all spiked samples (See Appendix).

4.3.9. Statistics and data analysis

Microplastic concentrations are reported in MP items kg^{-1}. Descriptive statistics including the range, mean and standard deviation for all sites were calculated on Minitab® 21.3 (64-bit). Spatial analysis techniques were conducted in geographical information systems using Esri® ArcGIS® in order to establish whether there was a relationship between the microplastic counts in each field and the distance to a road and an urban settlement. The "proximity" toolset was applied to analyse the distance between the sampling points and a main road, and within 1 km distance was considered as "close". Another potential source was urban settlements, of which are defined as urban areas with 1,500 people or more. The "proximity" toolset was applied to analyse the distance between the sampling points and each urban settlement, and "close" was considered as within 2 km distance. The data were transferred into Minitab® 21.3 (64-bit), and Kolmogorov-Smirnov tests and QQ-plots were conducted to test the normality of the data. All data were shown not normally distributed; thus, the statistical analysis was underpinned by non-parametric Kruskal Wallis tests to confirm the statistical significance of the differences in microplastic abundance based on the four farming land-use types, proximity to a road, and proximity to an urban settlement, with a significance threshold of ≤ 0.05 obeyed.

4.5. Results and Discussion

4.5.1. Meso- and microplastic films found in soils applied with plastic mulch films (PMFs)

Approximately 15.3 kg of wet soil was collected from the six-PMF fields and processed for meso- and macroplastics. In total, 77 mesoplastic films were recorded, with a mean length of 1.65 ± 0.5 cm. Plastic mulch film field (4) had undergone ten consecutive years of PMF application and contained the highest number of mesoplastic films with 9.1 mesoplastic films per kg of soil (wet weight) (Table 4.2). This was followed by PMF field (2) and (3) where 5.6 and 5.3 mesoplastic films were found respectively. More macroplastic films were retrieved from the same sample volume of soil with 115 macroplastic films recovered across the six fields (Table 4.3). Plastic mulch film field (2), had twenty years of PMF applications contained the most macroplastic films (20.4 macroplastic films per kg of soil (wet weight), with a mean length of 4.5 ± 1.6 cm). All meso- and macroplastic films were characterised as transparent films that resembled the types of films used for mulching. Several sample films (n = 8) were subject to Raman Spectroscopy analysis, revealing that all films were identified as polyethylene (PE). These are likely to have originated from the PMFs used at the sites, which are composed of photo-oxodegradable plastics. Such plastics have been banned for sale in Ireland since 2021, under the Single Use Plastics Directive (Directive EU 2019/904). These films are typically produced by combining conventional plastic polymers such as PE with pro-oxidant additives (e.g., trace metals) to trigger the breakdown of polyolefin under thermal or photochemical activation (Thomas et al., 2012). However, these PMFs have been scrutinised in the past because they do not ensure complete plastic degradation. Under specific laboratory conditions, they can become 'invisible' after a period of two years; however, in the field complete degradation is never fully achieved (Folino et al., 2023). Instead, they fragment into smaller macroplastics, mesoplastics and eventually microplastic and nanoplastic (Markowicz and Szymańska-Pulikowska, 2019), which is suspected in this study.

Table 4.2: The number and length (cm) of mesoplastic films recovered from soils in the six tillage fields utilising plastic mulch films (PMFs).

Field	Number of PMF application (out of 20 years)	Total mesoplastic film count	Weight of soil sampled (kg^{-1} wet weight)	Mesoplastic films per kg^{-1} soil (wet weight)	Mean length ± SD (cm)	Min length (cm)	Max length (cm)
PMF (1)	10/20	6	2.3	2.6	1.9 ± 0.3	1.4	2.3
PMF (2)	20/20	14	2.5	5.6	1.7 ± 0.4	1.1	2.1
PMF (3)	10/20	16	3	5.3	1.6 ± 0.6	0.6	2.5
PMF (4)	10/20	22	2.4	9.1	1.7 ± 0.6	0.8	2.5
PMF (5)	5/20	13	2.6	5	1.4 ± 0.6	0.7	2.5
PMF (6)	1/20	6	2.5	2.4	1.6 ± 0.4	1	2.2

Table 4.3: The number and length (cm) of macroplastic films recovered from soils in the six tillage fields utilising plastic mulch films (PMFs).

Field	Number of PMF application (out of 20 years)	Total macroplastic film count	Weight of soil sampled (kg^{-1} wet weight)	Macroplastic films per kg^{-1} soil (wet weight)	Mean length ± SD (cm)	Min length (cm)	Max length (cm)
PMF (1)	10/20	14	2.3	6.1	5.2 ± 2.4	3.2	11.1
PMF (2)	20/20	51	2.5	20.4	4.5 ± 1.6	2.6	11.5
PMF (3)	10/20	21	3	7	4.6 ± 1.9	2.6	10.4
PMF (4)	10/20	17	2.4	7.1	4.7 ± 1.7	2.9	8.9
PMF (5)	5/20	11	2.6	4.2	4.6 ± 1.5	2.7	7.6
PMF (6)	1/20	1	2.5	0.4	4.0 ± 0	4	4

4.5.2. Plastic mulch films (PMFs) and biosolids as sources of microplastics in Irish agricultural soils

Soils sampled from every field were contaminated with microplastics. Microplastic concentrations ranged from 200 to 4899 MP items kg^{-1} soil, with a mean concentration of 1851 ± 1222 MP items kg^{-1}. Soils applied with biosolids (2748 ± 529 MP items kg^{-1}) and PMFs (2448 ± 435 MP items kg^{-1}) contained significantly higher concentrations of microplastic than permanent grassland (pasture) soils (1508 ± 258 MP items kg^{-1}) and tillage soils (700 ± 194 MP items kg^{-1}) without PMFs or biosolids application ($p \leq 0.001$) (Figure 4.2).

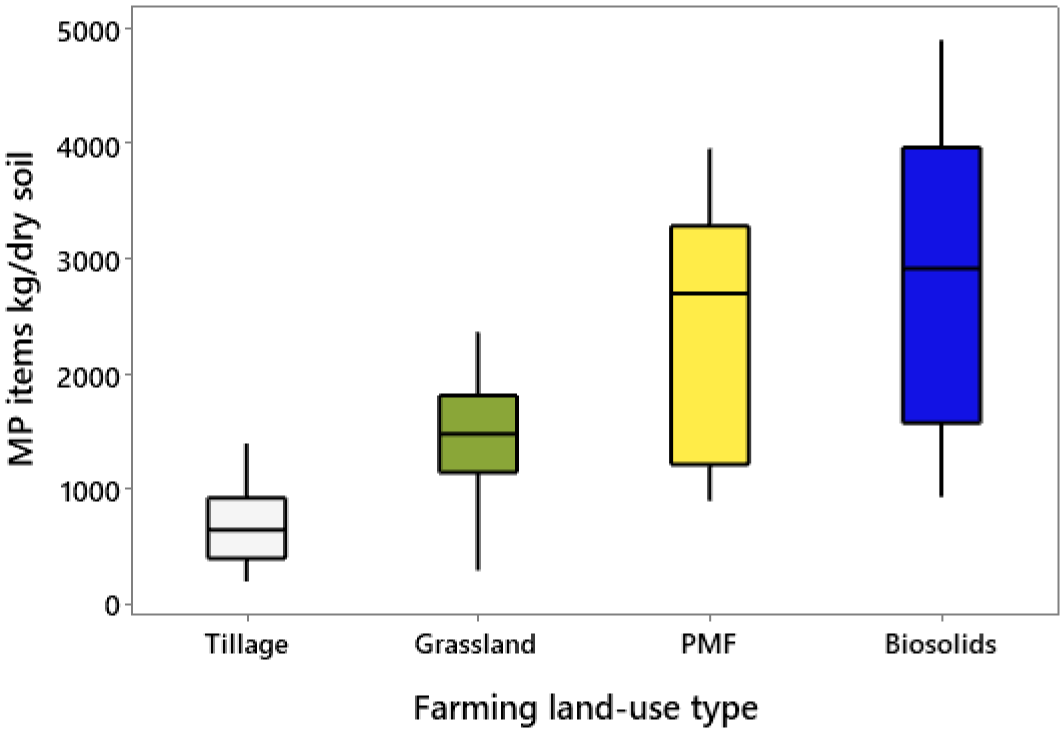

Figure 4.2: Microplastic abundance in agricultural soils across various farming land-use types

These results are comparable to recent and past findings on microplastics in agricultural soils (Adhikari et al., 2024; Naderi Beni et al., 2023; Yu et al., 2023). Most of the publications on microplastic abundance in agricultural soils have been conducted in China and Eastern Asia (Cai et al., 2023; Cao et al., 2021; Ding et al., 2020; Shi et al., 2024; Yu et al., 2021; Yu et al., 2023), and so available data on microplastic concentrations in European soils is limited. Only one other study has been carried out on microplastics in agricultural soils in Ireland (Heerey et al., 2023), where microplastics were found in concentrations of up to 2103 MPs items kg^{-1} and the sources of microplastic in the sampled soils were concluded to be from both biosolids, and plastic mulch sources. Other studies across Europe show that biosolid amended soils in Poland contained 200 - 1100 MPs items kg^{-1} (Medyńska-Juraszek and Szczepańska, 2023) and between 999 – 8658 MPs items kg^{-1} found in soils heavily applied with biosolids in Spain (van den Berg et al., 2020) (Table 4.4).

Table 4.4: Comparative summary of the sources and concentrations of microplastics found in this study and internationally in agricultural soils.

Sample	Potential Direct Sources of Microplastics	Abundance (range) (MP items kg^{-1} soil)	Country	Reference
Tillage soils growing crops	Biosolids	0 - 10,200	Chile	Corradini et al., (2019)
Tillage soils growing crops	Biosolids	999 - 8658	Spain	Van den Berg et al., (2020)
Tillage soils growing crops and vegetables	PMFs, greenhouse films, irrigation from sewage	50 - 880	Tunisia	Chouchene et al., (2022)
Tillage soils growing crops	Biosolids	200 – 1100	Poland	Medyńska-Juraszek, (2023)
Tillage soils growing crops	Biosolids, PMFs	0- 2103	Ireland	Heerey et al., (2023)
Tillage soils growing crops	Biosolids	360 - 500	USA	Adhikari et al., (2024)
Tillage soils growing crops	PMFs, fleece, nets	1320 – 8190	UK	Cusworth et al., (2024)
Tillage soils growing crops and grassland soils	Biosolids, PMFs	200 - 4899	Ireland	**This study**

This study shows that PMFs are a direct source of microplastic contamination in agricultural soils. Maize is the predominant crop grown under PMFs in Irish agriculture, although not widespread. In other European countries, PMFs are widely used for the production of crops with specific climatic and soil conditions, such as strawberries, asparagus, and leafy vegetables (Henseler, 2023). In other parts of the world, PMFs are used for growing wheat and potatoes (Qin et al., 2014; Amare and Desta, 2021). Traditionally maize growers used oxo-degradable PMFs. As mentioned, these are now banned for sale under the EU's Single Use Plastics Directive (2019/904) due to the risks of fragmentation into microplastics and/or chemical decomposition in the soil. These films do not guarantee proper biodegradation and thus contribute to microplastic pollution in the environment, which is demonstrated in this study. Farmers may continue using previously purchased oxo-degradable PMFs for up to five years after the directive comes into effect, and some farmers have been using PMFs on their land for nearly thirty years. However, obtaining historical data on their usage and the polymers used in past PMFs remains challenging. Research is needed to understand the long-term effects of PMFs buried in soils and the potential chemical migration from film additives to soils, their bioavailability and risks to food safety. This study provides evidence of microplastics entering Irish agricultural soils through biosolid application. Ireland has the highest reuse rate of biosolids in agriculture as a fertiliser to soil in the EU (up to 98%) (Uisce Éireann, 2023). At present, the total annual volume of dry sludge solids generated in Ireland is approx. 53,000 tonnes which is expected to increase to 96,000 tonnes p.a. by 2040 (Heerey et al., 2023). Biosolids and sewage sludge are heavily polluted with microplastic, with 4196 to 15,385 MPs kg^{-1} found in sources from Ireland (Mahon et al., 2017), and over 286,000 MPs kg^{-1} in the UK (Harley-Nyang et al., 2022). The removal efficiency of microplastics during wastewater treatment depends on the treatment techniques used and to date, there is no approach to remove all plastic residues including microplastics from sludge (Christian and Koper, 2023). This implies that significant amounts of microplastics may be entering Irish agricultural soils where biosolids are applied. Currently, there is no integrated approach or data system available with information the application rates and areas where biosolids are applied in Ireland, or in Europe. There are also no monitoring programmes on microplastics in biosolids, and no guidance or regulations in place on the presence of microplastics in biosolids and their associated risks to soil agro-ecosystems and the wider environment. If the use of biosolids in farming is controlled or restricted, this may put the biosolids market and a valuable source of nutrients to the agricultural sector at risk (Harley-Nyang et al., 2022). In addition to this, producers could face major challenges on tackling the disposal of wastewater sludges produced. Further strategies including environmental risk assessments should be carried out to minimise biosolids as a pathway of microplastics into agricultural soils. Most of the agricultural land in Ireland (82%) is dominated by permanent grasslands that are not applied with PMFs, and none of the permanent grassland soils in this study were applied with biosolids suggesting that other sources of microplastics in soils exist. Microplastic characteristics such as shape, colour and polymer type can offer insights into the potential other sources of microplastic in soils.

4.5.3. Microplastic characteristics potential risks and sources

In this study, the category that represented the vast majority of microplastics found across all fields sampled were fibres, with on average 91% of samples containing fibres, followed by fragments (6%) and films (3%). No beads or foams were identified in any samples (Figure 4.3).

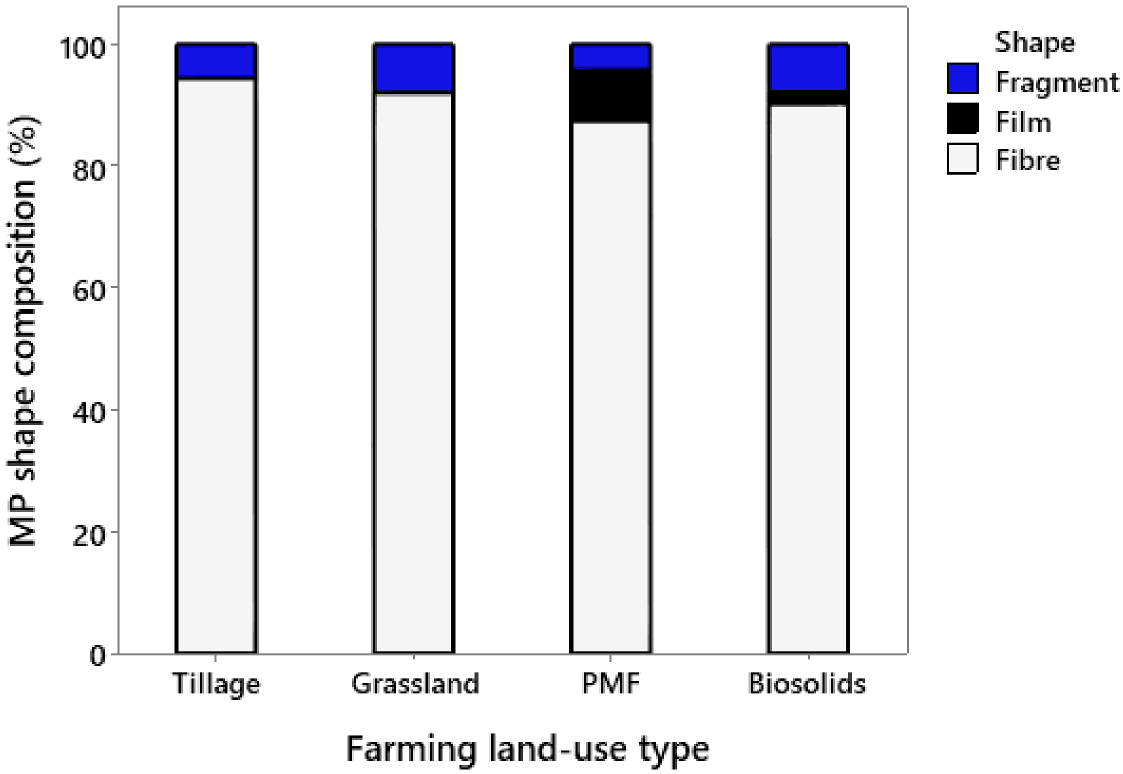

Figure 4.3 Microplastic shape compositions found in agricultural soils across different farming land-use types.

Fibres are abundant in biosolids from sources such as domestic and industrial wastewater (Gkika et al., 2023; Sivarajah et al., 2023), and when biosolids are applied on-land, contribute to the number of microfibres found in soils (Harley-Nyang et al., 2022). In this study, fibres were found across all farming land-use groups, regardless of whether biosolids had been applied. This finding was expected as fibres are typically the most abundant type of microplastics found in various environmental settings (de Oliveira et al., 2023; Sait et al., 2021; Santonicola et al., 2023), including the air (Torres-Agullo et al., 2021). This suggests that microplastic fibres may enter soils through atmospheric deposition (Enyoh et al., 2019; Klein and Fischer, 2019), as well as other potential pathways, such as inputs from chemical and organic fertilisers (Yang et al., 2021; Guo et al., 2023; Surendran et al., 2023). Each year, over 40 million tonnes of animal waste and slurry are spread on agricultural land in Ireland (Health and Safety Authority, 2018). Microplastic fibres have been detected in faecal matter (Pérez-Guevara et al., 2021) and the widespread practice of applying raw or treated animal manure to fields may serve as a pathway for microplastic entry to agricultural soils. In this study, 21 out of the 24 sampled fields had received either organic fertiliser such as cattle slurry or farmyard manure, inorganic chemical fertilisers, or both spread within the same year of sampling. A study in the UK found that microplastic concentrations in soils increased following the application of organic and inorganic fertilisers, highlighting their role as contributor to microplastic pollution in agricultural soils over-time (Cusworth et al., 2024). This suggests that chemical and organic fertilisers may also be a potential source of microplastics in agricultural soils. However, this study does not provide direct evidence, and further research is warranted in order to confirm this source. Film-shaped microplastics were found exclusively in soils where PMFs or biosolids had been applied. This was expected due to material properties of PMFs and their ability to break down into microplastic films through weathering, biodegradation, repeated tillage and mechanical stress, which is likely to accelerate the fragmentation process (Qiang et al., 2023). In a study by Lehmann et al., (2021) microplastic films from plastic mulching modified soil structure by introducing artificial pores, preventing the formation of large soil aggregates. In a separate study, fibre shaped microplastics decreased the formation of soil aggregates by 29% due to the introduction of fracture points into aggregates (Lozano et al., 2021). Not only does the shape of microplastics have the potential to physically alter the structure of soils but also shape (as well as size and colour) may potentially influence ingestion by soil biota, which has been demonstrated in aquatic research (Casagrande et al., 2024; Xiong et al., 2019).

Most microplastics (66%) found across all samples were between 1000 – 5000 µm, and 34% were less than 1000 µm in size (Figure 4.4). Depending on the size of microplastics, they can have different effects on soil processes (Chen et al., 2022). In the terrestrial environment, smaller sized microplastics may potentially block soil micropores, be absorbed by plants or ingested by soil organisms in comparison to larger microplastics. Soil microarthropods can facilitate the uptake of microplastic particles in soils in the smaller size ranges (2 – 34 µm) (Kim and An, 2020; Lahive et al., 2022), and due to their greater surface area have a higher capacity to adsorb pollutants in the soil (Fu et al., 2022; Wang et al., 2019). However, studies have shown that *Lumbricus terrestris* can ingest larger microplastics up to 1000 µm, with many deposited in casts via excretion (Rillig et al., 2017). The ingestion of microplastics and nanoplastics by smaller terrestrial organisms such as collembola, nematoda and gastropoda has shown varying effects on growth, reproduction, tissue damage and intestinal blockages (Zhang et al., 2022). Different sized microplastics can provide microhabitats for soil microbes (Chen et al., 2022; Ya et al., 2022), and although this has the potential to enhance microbial diversity (Zhang et al., 2022); it can disrupt the microbial communities already present in the soil (Wu et al., 2023). Given the broad range of microplastic sizes found in this study, all of these factors could be considered a risk on soils, regardless of farming land-use type.

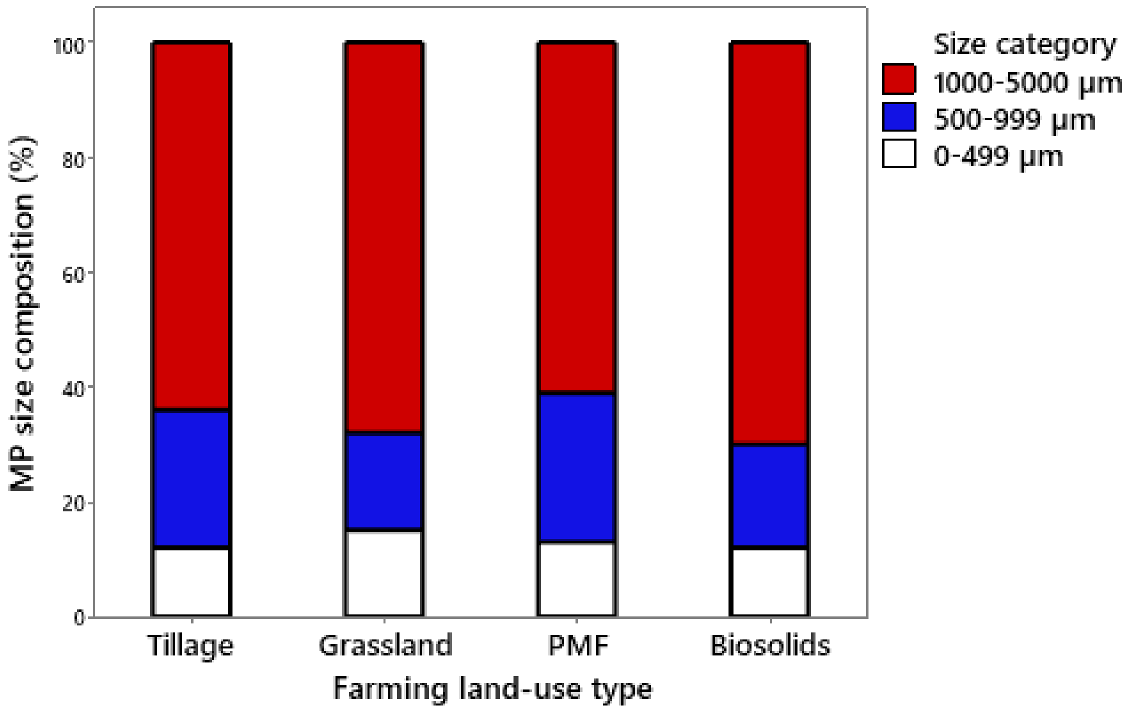

Figure 4.4: Microplastic sizes found in agricultural soils across different farming land-use types.

Nine subsets of colours were characterised and their abundance relative to farming land-use type is captured in Figure 4.5. Black microplastics were the most commonly identified colour group (38%), with more found in tillage (cereal crop) soils and grassland (pasture) soils, followed by transparent (26%) and blue microplastics (24%). Across all groups, red and green coloured microplastics were present, but only made up < 10% of all microplastic colours recorded. Colours such as yellow, brown, multi-coloured and others were found in < 2% of samples analysed. Black coloured microplastics may be indicative of other microplastic sources including road-tyre particles, like has been reported elsewhere (Giechaskiel et al., 2024; Worek et al., 2022), or potentially from farm plastic materials. To ensure consistent feed for cattle during periods of reduced pasture growth, many countries with temperate climates engage in large-scale silage production (Teagasc, 2024). In Europe, cattle farming is significant part of agricultural systems, and silage provides feed supplies year-round (Wilkinson and Rinne, 2018). Approximately one third of silage in Ireland is wrapped in conventional PE black plastic, which reflects a trend seen in various temperate regions and may be contributing to microplastic contamination in soils. The highest number of transparent microplastics were found in biosolid-applied soils (35%) and PMF soils (30%) in comparison to grassland (pasture) soils (16%).

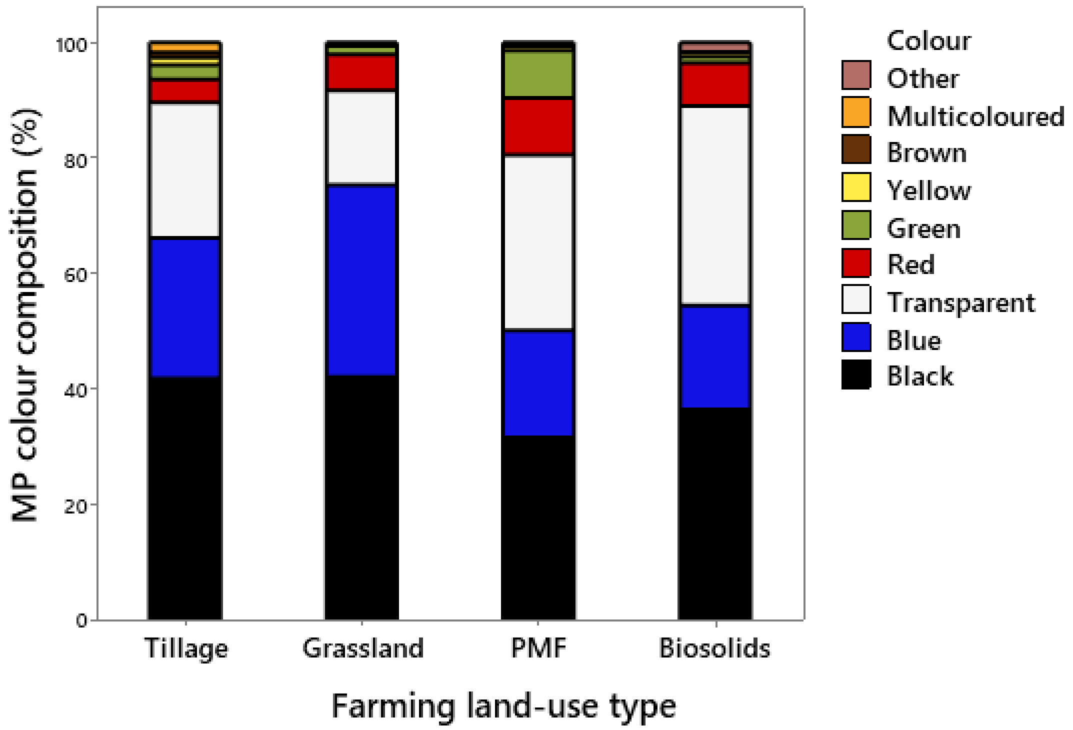

Figure 4.5: Microplastic colour compositions found in agricultural soils across different farming land-use types**.**

Polyethylene (PE) was the most commonly identified polymer across samples (22%), followed by polyethylene terephthalate (PET) (21%), polyvinyl chloride (PVC) (17%), polyurethane (PU) (15%), polyamide (PA) (8%), polypropylene (PP) (7%), polybutyl methylacrylate (PBMA) (6 %), and polystyrene (PS) (4%) (Figure 4.6). The higher proportions of PE were expected as it the most common type of plastic used worldwide (Lise et al., 2014; Zhang et al., 2017), and particularly on farms where PE plastics are used to mulch crops and wrap and store silage (Shah et al., 2020; Picuno, 2021). Polyethylene terephthalate was the second most common polymer identified in the soil samples. Polyethylene terephthalate is the sixth most-produced plastic globally; it is the fastest-growing plastic produced from fossil fuels, and the third most commercially used in packaging material (Bohre et al., 2023). The number of microplastics identified as PP were lower than expected. Globally, PP is the second most manufactured plastic produced after PE (Parku et al., 2020) however, it was the most produced polymer in European plastics production in 2022 (Plastics Europe, 2023). It is also one of the main polymers used in agricultural practices.

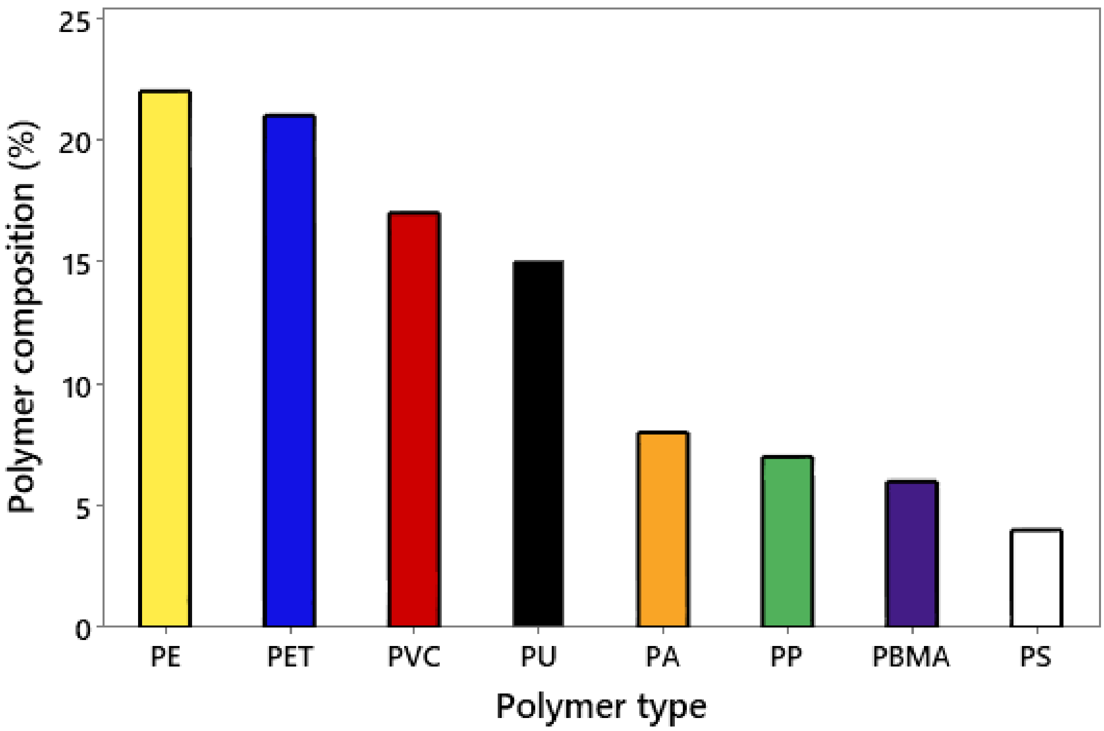

Figure 4.6: Polymer classifications of microplastics found in Irish agricultural soils.

Polyethylene terephthalate and PU made up 60% of the polymers detected in tillage (cereal crop) soils without PMFs or biosolids (Figure 4.7a). A mix of polymers (PE, PET, PU and PVC) were distributed almost equally in permanent grassland (pasture) soils, and PP was present in over 10 % of samples from this group (Figure 4.7b). Polyethylene was the dominant polymer found in soils from plastic mulch fields (64%), followed by PVC (18%), PP (9%) and PA (9%) (Figure 4.7c). Soils applied with biosolids contained over 50% of PET and PA which are the dominating polymers used in the production of synthetic textiles and clothing materials (Figure 4.7d). This result was expected due to previous work carried out showing that PET and PA are ubiquitous in biosolid and sewage sludge samples that come from the shedding of microplastics during synthetic clothes washing cycles that enter into the wastewater stream (Marchuk et al., 2023). In this study, PU was the third most detected polymer when combining results across all sample sites. According to Plastics Europe, (2019), PU was the fifth most manufactured plastic produced in the European Union (EU). Polyurethane is a major polymer used within the automotive industry, where it is incorporated into dashboards and door liners, but also in vehicle tyres. Moreover, PU is also widely used in the production of coatings and adhesives (Iordachescu et al., 2024), and in the coating of typical controlled-release fertilisers used in agriculture. It is unknown whether these types of fertilisers were utilised by farmers in this study. Nonetheless, all of the aforementioned may all represent potential sources of microplastics found in the soils sampled in this study.

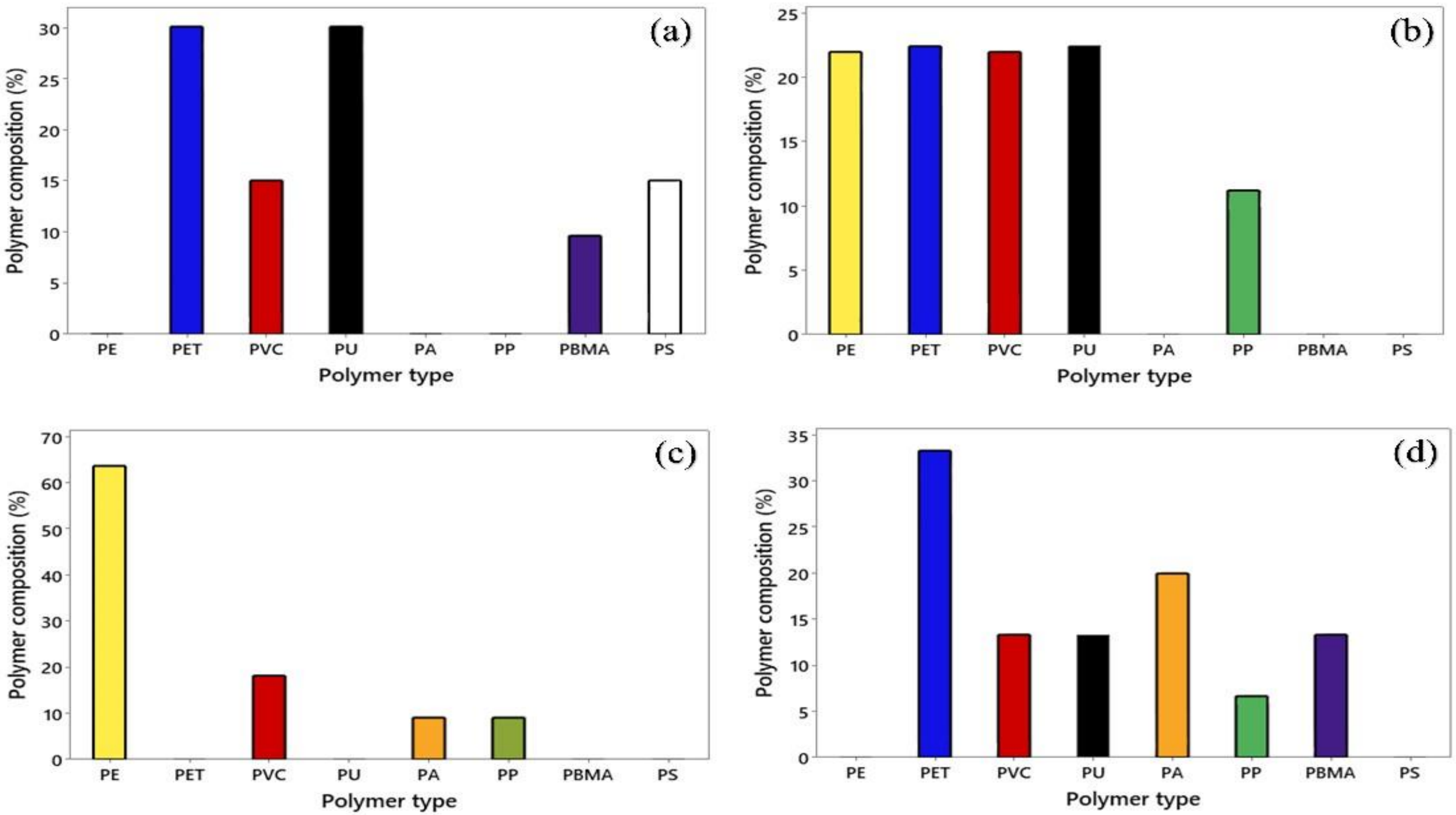

Figure 4.7: Polymer classifications of microplastics found in agricultural soils across different farming land-use types: (a) tillage, (b) grassland, (c) PMF and (d) biosolids.

4.5.4. Practical implications of this study

This study revealed that microplastics were found across all fields sampled, irrespective of which land-use practices were in place on the farm. This finding potentially poses implications for the sustainable development of agriculture. Globally, agriculture has been the focus of concerns around environmental and economic sustainability, and the demands of modern farming can burden farmers with substantial stress (Brennan et al., 2022). Farmers need to be able to feed the growing population, and food production needs to double by 2050 (Sands et al., 2023). However, farmers are facing major challenges due to the changing climate, unpredicted weather patterns, soil erosion and depletion, nutrient losses and pollution, and compliance with government regulations (Attorp, 2022; Hammersley et al., 2021; Wheeler and Lobley, 2021). Meanwhile enough produce needs to be generated to cover the financial outgoings on the farm, as well as their own cost of living (Brennan et al., 2022). As a result, farmers are using chemical fertilisers, organic fertilisers, biosolids, polymer coated seeds, and PMFs in order to enhance yields and increase productivity (Hofmann et al., 2023). Consequently, the use of these soil amendments can lead to the accumulation of microplastics in the soil, which may potentially alter the physical, chemical and biological properties of soil (de Souza Machado et al., 2018; Lian et al., 2021; Yu et al., 2022). Microplastics are very persistent in the environment, their effects on soils are highly dependent on the properties of the microplastic, including size, shape, and chemical composition, along with the release of toxic additives from microplastic and the transport of other pollutants through the soil as chemical and biological contaminants can bind well to microplastic particles (Wang et al., 2022). All of these factors may pose potential risks to human health and well-being as studies have documented the ability of certain animals and plants to ingest/uptake microplastics, promoting trophic transfer of microplastics and their contaminants, which make their way into the human food chain (Mamun et al., 2023). There is currently not enough scientific evidence to link human disease with microplastic consumption due to the complexity and multifaceted nature of microplastics. Despite this, many of the constituents of microplastics have been shown to directly affect the health and well-being of humans (Lal et al., 2021). It is known that soil can have a profound impact on human health, which can be positive or negative, direct or indirect. For example, utrient imbalances in soil and the presence of biological pathogens can directly cause negative effects to human health (Brevik et al., 2020; Steffan et al., 2017). In the last century there has been progress made in understanding the links between soil and human health, however, like microplastics, these interactions are complicated and further require a considerable amount of research attention. Therefore, investigating the risks associated with microplastics found in agricultural soils and human health is warranted since soils are a major sink and source of microplastic in the environment.

An extensive amount of research has been conducted on microplastics in water and the marine environment and there has been great emphasis on the role that wastewater treatment facilities play in removing microplastics from water to prevent entry into aquatic ecosystems (Acarer, 2023; Iyare et al., 2020; Sun et al., 2019). However, frequently this sludge which is laden with microplastics is applied to agricultural soils, creating a feedback loop that reintroduces microplastics into aquatic ecosystems via run-off or by seeping into groundwaters. The circular fate and transport of microplastics from biosolids into soils and later to water sources may pose significant environmental risks (Figure 4.8).

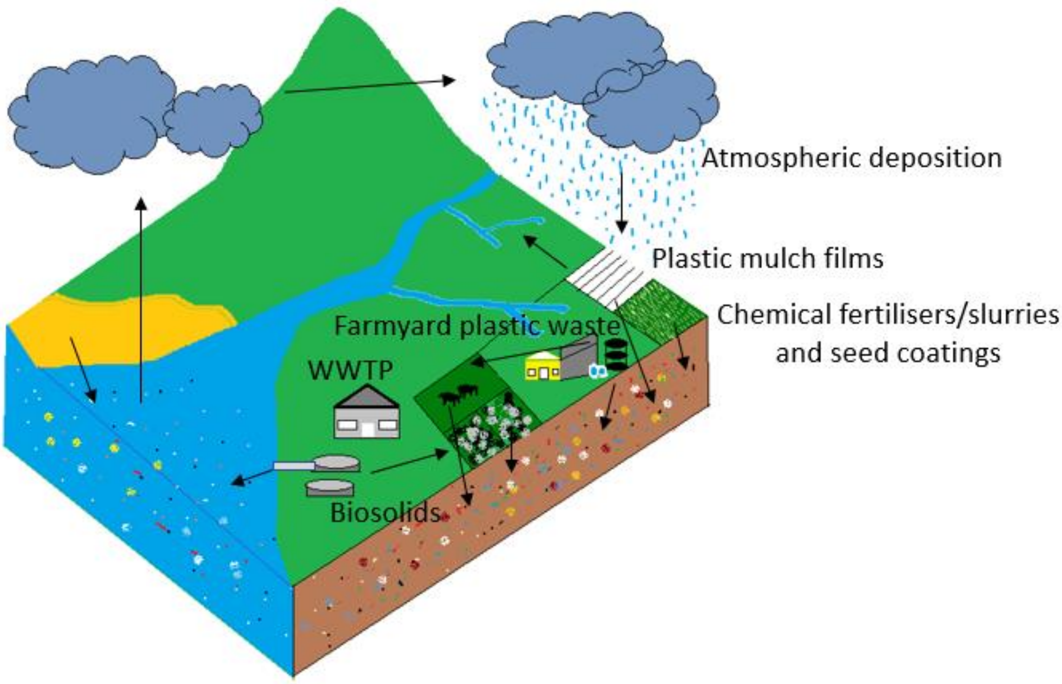

Figure 4.8: Schematic diagram of the potential direct and indirect sources of microplastics into agricultural soils and the circular fate of microplastics from terrestrial to aquatic ecosystems.

Once introduced into soil systems, microplastics can alter key physicochemical properties such as soil structure (Han et al., 2024), pH (Zhao et al., 2021) and water retention capacity (Wang et al., 2023), which may disrupt soil microbial communities responsible for nutrient cycling (Seeley et al., 2020), with potential knock-on effects to soil fertility and crop productivity (Rillig et al., 2019; Gao et al., 2022; Tunali and Rillig, 2025). Moreover, as microplastics migrate into freshwater or groundwater sources through leaching or surface run-off they can affect aquatic organisms, including those in the food webs, which are relied on by humans (Yuan et al., 2022; Nash et al., 2023 Rivas-Mena et al., 2024). Aquatic species can ingest microplastics, leading to physical blockage, oxidative stress, inflammatory responses or bioaccumulation and biomagnification of substances in tissues (Wright et al., 2013; de Sá et al., 2018; Miller al., 2020; Cao et al., 2023). This raises potential implications for overall ecosystem health, and human exposure via trophic transfer or microplastics present in drinking water supplies. A revision of biosolid application policies is necessary to prevent microplastic accumulation in and export from soils to aquatic environments, ensuring that wastewater treatment processes are improved to reduce microplastic load before land application.

4.6. Conclusions

The main conclusions from this study are:

1. This study finds microplastics in all sampled Irish agricultural soils (200 – 4899 MP items kg^{-1}), with higher concentrations in soils treated with biosolids and PMFs, though these are not the sole sources. The lack of standardised sampling and analysis methods continues to hinder the accurate detection and comparison across studies, underscoring the need for standardisation.

2. At present, there are no specific regulations or monitoring requirements for microplastics in biosolids in Ireland, or across the EU. This study highlights the importance of incorporating microplastic surveillance within the scope of the new EU Soil Monitoring Law, expected to be enforced by 2030. Microplastic contamination should be included as a key indicator in soil health reporting and formally integrated into Annex I, Part B of the proposed legislation. Member states should undertake national-level assessments of microplastic contamination in soils, focusing on high-risk areas such as land receiving frequent applications of biosolids and PMFs.

3. There is a need for regulatory oversight concerning the land application of biosolids and the use of PMFs (which in the case of PMFs has already been done to some extent). In addition to these measures, long-term research is required to understand the persistence and ecological impacts of PMFs in soils, in order to inform sustainable land management practices.

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Chapter 5: Microplastic-induced changes in soil chemistry, enzymatic activity and biomass in grassland mesocosms

5.1. ABSTRACT

Microplastics (plastic particles ≤ 5 mm) are widespread contaminants in agricultural soils that can affect soil properties, enzymatic activities, and plant growth, depending on their polymer type, size, shape and concentration. This study investigates the impact of polyethylene (PE), polypropylene (PP), and polyester (PES) microplastics on soil-plant systems using mesocosm experiments focusing on two common grassland species: *Lolium perenne* (*L. perenne* L.) and *Trifolium repens* (*T. repens*). Experiment A assessed the effects of varying concentrations (0.01, 0.1 and 0.5 % w/w) of PE, PP, and PES on *L. perenne*, while Experiment B evaluated PE and PP at 0.1 % in *T. repens* and *L. perenne* monocultures and mixed swards with *L. perenne*. Polyester at higher concentrations (0.5 %) enhanced *L. perenne* aboveground biomass in growth by 23%, while PE microplastics significantly reduced *T. repens* biomass. Polyethylene microplastics significantly increased soil pH compared to control soils. Enzymatic responses varied by polymer: PE reduced β-glucosidase activity by up to 39 %, while PP increased it. Acid phosphatase and arylsulfatase activities fluctuated depending on the polymer type, suggesting complex microplastic-microbe-soil interactions. The main findings from this study indicate that microplastics, depending on their characteristics can both enhance and impair soil and plant functions. From a practical perspective, this underscores the need for agricultural management strategies to consider microplastic contamination of soils. The disruption of soil pH and enzyme activity by specific polymers (such as PE) suggests potential impacts on nutrient cycling and soil fertility. Further research should be conducted to investigate microplastic effects under field conditions, in order to inform soil protection policies and regulatory frameworks such as the forthcoming European Union (EU) Soil Monitoring Directive.

5.2. Aims and Objectives

The main aims and objectives of this study were to:

1. Conduct two mesocosm experiments to examine the effects of microplastics commonly found in agricultural soils on the growth of two widespread Irish grassland species, *Lolium perenne* (Perrenial ryegrass) and *Trifolium repens* (White clover).

2. Evaluate the impacts of these microplastics on a sandy loam soil representative of grassland systems found in parts of Ireland.

5.3. Methodology

5.3.1. Experimental design

Two mesocosm experiments were conducted between July and November 2022 in a controlled polytunnel setting in the Research Centre for Freshwater and Environmental Studies at Dundalk Institute of Technology (53.98° N, 6.39° W). Experiment A consisted of a 4x3 factorial design, whereby four different types of microplastics (PE, PP, and PES of two different sizes), were incorporated into soil at three different concentrations (0.01, 0.1 and 0.5 %), with one model plant species (*L. perenne*), resulting in twelve treatments in addition to the control (without added microplastics). There were six replicates per treatment (72 mesocosms), plus an additional six mesocosms (controls containing no added microplastics), giving 78 mesocosms in total. Experiment B had a 3x3 factorial design, with three botanical compositions evaluated in the plant factor, including *L. perenne* and *T. repens* monocultures and an *L. perenne /T. repens* mixed sward. Plastic was also considered a factor, with three levels (PE, PP and no plastic) at one concentration for PE and PP (0.1 %). There were six replicates per treatment, resulting in 54 mesocosms in total (Table 5.1). Microplastics were added based on the fresh weight of the soil and exceeded any naturally occurring levels already present. *L. perenne* and *T. repens* were chosen because they represent the most commonly sold agricultural grass and clover seeds in Ireland. A sandy loam soil (sand 66.1 %; silt 28.6 %; clay 5.3 %) (pH 6.2; OM(LOI) 6.5 %; Total N: 5319 mg/kg; P (Morgan's) 14.1 mg/L; K (Morgan's): 557 mg/L) was used for both experiments, which was taken from a permanent grassland field, with a long history of pasture (grass-clover swards), located in (53.912, -6.725). Soils were then sieved using a 5 mm (mesh size) stainless steel sieve and air-dried before microplastic addition to soil. The trial lasted 102 days and temperature range during this trial was between 5.5 and 26.5 °C (Figure 5.2).

Table 5.1: Summary of the experimental designs employed for Experiment A and Experiment B.

	Experiment A	**Experiment B**
Plant	1 sward: *L. perenne* monoculture	3 swards: *L. perenne*, *T. repens* as monoculture and in a mixed sward
Microplastic type	4 types: PE (400 µm), PP (3000 µm), PES (3000 µm PES (250 µm)	3 types: PE, PP, no plastic
Concentration (of added microplastics – excluding the control)	3 concentrations: 0.01, 0.1, 0.5 %	1 concentration: 0.1 %
Factor design	4x3	3x3
Number of pots (units)	72 (including controls)	54 (including controls)

Figure 5.1: All pots set up in the polytunnel at the beginning of the trial.

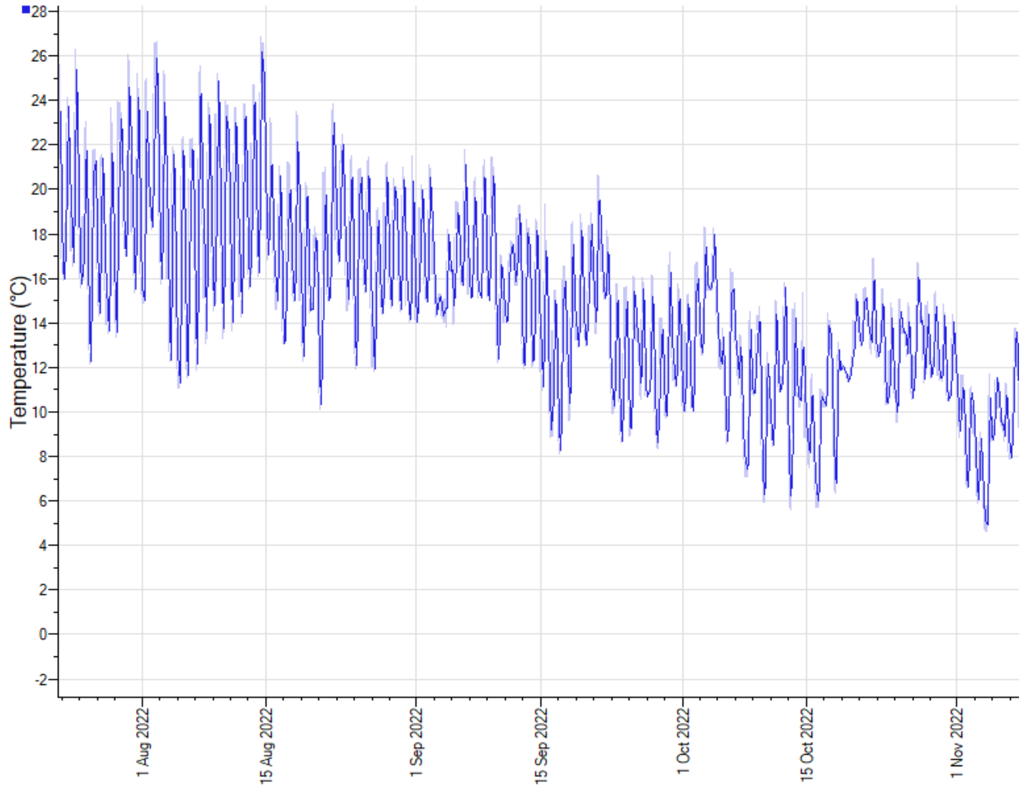

Figure 5.2: Temperature range inside the polytunnel during the pot trial experiment.

5.3.2. Microplastics description and preparation

All microplastics used for the experiments were fibre shaped to represent the most abundant type of microplastic recovered in the environment (Chubarenko et al., 2016; Gallagher et al., 2016), despite most previous controlled studies on microplastics in soils being carried out on beads and fragments. Moreover, fibres were selected as they represented the most dominant shape of microplastic detected in the agricultural soils from Chapter 4. Both PE and PP were selected for this study because they are the most commonly used polymers in agriculture (European Commission, 2021). Furthermore, PES fibres were chosen as these 'synthetic clothing fibres' are ubiquitously found in sewage sludge, or biosolids, which is a known contributor to microplastic contamination of agricultural soils (Yang et al., 2021; Zhou et al., 2020), and water (Gunaalan et al., 2023; Kanhai et al., 2017). In Ireland, 98 % of sewage sludge is used as agricultural fertiliser (Gavigan et al., 2020; Irish Water, 2015). The microplastic concentration range was selected based on publications reporting these concentrations as the baseline level of microplastic contamination in some agricultural soils (De Souza Machado et al., 2018; Fuller and Gautam, 2016; Xu et al., 2020; Brouwer et al., 2024). Microplastics were sourced from ©Goonvean Fibres (Devon, England). The following polymer types and sizes of microplastics were selected: PE (400 µm), PP (3000 µm), PES (250 µm) and PES (3000 µm). Microplastics were sourced fresh and initially did not represent the aged-microplastics that are typically found in the environment; therefore, they were rinsed thoroughly x3 with deionised water to remove any chemicals from their surface that may interfere with the soil chemistry. In addition to this, microplastics were dried in the oven at 40 °C for 6 hours and then microwaved for 1 minute to minimise any potential microbial contamination on their surface.

5.3.3. Microplastic addition to soil mesocosms

Approximately 1050 g of soil was used for each mesocosm, and six mesocosms were made at one time for each treatment by adding microplastics to 6300 g soil in a large container and mixing thoroughly for 10 minutes by manually stirring with a stainless-steel spoon to ensure a homogenous mixture (Yu et al., 2021). The same stirring was carried out for the controls. Soils spiked with microplastics were then transferred into 1.5 L PP plastic pots that were 15 cm (top diameter), 10 cm (bottom diameter) and 12.4 cm (height). The water holding capacity (WHC) of the soils were pre-determined, and thereafter, deionised water was added to each pot and kept to a minimum of 60 % WHC for the length of the experiment (Boots et al., 2019; Moorberg and Crouse, 2021).

In total, 20 *L. perenne* seeds and 63 *T. repens* seeds were added to each pot of the mixed swards, and in monocultures only 20 *L. perenne* seeds were added, and 63 in *T. repens* monocultures, these figures were extrapolated from typical agricultural field application rates. The seed application rate of *L. perenne* (variety – Bowie) to typical grassland soils in Ireland is 32-40 kg^{-1} ha^{-1} (Grogan, 2012), and 1-2 kg^{-1} ha^{-1} for sowing *T. repens* (variety – Iowa) (Germinal Ireland, 2022). All seeds were weighed individually and carefully chosen for the experiment (damaged or smaller seeds were excluded). Mesocosms were set up in a randomised design in the polytunnel, using a series of points and a random number generator. Mesocosms were rearranged using similar randomisation at three times during the length of the experiment.

5.3.4. Seedling emergence and plant biomass

The emergence of *L. perenne* and *T. repens* were assessed according to guidelines in the OECD Test 208: on Terrestrial Plant Seedling Emergence and Growth Test (OECD, 2006). On day 50, a first harvest was performed by cutting each plant 2 cm from their root with the use of a ruler and scalpel/scissors. All plant biomass was weighed, dried in an oven at 95 °C for 15 hours, and then re-weighed to determine the dry biomass yield per pot (Beecher et al., 2013). These measures were repeated for the second harvest on day 102 at the end of the experiment.

5.3.5. Soil pH

Soil samples were sieved using a 2 mm stainless steel sieve and were oven dried at 40 °C for 48 hours in preparation of soil pH testing in accordance with procedures carried out by Teagasc (2020). Aliquots of 10 g soil were weighed and transferred to 50 ml beakers. Twenty ml of deioniwed water was added to each beaker and samples were mixed using a glass rod and left to sit for 5 min. A pH meter (Mettler Toledo™) was calibrated using buffers with pH values of 4.0, 7.0 and 10.0. The electrode probe was rinsed down with deionised water, dried with a paper towel and submerged into each sample. The pH reading was taken after the electrode produced a stable reading from the sample. This was performed in duplicate per pot. The electrode probe was rinsed with deionised water in between samples.

5.3.6. Estimation of soil enzyme activity

On day 102, mesocosms were dissembled and subsamples from each experimental unit were stored at -20 °C prior to conducting enzyme activity assays. They were stored for a period of of two weeks before assays were performed. While analyzing fresh soil samples provides the most accurate assessment of enzyme activity, this was not feasible due to time constraints, logistical challenges, and the large number of samples. However, the relative differences among treatments remained consistent during storage. According to Peoples and Koide, (2012) it is acceptable for storage methods to reduce enzyme activity, as long as the relationships between treatments and soils remain unaffected. The activity of β-glucosidase, acid phosphatase and arylsulfatase were determined spectrophotometrically by measuring the amount of *p*-nitrophenol released by *p*-nitrophenyl- β-glucopyranoside, *p*-nitrophenyl phosphate and *p*-nitropheyl sulfate (Chen et al., 2021). For β-glucosidase activity, 0.5 g of dried soil, 0.2 ml of toluene, 0.5 ml of 25 mM *p*-nitrophenyl-β-glucopyranoside and 2 ml of modified universal buffer (MUB) were used and incubated at 37 °C for 1 hour. The MUB was composed of Tris, boric acid, maleic acid, sodium hydroxide (NaOH), hydrochloric acid (HCl) and deionised water and made up to a pH of 6.5. The reaction was terminated by adding 2 ml of 0.5 M

Tris buffer (pH 12) and 0.5 ml of 0.5 M $CaCl_2$. For acid phosphatase activity, 0.5 g of dried soil, 0.2 ml of toluene, 0.5 ml of 115 mM *p*-nitrophenyl phosphate and 2 ml of modified universal buffer (MUB) were used and incubated at 37 °C for 1 hour. For arylsulfatase activity, 0.5 g of dried soil, 0.2 ml of toluene, 0.5 ml of 25 mM *p*-nitrophenyl sulfate and 2 ml of modified universal buffer (MUB) were used and incubated at 37 °C for 1 hour. The reactions for acid phosphatase and arylsulfatase activities were terminated by the addition of 2 ml of 0.5 M and 0.5 ml of 0.5 M $CaCl_2$. All assays were carried out in glass test tubes and after reactions were interrupted, the mixtures were filtered using filter paper (©Satorius, 125 mm, 5-8 µm pore size) and absorbance was detected at OD_{400} using a UV/Vis spectrophotometer (JENWAY® 6300). Assays were performed in duplicate per experimental unit (pot/mesocosm) with appropriate controls and enzyme activities (µmol PNP g^{-1} h^{-1}) were calculated with reference to a *p*-nitrophenol standard curve (range: 0, 0.25, 0.5, 0.75, 1 mM).

5.3.7. Statistics and data analysis

All data were expressed as mean ± standard error (SE), and all statistical analyses were conducted using Minitab® 21. 1. 1. Data normality was assessed using the Komogorov-Smirnov test, which is appropriate for larger sample sizes (N = ≥ 50). For data that did not meet normality assumptions, non-parametric Kruskal-Wallis tests were used to evaluate significant differences between dependent and independent variables for each treatment individually ($p \leq 0.05$). For Experiment A, a Generalised Linear Model (GLM) was applied to data that met the assumptions of normality. Plastic type and concentration were included as fixed factors, and the comparison between control and treated groups (‘Control_vs_treated’) was nested within these factors. Two-way interactions were included to evaluate potential combined effects. Statistical significance was determined at $p \leq 0.05$. For Experiment B, a Two-way ANOVA was performed on normally distributed data to assess treatment effects for each response variable. Where significant differences were detected ($p \leq 0.05$), Tukey’s post hoc tests were conducted to compare group means.

5.4. Results

5.4.1. Microplastics effects on seedling emergence and plant biomass

In experiment A, there were no significant differences in seedling emergence between the control and treated soils on average; however, plastic type had a significant effect ($p = 0.028$), with PES (3000 µm) microplastics reducing seedling emergence by up to 15 % compared to other microplastic treatments. At Harvest 1, microplastic concentration significantly affected plant biomass, with soils containing 0.1 % microplastics increasing the growth of *L. perenne* compared to lower (0.01 %) and higher (0.5 %) concentrations of microplastic ($p = 0.016$). By Harvest 2, microplastic-treated soils showed significantly greater *L. perenne* biomass than control soils ($p = 0.008$). Notably, control soils and soils treated with PES (3000 µm) (0.5 %) produced the lowest and highest yields on Harvest 2, with 1.14 ± 0.04 and 1.49 ± 0.08 (mean ± SE) biomass (g . pot^{-1} DW), respectively. Although PES (3000 µm) across combined concentrations decreased seedling emergence, there was a 23 % increase in plant biomass at Harvest 2 with soils containing 0.5 % PES (3000 µm) microplastics compared to other microplastic treatments (Table 5.2).

Table 5.2: Microplastics effects on seedling emergence (%) and plant biomass (g^{-1} DM^{-1}), of *L. perenne*, means, standard errors and p-values are shown.

Polymer	Concentration (w/w)	Seedling emergence (%)	Biomass (g . pot^{-1} DW) Harvest 1	Biomass (g . pot^{-1} DW) Harvest 2
Control (no plastic)	(0 %)	92.50 ± 2.14	3.31 ± 0.19	1.14 ± 0.04
PE (400 µm)	(0.01 %)	93.33 ± 3.57	3.26 ± 0.06	1.31 ± 0.06
	(0.1 %)	92.50 ± 2.14	3.28 ± 0.12	1.22 ± 0.08
	(0.5 %)	88 ± 2.55	3.60 ± 0.25	1.37 ± 0.07
PP	(0.01 %)	100 ± 0	3.16 ± 0.22	1.26 ± 0.06

(3000 µm)	(0.1 %)	92.50 ± 2.14	3.79 ± 0.14	1.40 ± 0.08
	(0.5 %)	85.83 ± 2.38	3.30 ± 0.15	1.27 ± 0.05
PES (3000 µm)	(0.01 %)	84.17 ± 7.23	3.41 ± 0.20	1.24 ± 0.06
	(0.1 %)	85.00 ± 2.58	3.88 ± 0.21	1.46 ± 0.06
	(0.5 %)	84.17 ± 3.75	3.35 ± 0.16	1.49 ± 0.08
PES (250 µm)	(0.01 %)	84.17 ± 2.00	3.24 ± 0.20	1.47 ± 0.08
	(0.1 %)	85.00 ± 3.87	3.75 ± 0.21	1.44 ± 0.09
	(0.5 %)	90.83 ± 2.00	3.45 ± 0.29	1.19 ± 0.06
Control v Treated p-values		0.332	0.470	0.008
Main effects p-values (plastic)		0.028	0.748	0.301
Main effects p-values (concentration)		0.184	0.016	0.431
Interaction p-values		0.083	0.395	0.009

In experiment B, microplastics had no effect on seedling emergence, but significantly influenced plant biomass at Harvest 1 ($p = 0.049$). Irrespective of the plant type, the addition of PE microplastics significantly reduced plant biomass compared to PP and control soils. PP microplastics enhanced growth in *L. perenne* monocultures, mixed swards and *T. repens* monocultures by over 15 %, 17 % and 18 %, respectively, compared to PE-treated soils. In *T. repens* monocultures, PE addition reduced plant biomass by

20.7 % when compared to the control. Moreover, in the mixed sward, plant biomass was reduced by 17.2 % in PE treated soils. By Harvest 2, microplastics did not significantly affect the growth of forage, however, the results were trending towards significance (p = 0.060). The addition of PE microplastics in the mixed sward decreased plant biomass by 32.6 % in comparison to the control (Table 5.3).

Table 5.3: Microplastics effects on seedling emergence (%) and plant biomass (g . pot^{-1} DW) of *L. perenne*, means, standard errors and p-values are shown.

Treatment	Seedling emergence (%)	Biomass (g . pot^{-1} DW) Harvest 1	Biomass (g . pot^{-1} DW) Harvest 2
WC Control (no plastic)	54.57 ± 3.91	2.60 ± 0.06	0.99 ± 0.15
WC PE (0.1 %)	44.09 ± 3.34	2.06 ± 0.14	0.92 ± 0.05
WC PP (0.1 %)	42.20 ± 2.74	2.51 ± 0.23	0.87 ± 0.05
WC_PRG Control (no plastic)	59.15 ± 1.36	3.59 ± 0.30	1.39 ± 0.18
WC_PRG PE (0.1 %)	53.05 ± 3.64	2.87 ± 0.18	0.94 ± 0.007
WC_PRG PP (0.1 %)	47.56 ± 2.15	3.47 ± 0.20	1.60 ± 0.09
PRG Control (no plastic)	92.50 ± 2.14	3.31 ± 0.19	1.14 ± 0.04
PRG PE (0.1 %)	92.50 ± 2.14	3.28 ± 0.12	1.22 ± 0.08
PRG PP (0.1 %)	92.50 ± 2.14	3.88 ± 0.21	1.40 ± 0.08
Main effects p-values (plastic)	0.158	0.049	0.060
Main effects p-values (plant)	≤ 0001	≤ 0001	≤ 0.001

WC = *T. repens*, WC_PRG = *T. repens* / *L. perenne* sward, PRG = *L. perenne*

5.4.2. Microplastics effects on soil pH

In experiment A, microplastics significantly altered soil pH in comparison to the control ($p \leq 0001$), and it was evident that the addition of PE microplastics increased soil pH up to 7.08 in higher concentrations (0.5 %), in contrast to control soils with a mean pH of 6.5. Concentration contributed towards a significant difference in soil pH among treatments and particularly when higher concentrations of PE, PES (250 µm) and PP, were added to soils the pH increased ($p \leq 0001$) (Figure 5.3). In experiment B, the effects of microplastics on soil pH were trending towards significance ($p = 0.051$). Soils containing *T. repens* monocultures and PE microplastics (0.1 %) increased soil pH (7.5) in comparison to control soils (6.4) and soils with PP (0.1 %) (5.6) (Figure 5.4).

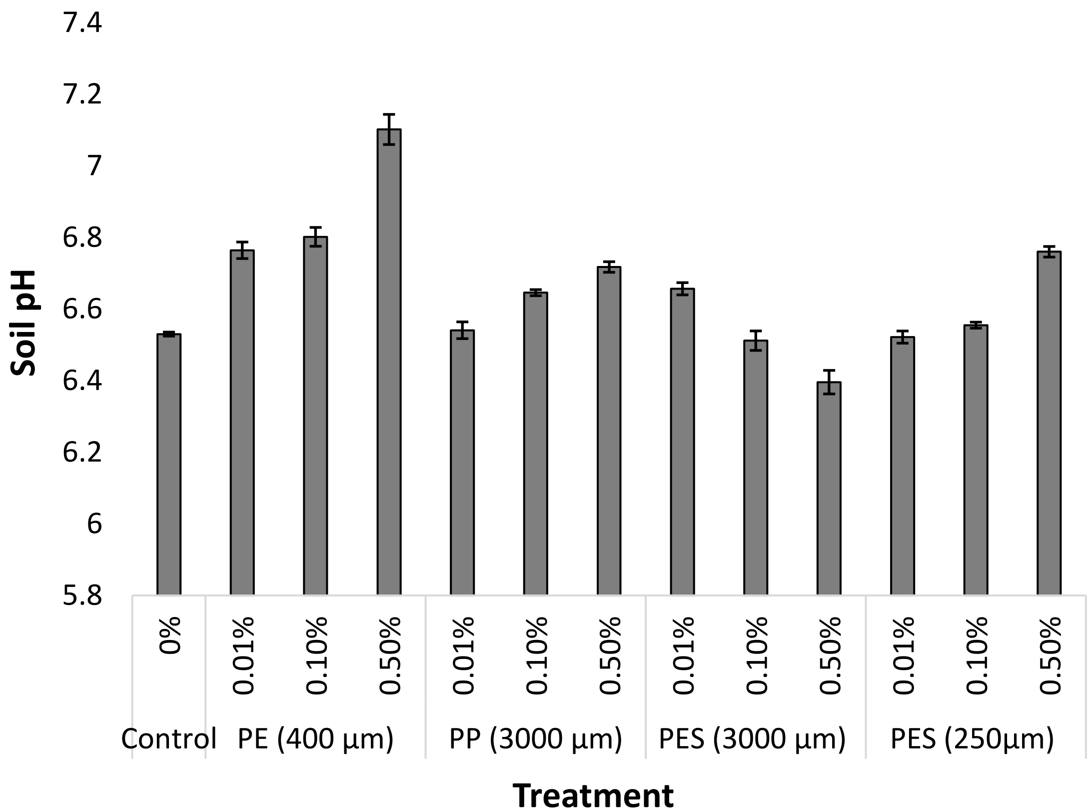

Figure 5.3: The effects of microplastics on soil pH in Experiment A

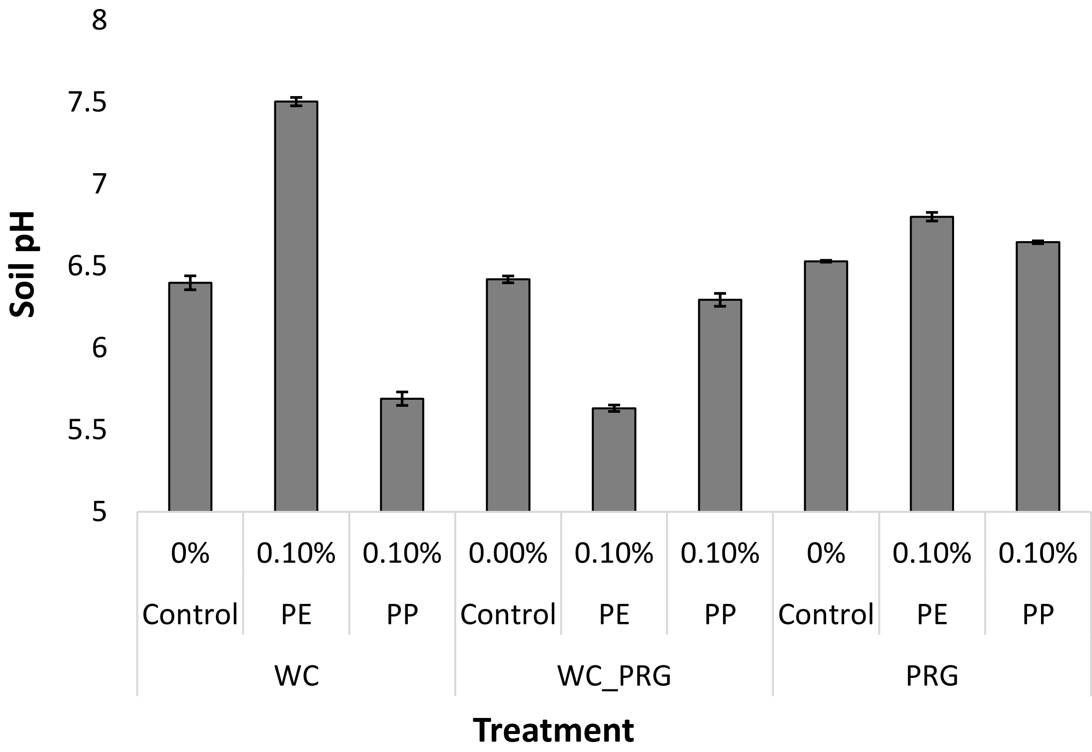

Figure 5.4: The effects of microplastics on soil pH in Experiment B.

5.4.3. Microplastics effects on soil enzyme activity

In experiment A, microplastics significantly altered the activity of the three soil enzymes measured in this study. The effects of treatments on the activity of β-glucosidase ($p \leq 0001$), were polymer and concentration dependent. For example, soils spiked with PP microplastics showed a decreasing trend in β-glucosidase activity as concentration increased, however, this trend was not evident in soils treated with PES (250 µm). Despite this, on average, soils exposed to PP promoted the highest activity of β-glucosidase (706.29 ± 89.28 µmol PNP g^{-1} h^{-1}) in comparison to soils with PE microplastics added (431.04 ± 31.96 µmol PNP g^{-1} h^{-1}) and control soils (564.31 ± 31.96 µmol PNP g^{-1} h^{-1}) ($p \leq 0001$). Overall, when combining concentrations, the activity of β-glucosidase was inhibited by 38.9 % and 23.5 % in soils with PE in comparison to soils with PP and the control soils, respectively (Figure 5.5). In experiment B, β-glucosidase activity in control soils was increased in soils containing PE and PP microplastics ($p \leq 0001$). PE and PP reduced β-glucosidase activity in *T. repens* monocultures by 33.1 % and 30.1 %, respectively compared to the control. In mixed swards, activity was reduced by 36.3 % (PE) and 50.2 % (PP) compared to controls (Figure 5.6).

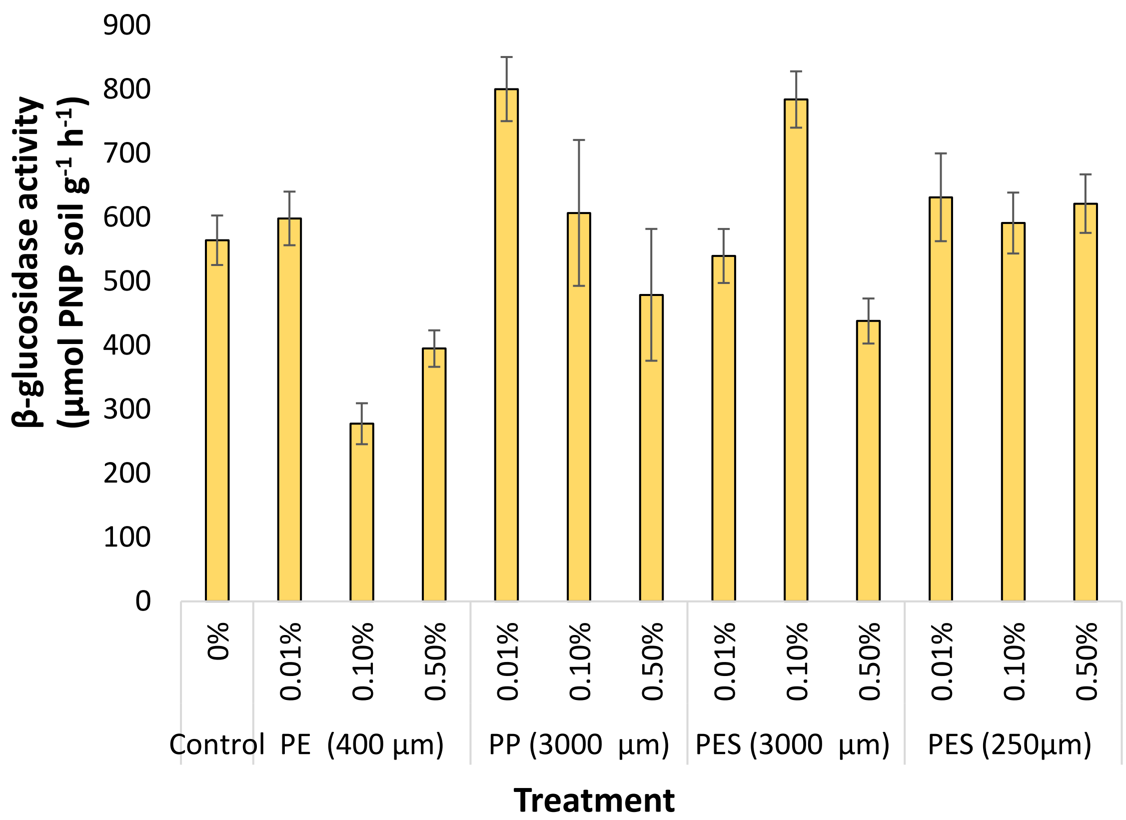

Figure 5.5: The effects of microplastics on β-glucosidase activity in soil in Experiment A.

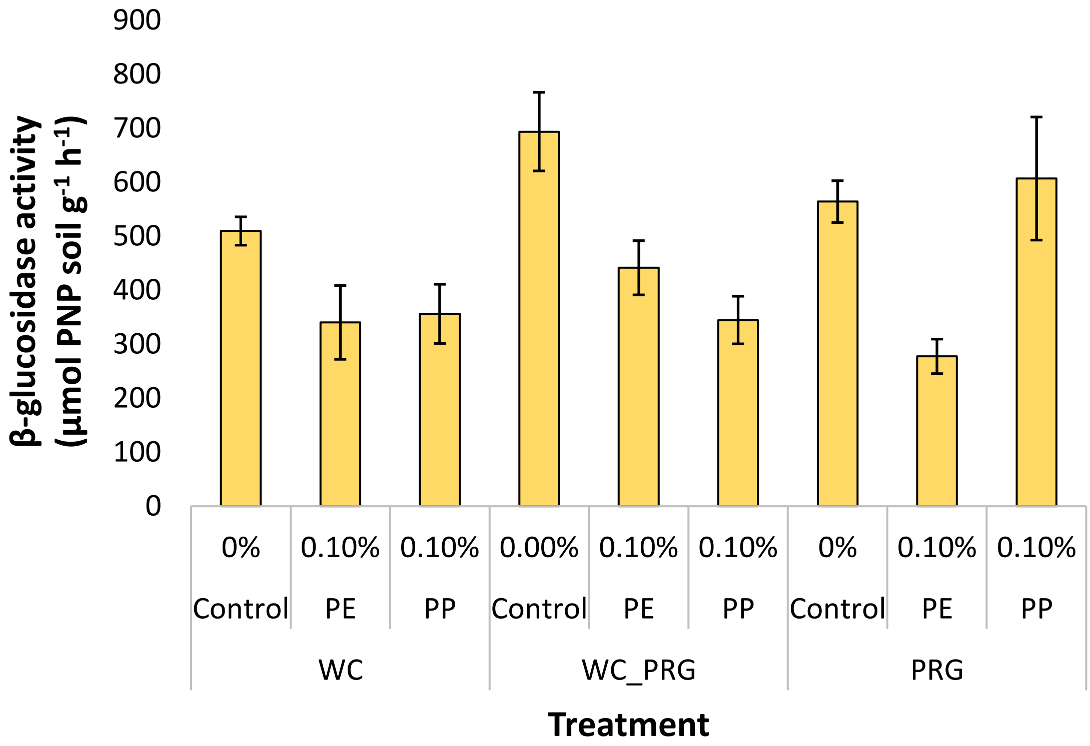

Figure 5.6: The effects of microplastics on β-glucosidase activity in soil in Experiment B.

In experiment A, acid phosphatase activity was significantly reduced from 961.03 ± 41.35 µmol PNP g^{-1} h^{-1} in soils with microplastics to 1078.8 ± 3.05 µmol PNP g^{-1} h^{-1} in control soils ($p = 0.037$). Treatments with PP microplastics had the highest acid phosphatase activity with 1087.78 ± 3.05 µmol PNP g^{-1} h^{-1} released which was significantly higher than treatments with PES (3000 µm) (836.52 ± 30.9 µmol PNP g^{-1} h^{-1}), representing a 24 % increase ($p \leq 0001$). Concentration of microplastics had a significant effect on acid phosphatase activity, and in particular, PE (0.1 %), PES (3000 µm) (0.1 %) and PE (0.5 %) were significantly lower than other treatments, including the control ($p = 0.003$). Lower concentrations (0.01 %) across all types of microplastics made no significant impact on acid phosphatase activity when compared with controls (Figure 5.7). In experiment B, the addition of PE significantly reduced the activity of acid phosphatase in soil (656.2 ± 28.7 PNP g^{-1} h^{-1}) compared to addition of PP microplastics (869.9 ± 28.7 PNP g^{-1} h^{-1}) and the control soils (899.4 ± 28.7 PNP g^{-1} h^{-1}), with control soils containing 27 % more acid phosphatase activity than PE soils (Figure 5.8).

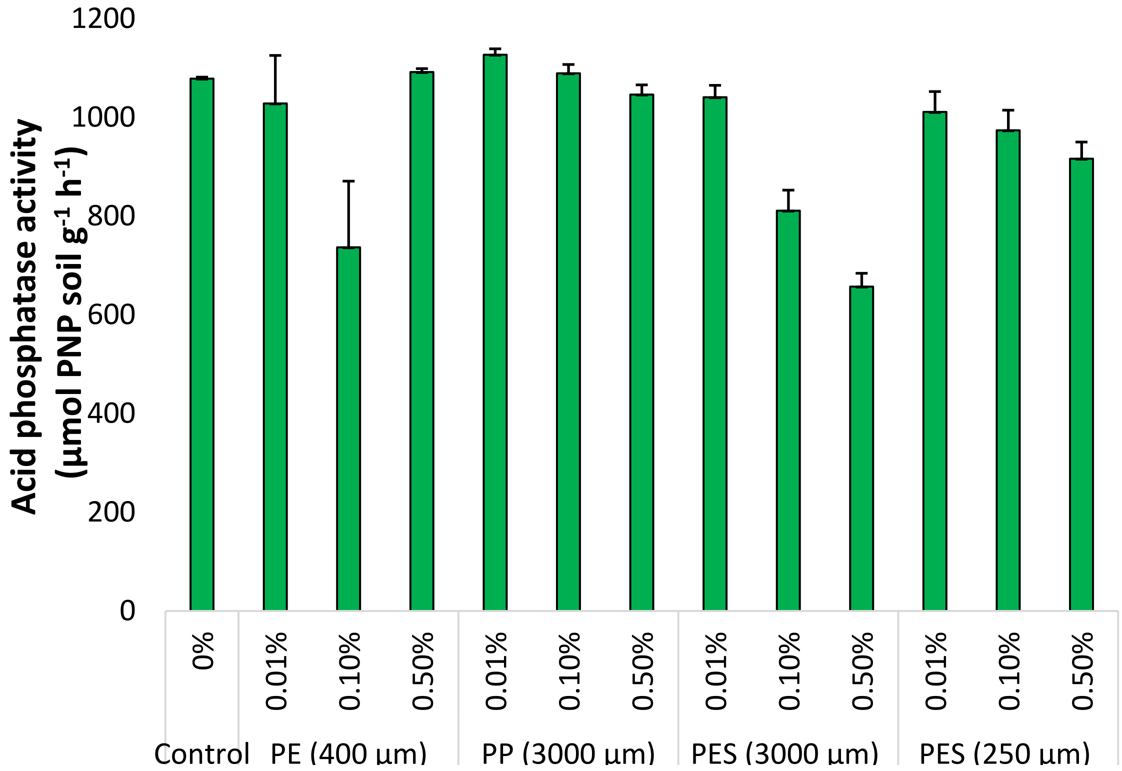

Figure 5.7: The effects of microplastics on acid phosphatase activity in soils in Experiment A.

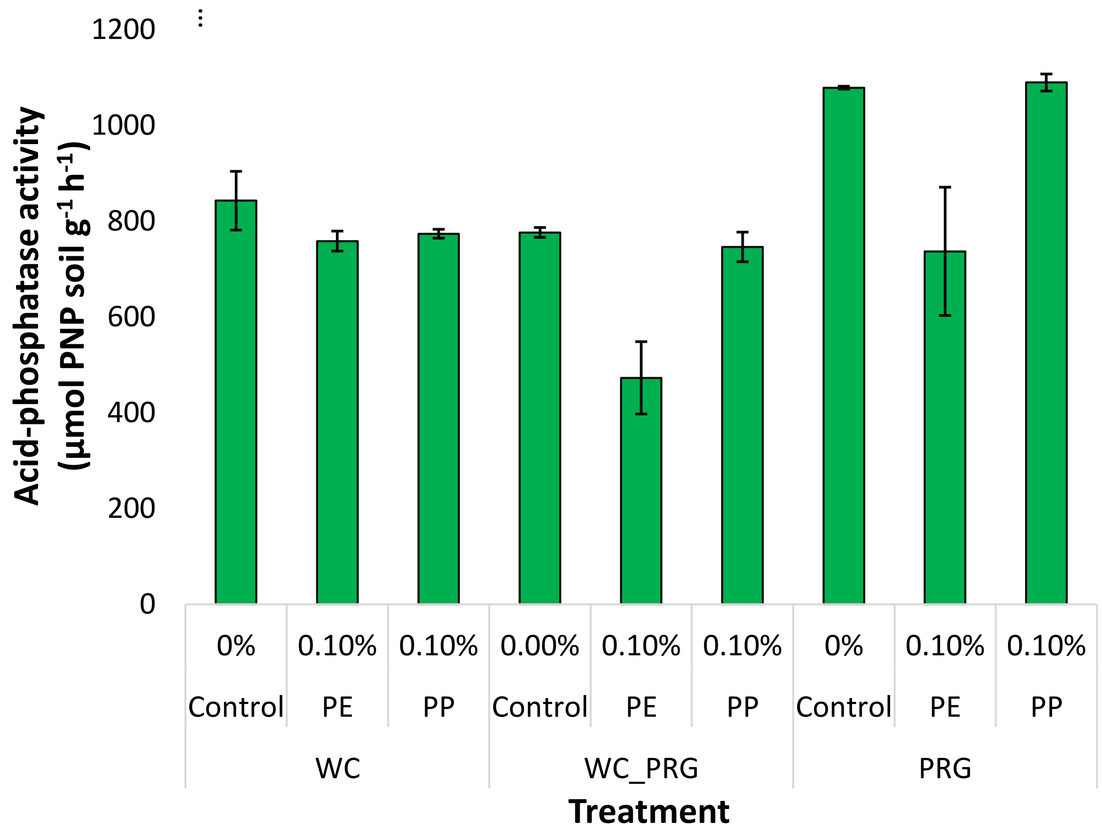

Figure 5.8: The effects of microplastics on acid phosphatase activity in soils in Experiment B.

In experiment A, combined microplastic treatments did not significantly affect arylsulfatase activity ($p = 0.060$), but polymer-specific differences were significant ($p \leq$ 0001). Polyester (250 µm) (1214.75 ± 71.5 µmol PNP g^{-1} h^{-1}) and PP (1106 ± 76.6 PNP g^{-1} h^{-1}*)* increased activity compared to controls (787.33 ± 3.05 µmol PNP g^{-1} h^{-1}). In contrast, PES (3000 µm, 0.5 %) and PE (0.5 %) had the lowest activity (287.7 and 466.6 µmol PNP g^{-1} h^{-1}, respectively) (Figure 5.9). Overall, arylsulfatase was most negatively affected in treatments of PES (3000 µm) (0.5 %) (287.7 ± 76.6 PNP g^{-1} h^{-1}) and PE (0.5 %) (466.6 ± 28.7 PNP g^{-1} h^{-1}). In experiment B, there were no significant differences between arylsulfatase activity in treatments applied with microplastics and the control soils ($p = 0.097$) (Figure 5.10).

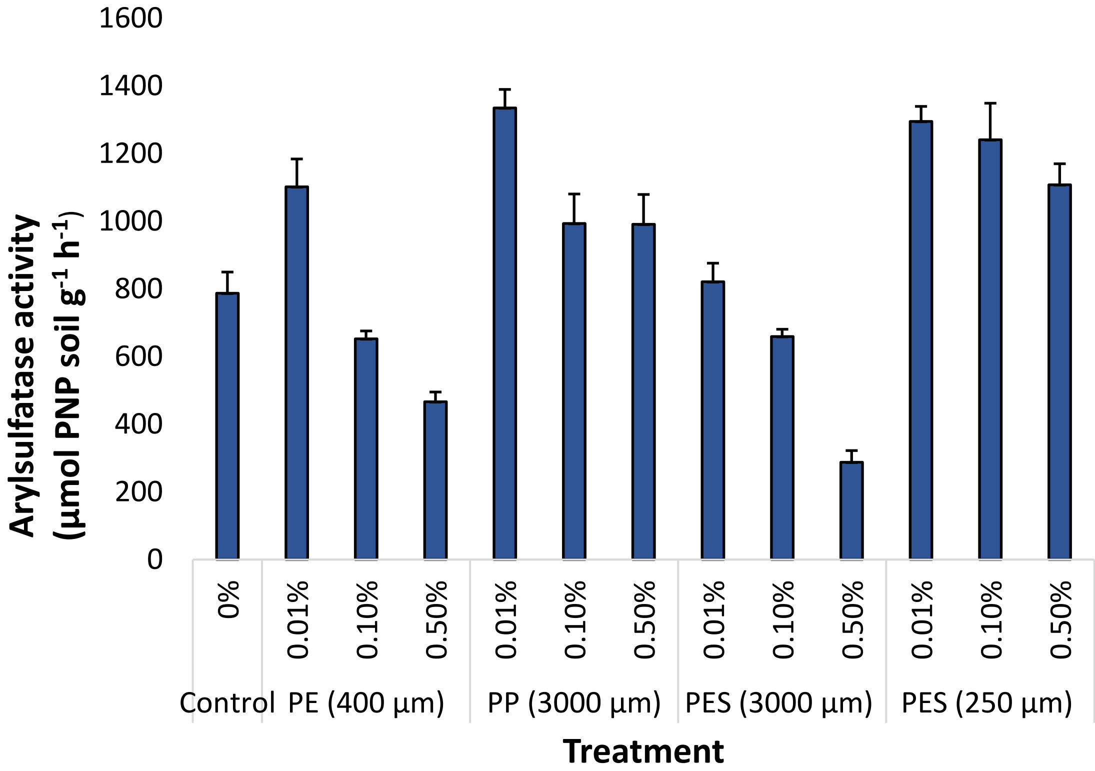

Figure 5.9: The effects of microplastics on arylsulfatase activity in soils in Experiment A.

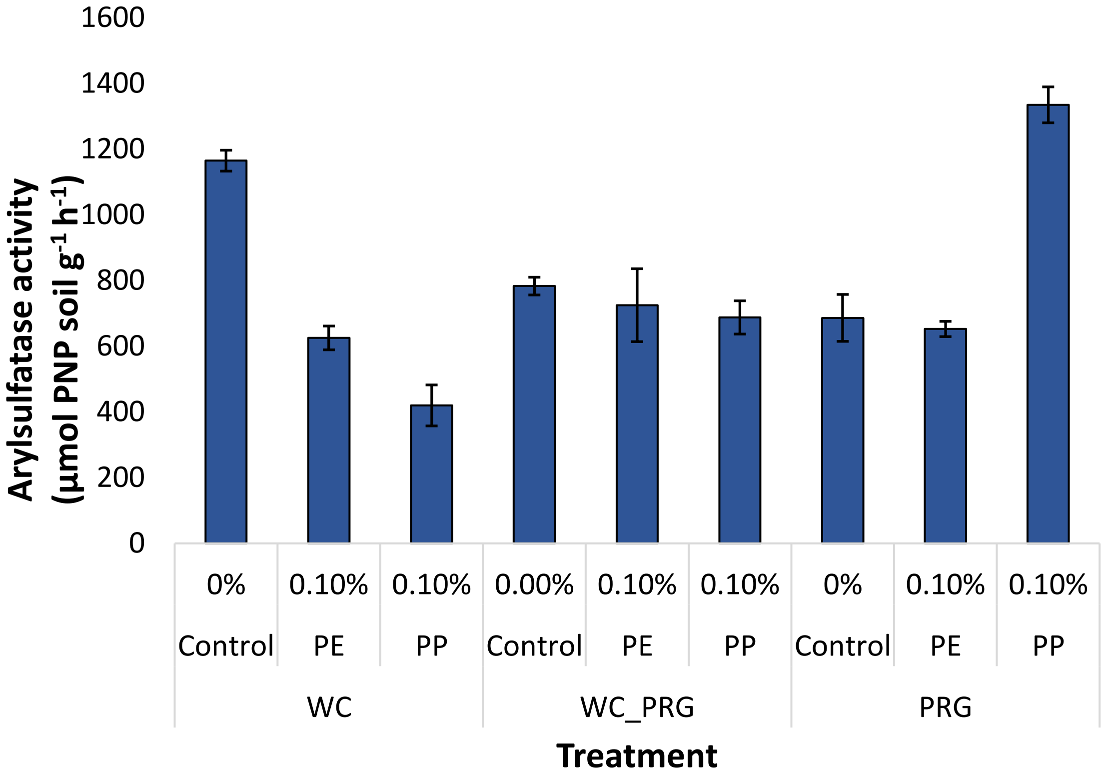

Figure 5.10: The effects of microplastics on arylsulfatase activity in soils in Experiment B.

5.5. Discussion

This study demonstrates that microplastics can significantly influence plant growth, soil properties and key enzyme activities. These effects are not uniform, highlighting the complexity of microplastic-soil-plant interactions.

5.5.1. Microplastics and plant biomass

The addition of microplastics, especially PES (3000 μm) fibres, significantly increased *L. perenne* biomass at Harvest 2, with a 23 % increase, compared to controls. These findings align with previous studies, where PES fibres at 0.4 % and 0.2 % significantly increased shoot biomass in *Daucus carota* (Lozano et al., 2021) and total biomass in *Allium fistulosum* (De Souza Machado et al., 2018). Microplastics can influence plant growth by multiple indirect mechanisms. Depending on the shape of the microplastics, different shapes can behave differently in soil (Zhang et al., 2022) and exert different effects (Lozano et al., 2021; Sun et al., 2022). This study only used fibre-shaped microplastics, which are known to integrate more readily into soil aggregates (De Souza Machado et al., 2018; Ingraffia et al., 2022; Zhang et al., 2019; Zhang and Liu, 2018). Fibres can enhance water-holding capacity (De Souza Machado et al., 2018; Lozano et al., 2021), reduce bulk density, and increase porosity (De Souza Machado et al., 2018; Zhang et al., 2019). These changes can lower resistance to root growth and often lead to improved plant growth, although this is not always the case (Stirzaker et al., 1996; Strock et al., 2021).

However, this stimulatory effect was not observed in *T. repens*. In experiment B, the addition of PE microplastics reduced plant aboveground biomass in *T. repens* monoculture and the mixed sward. The direct negative effects of PE on the growth of *T. repens* are unknown, but may be a result of a variety of indirect mechanisms by which PE affects other soil properties which feedback to the plant system. Moreover, the different physiology of *T. repens* and *L. perenne* may be somewhat responsible for the negative effects of PE microplastics on the growth of *T. repens,* but not on *L. perenne*. For example, *T. repens* develops a shallow root system that facilitates its symbiotic relationship with *Rhizobium* bacteria housed in root nodules. In contrast, *L. perenne* forms a deeper, fibrous root system that enhances access to water and nutrients at greater soil depths. As a result, *L. perenne* contributes more significantly to soil structural stability compared to *T. repens* (Ren et al., 2017). In this experiment, the reduced aboveground biomass of *T. repens* may potentially be explained by disruption the root-rhizobium interface or leaching of PE additives that may alter soil microbes, including rhizobia, which has been demonstrated in other studies (Singh and Singh, 2022). Limited existing studies on microplastic impacts on *T. repens* (Li and Xiao, 2024), underscore a need for more targeted research on the effects of microplastics on nitrogen-fixing legumes.

5.5.2. Microplastics effects on soil pH

In experiment A, PE microplastics significantly increased soil pH in comparison to the control soils and other treatments. In experiment B, the results were trending towards significance with PE slightly increasing soil pH on average. Soil pH is a key factor that influences a range of other soil properties, as it affects numerous processes such as nutrient availability (Barrow and Hartemink, 2023; Penn and Camberato, 2019), the composition of microbial communities (Bartram et al., 2014), and, consequently, enzyme activity. One possible explanation for the observed increase in pH is that PE may leach additives into the soil (Fajardo et al., 2022), potentially disrupting and altering soil microbial communities (Cao et al., 2023). However, identifying which substances leach from microplastics is challenging due to the variety of chemicals they may contain (Do et al., 2022; Gulizia et al., 2023). There are approximately 16, 000 known chemicals in plastics, with only 6 % currently subject to international regulation (Wagner et al., 2024) and more than 4,200 concerning as they are persistent, mobile, bioaccumulative and/or toxic (Wagner et al., 2024). While plastic polymers do not possess an inherent pH, their interaction with environmental factors can influence the pH of surrounding matrices, such as soil and water. In soils, microplastics have been shown to increase soil pH (Zhou et al., 2021); however, in marine environments they have been associated with a decrease in water pH, which highlights the complex and context-dependent effects of microplastics across different ecosystems (Romera-Castillo et al., 2023).

In this study, the increase in pH may be associated with PE altering soil microbial diversity, like the suppression of nitrifying bacteria. Such a reduction could influence N-transformation potentially reducing nitrification rates thereby decreasing the production of H^+ ions, which may contribute to an increase in soil pH. PE addition has been reported to lead to a decline in the relative abundance of Acidobacteria (Fei et al., 2020), which are one of the richest phyla found in soils (Zhang et al., 2014). Some have the ability to produce organic acids (Sindhu et al., 2022) as metabolic by-products, which can contribute to a release of H+ that decreases soil pH (Lauber et al., 2009; Wang et al., 2019). However, the inhibition of these bacteria could potentially have opposite effects therefore increasing soil pH. Qiu et al., (2023) found that PE (0.5 %) reduced the relative abundance of Acidobacteria, which they related to an increase in soil pH. Moreover, Wang et al., (2024) found that Acidobacteria were negatively correlated with the abundance of PE microplastics.

The plant species *L. perenne* is tolerant to both acidic and alkaline soils, and can grow in soils with a pH range between 5.5 and 7.5 (Hannaway et al., 1999), however, maintaining grassland soils at a pH of 6.5 is recommended (Teagasc, 2022). In this study, higher concentrations of PE led to an overall significant increase in soil pH. Further increases in soil pH could lead to an alkaline soil forming, and although many plants are tolerant to surviving in alkaline soil conditions, they may struggle to thrive in highly alkaline soils and show signs of nutrient deficiency (Barrow and Hartemink, 2023; Msimbira and Smith, 2020). Further work, using a longer exposure period than the current study is required determine if continued increases in soil pH occur over-time after the addition of PE microplastics to soil, and if microbial community structure is affected by microplastics and influences soil pH.

5.5.3. Microplastic effects on soil enzyme activity

Microplastics significantly influenced soil enzyme activities but the effects varied with polymer type and concentration. In both experiments, the addition of PE microplastics led to a decrease in β-glucosidase activity compared the control soils. In contrast, in Experiment A, the addition of PP microplastics increased β-glucosidase activity, compared to the control with lower concentrations of PP promoting higher activity, but activity decreased as the concentration of PP increased. β-glucosidase is involved in the final degradation of cellulose in soil and is responsible for the final step in the hydrolysis of lignocellulose which converts cellobiose into glucose (Zhang et al., 2020). It is an important component of cellulose enzyme complex that plays a significant role in the soil carbon cycle, providing an important energy source in the form of glucose for microorganisms (Dai et al., 2021), and therefore is often used as an indicator for monitoring biological soil quality (De Almeida et al., 2015). It may be that those enzymes were inhibited earlier in the process, or that glucose may have been released earlier and used up. It is unknown what caused the negative and positive responses to β-glucosidase in this study. Again, PE and PP microplastics may have caused shifts in microbial community structures by changing soil physical properties (Han et al., 2024), by providing microbial habitat (Kublik et al., 2022), binding to functional groups (Aralappanavar et al., 2024), or from releasing certain additives into the soil (Kong et al., 2012; Omidoyin and Jho, 2023). Polyethylene microplastics are considered a source of persistent organic pollutants in soil and can be toxic to soil microbes (Tziourrou and Golia, 2024). Moreover, the effects may likely be connected to the alterations in soil pH that were observed. Recent publications have demonstrated microplastics can negatively affect β-glucosidase in soils. For example, Shah et al., (2023) reported the addition of PE and other polymers reduced the activity of β-glucosidase d in soil, although this did not adversely affect the growth of *Glycine max* in those soils. Qiu et al., (2023) linked PE-induced changes in enzyme activity to alterations in microbial biomass and bacterial community composition, ultimately leading to a decline in microbial carbohydrate metabolism.

In experiment A, PE and PES reduced the activity of acid phosphatase and in experiment; B PE reduced the activity of acid phosphatase. Acid phosphatases produced by plants and microbes play a fundamental role in recycling soil phosphorus (Park et al., 2022). They catalyse the cleavage of phosphate bonds, releasing the phosphate through hydrolysis for uptake by plants (Anand and Srivastava, 2012). Acid phosphatase is influenced by soil pH and is most active in acidic conditions (Cai et al., 2021). In this study, the increase in soil pH may be associated with the reduced activity of acid phosphatases. Generally, an increase in soil pH tends to impair the function of phosphatases as microbial populations in soil are sensitive to changes in pH (Dick et al., 2000). Several studies have shown that microplastics in soil can reduce acid phosphatase activity. This reduction has been attributed to the potential of microplastics to alter the enzyme's tertiary structure (Dong et al., 2021; Yi et al., 2021). In contrast, Fei et al., (2020) observed that higher concentrations of PE microplastics (1 % and 5 %) increased acid phosphatase activity in acidic soils. These constrasting findings underscore the difficulty of comparing results across studies, as Yi et al., (2021) examined a different polymer type, while Fei et al., (2020) used the same polymer but with a different microplastic shape, and both studies were conducted using different soil types.

In experiment A, PES (250 µm) and PP microplastics increased the activity of arylsulfatase and PE and PES (3000 µm) decreased arylsulfatase activity in comparison to the control. Arylsulfatases are involved in sulphur cycling in soil; they are responsible for the breakdown of organic sulfate esters into inorganic sulfates and residual compounds (Chen et al., 2016; Germida et al., 1991). These compounds are crucial for certain microbes to biosynthesise proteins, vitamins and coenzymes (Kertesz and Frossard, 2024). Few studies have explored the impacts of microplastic on arylsulfatase in soil (Palucha et al., 2024). Dong et al., (2024) reported that PS microplastics had a reducing effect on the activity of arylsulfatase across different soil types. Overall, this research illustrates that microplastic impacts on soil-plant systems are multifaceted, with both positive and negative effects that depend on polymer type, particle size, concentration and plant species. These mixed results are potentially attributed to the constituents and concentrations of microplastics, plastic additives, and plant-soil properties and mirror findings in broader literature. Thus, underscoring the complexity of microplastic-plant-soil interactions.

5.5.4. Wider implications and prospective future research

In this study, the effects of microplastics on soil and plant performance indicators varied, with some being positive, some negative, and others insignificant. Although some results appear to be nominally positive, they are not necessarily desirable because they still represent deviations from the natural state of soils. Rillig et al, (2021), proposed the theory that microplastics should be seen as a factor of global change based on evidence that they are linked with human activity, they affect biota across different ecosystems and the effects are apparent on a global scale. Identifying microplastics as a global change stressor suggests that microplastics have the potential to act on agricultural soils, like other physical (e.g. temperature warming), chemical (e.g. chemical fertiliser and pesticides) and biological pressures (e.g. weeds and other invasive plant species). Moreover, microplastics may be causing shifts in the soil carbon pool (Zhang et al., 2020). The impact of microplastics on soil enzymatic activities was highly variable and depended on the type of polymer, plant, enzyme and concentration of microplastics. For example, PE microplastics decreased the activity of β-glucosidase, while PP microplastics increased it. This particular finding highlights the complexity of interactions between microplastics and soil properties, including potential changes in microbial community structures, which can have positive and negative indirect effects on plant growth. Further studies on how microplastics influence soil microbial communities are required to understand the underlying mechanisms.

Microplastic leachates may contribute to soil carbon emissions, as they represent a more active source of carbon than plastic solids and certain organic matter (Chen et al., 2024). The leachates contain a broad range of additives such as stabilisers, antioxidants, flame-retardants and pigments that are added to improve the quality of plastic (Sendra et al., 2021). The most frequently detected additive in the environment associated with plastic production include phthalates, brominated flame-retardants and bisphenol-A, all of which are classified as both endocrine-disrupting chemicals and carcinogens (Hermabessiere et al., 2017). The combined effects of micro- and nanoplastics, and their associated chemical additives on soil, plants and biota are not well understood. Microplastics also readily combine with other pollutants already present in the soil matrix, such as heavy metals and pesticides (Kinigopoulou et al., 2022), potentially affecting the distribution and bioavailability of these contaminants in agroecosystems. A clearer understanding of how plastics and the leachates from plastic additives accumulate in plants and enter the human food chain is required to address food safety concerns. In addition to further research on the role of different polymers in transporting pollutants through soil. This study involved two mesocosm experiments, which took place for 102 days in an outdoor polytunnel setting. This is coupled with benefits, as well as limitations. One of the main challenges is extrapolating the findings from mesocosm experiments to real-world scenarios as controlled conditions often fail to capture complex field interactions, and exclude most environmental variability. Further research examining the long-term effects of microplastics on soil properties, soil types, plant growth, soil biota and microbial communities over multiple growing seasons should be undertaken. In addition to large-scale field trials to determine whether microplastics can alter soils in-situ and the productivity of natural Irish grassland systems.

5.6. Conclusions

The main conclusions from this study are:

1. Microplastics had varied effects on plant growth with some types of microplastics stimulating the growth of *L. perenne*, particularly in higher concentrations, while others, such as PE microplastics negatively affected the growth of *T. repens*. Moreover, microplastics were shown to alter soil pH, particularly PE which increased soil pH in comparison to the control.

2. The effects of microplastics on soil enzymatic activities were variable; for example, PE decreased the activity of B-glucosidase, but PP microplastics increased it. This demonstrates that the effects of microplastics on plant performance and soil health indicators depend on the type of polymer, plant species, enzyme and concentration of microplastics. The specific causes for the observed effects of microplastics on *L. perenne*, *T. repens* and the soil are unknown. Although some results may seem positive, they represent deviations from the natural state of soils. Longitudinal studies could determine if microplastics have lasting negative or positive effects on soil and plant productivity.

3. Further research is also needed to understand the specific changes in microbial community composition and function in response to different types and concentrations of microplastics. Since microplastics are carriers for other chemical contaminants in the environment and contain a wide range chemical toxic additive that have can leach into soil and water, additional studies should be carried out to investigate the relationship between microplastics and their associated contaminants, as well as the uptake by plants and soil biota, in a field study setting.

5.7. References

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Chapter 6: The abundance, characteristics and removal efficiency of microplastics from an Integrated Constructed Wetland (ICW) system in Glaslough, Co. Monaghan, Ireland

6.1. ABSTRACT

Microplastic pollution is a growing environmental concern due to its pervasive presence in soils and waterbodies and its potential impact on ecosystems and human health. This study evaluates the abundance, characteristics, and removal efficiency of MPs in an Integrated Constructed Wetland (ICW) system located in Glaslough, Co. Monaghan, Ireland. The ICW, which serves a rural community, treats raw domestic wastewater. Raw wastewater influent, sludge pond effluent and treated water were sampled under different environmental conditions, including periods of lower inflow (dry days) and higher inflow (wet days). Additionally, pond sediments were collected and analysed for the presence of microplastics. In water samples, microplastic concentrations ranged from 29 – 83 MP items L^{-1} in raw wastewater influent, 34 – 71 MP items L^{-1} in sludge effluent, and 0.9 – 1.9 MP items L^{-1} in treated effluent discharged into the adjacent Mountain Water River. The ICW achieved an overall removal efficiency of 96.6%, effectively reducing microplastic concentrations regardless of inflow pressure. Microplastic retention in sediments was notably higher than in water samples, with concentrations decreasing across the sequential ponds. The first vegetative pond exhibited the highest microplastic contamination (2232 ± 193 MP items kg^{-1} dry sediment), while the fifth pond had the lowest (683 ± 229 MP items kg^{-1}). The decreasing trend reflects the ICW's capacity to capture and immobilise microplastics through sedimentation, plant and substrate interactions. The design of the ICW and operational characteristics including the long hydraulic retention time, shallow flow paths and vegetative coverage may have all contributed to high microplastic removal efficiency. Sediment contamination with microplastics poses environmental and agricultural risks, as ICW sludge and similar are often applied back on to agricultural land, potentially reintroducing microplastics into terrestrial ecosystems. Moreover, further research efforts should focus on the long-term effects sediment contamination, seasonal variations and potential bioaccumulation of microplastics in ICW biota.

6.2. Aims and Objectives

The main aims and objectives of this study were to:

1. Quantify the abundance and characteristics of microplastics in an ICW system that receives untreated domestic wastewater from a rural community in Ireland, focusing on microplastics in raw wastewater, treated water and sediments.

2. Determine the overall removal efficiency of microplastics from an ICW system that receives untreated domestic wastewater from a rural community in Ireland.

6.3. Methodology

6.3.1. Study site description

The ICW treatment system at the centre of this study is a horizontal flow free-surface wetland system that is located on the grounds of Castle Leslie Estate. The site is on the banks of the Mountain Water River at Glaslough in County Monaghan, Ireland (6°53'37.94" W,54°19'6.01" N). The site comprises of 5.76 ha land-cover and 3.3 ha in functional water area (Figure 6.1).

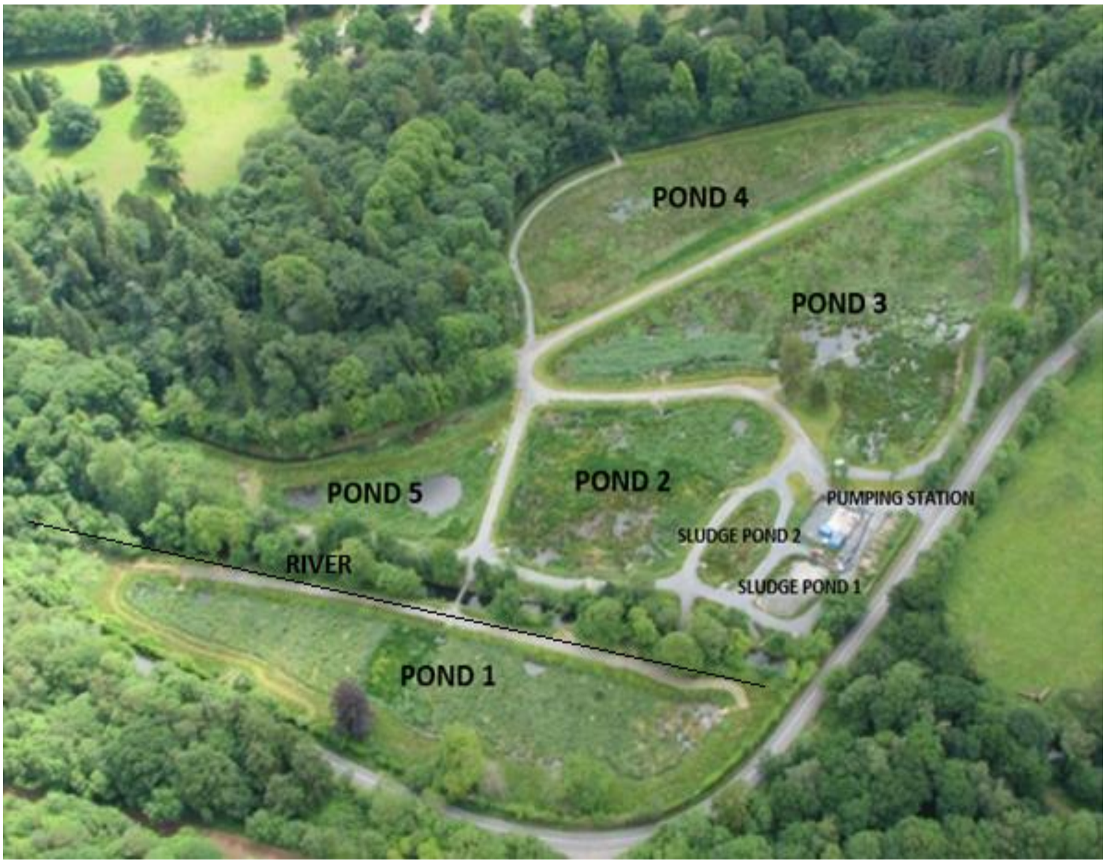

Figure 6.1: Aerial photograph of the ICW in Glaslough, Co. Monaghan, Ireland. Photograph was taken by Dr. Rory Harrington, Waterford County Council, Ireland.

The ICW was established in 2008 and it is based on a universal design, using operational and maintenance guidelines for farm-constructed wetlands in temperate climates and was previously studied as part of a Ph.D. study that focused on nutrient retention (Dzakpasu, 2014). The system is designed to treat domestic wastewater from the inhabitants of the village, as well as managing stormwater surges to help control flows, and prevent flooding by slowly intercepting stormwater back into surface water systems. The ICW uses a maximum influent BOD_5 loading per nominal population equivalent (p.e.) of 60 g d^{-1} (European Union Council Directive 91/271EEC), and the average influent BOD_5

concentration is 875 mg L^{-1}. The design capacity of the CW is 1,750 (p.e.); however, it currently serves approximately 700 (p.e.). The average wastewater inflow volume rate is 120 m^{-3} d^{-1} (Dzakpasu, 2014).

The ICW operates as a continuous flow system; it is a full-scale horizontal flow free-surface water wetland system, with the ICW cells interconnected by PVC piping. Influent primary domestic wastewater pumps directly from the pumping station into one of the two receiving sludge ponds (sedimentation ponds). Thereafter it flows by gravity through five sequential earth-lined vegetative cells (which will be referred to as ponds throughout the document), during an average period of 92 days. The final treated effluent from the fifth pond is discharged directly into the adjacent Mountain Water River (Figure 6.3a). The purpose of the initial sludge pond is to separate and retain solids to prevent sludge accumulation in the wetland vegetative ponds, which could otherwise reduce their capacity. The two sludge ponds are used alternately every five years, allowing one to be de-sludged without disrupting the overall treatment process. The sludge is scraped from the bottom of the cells using a digger and is placed on the banks to dry and decay back into the soil over a period of five years.

Each cell was constructed with no bottom slope or artificial lining. The excavated subsoil was used to construct the base of each cell, and it was compacted to 500 mm to produce a low permeability liner. Soils in the study area comprise of coarse and fine-grained materials. The subsoil in the sludge ponds were classified as sandy gravelly clay, and the subsoil surrounding the vegetated ponds were classified as sandy silt and silty clay (Dzakpasu, 2014). The dimensions of the ICW cells are presented in Table 6.1. Initially when the site was constructed, the ponds were planted in a club pattern using *Carex riparia* Curis, *Phragmites australis* (Cav.) Trin. Ex Steud., *Typha latifolia* L., *Iris pseudacorus* L., and *Glyceria maxima* (Hartm.) Holmb. This included a mixture of *Glyceria fluitans* L. R.Br., *Juncus effuses* L., *Sparganium erectum* L. emend Rchl, *Elisma natans* (L.) Raf., and *Scirpus pendulus* Muhl (Dzakpasu, 2014).

Table 6.1: Dimensions of cells at ICW treatment system, Glaslough, Co. Monaghan, Ireland (Dzakpasu, 2014).

Section	Area (m^2)	Depth (m)	Volume (m^3)
Sludge pond 1	285	0.45	128.3
Sludge pond 2	365	0.45	164.3
Pond 1	4664	0.42	1958.9
Pond 2	4500	0.38	1710.0
Pond 3	12660	0.32	4051.2
Pond 4	9170	0.36	3301.2
Pond 5	1460	0.29	423.4

6.3.2. Experimental Design

Water samples were collected over multiple campaigns to capture environmental variations such as rainfall and different inflow pressures. For water sampling, four campaigns were conducted between March 2022 and March 2023. Sampling points were chosen to represent key stages of the treatment process (i) raw wastewater influent entering the system, (ii) effluent exiting the initial sludge pond (1), and (iii) treated water effluent discharged directly into the Mountain Water River. Samples were collected under both "dry" and "wet" weather conditions to account for the potential impact of different inflow pressures on microplastic abundance (Table 6.2). The cumulative inflow rate was calculated based on the number of litres of wastewater that entered the ICW on an hourly basis on the day prior to sampling, and the day of sampling. On both days that were considered wet, the cumulative flow rates were higher than on the sampling days considered dry, demonstrating a difference in the amount of wastewater entering the ICW under two different inflow pressures which may influence the concentration of microplastics found in the ICW (Figure 6.2).

The total number of samples, and total volume of raw wastewater and treated water analysed are presented in Table 6.3. Sediment sampling occurred on a single occasion in January 2022, targeting the inlet and outlet of the five vegetative ponds, although some access limitations prevented collection from specific points. In total, 16 surface sediment samples were retrieved, with approximately 300 g of wet sediment collected in each sampling location.

Table 6.2: The amount of precipitation on the dates when water sampling took place.

Date	Precipitation (mm) on the day prior to sampling	Precipitation (mm) on the day of sampling	Inflow pressure
11/03/22	9.8	5.6	Higher
20/05/22	0.2	0.9	Lower
20/10/22	0.6	15.6	Higher
30/03/23	1.2	1.6	Lower

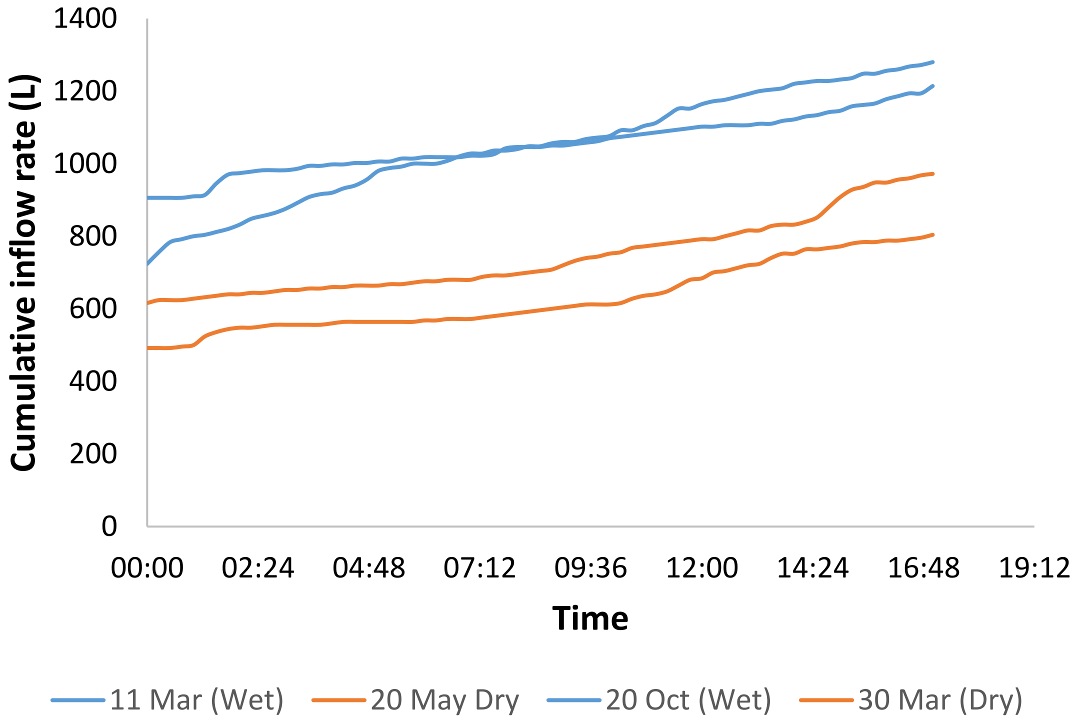

Figure 6.2: The cumulative inflow rate (L) of wastewater entering the ICW calculated from the day before sampling.

Table 6.3: Experimental design: including, sampling locations, types of samples, number of samples, total volume/mass and analysis performed on samples.

Location	Type of sample	Number of samples	Total volume/mass	Analysis carried out
Sludge pond 1 (inlet)	Wastewater	8	40 L	Sequential sieving, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Sludge pond 1 (outlet)	Wastewater	8	40 L	Sequential sieving, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Pond 5 (outlet)	Treated water	8	800 L	Sequential sieving, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Pond 1 (outlet)	Sediment	2	0.6 kg (approx.)	Density separation, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Pond 2 (inlet and outlet)	Sediment	4	1.2 kg (approx.)	Density separation, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Pond 3 (inlet and outlet)	Sediment	4	1.2 kg (approx.)	Density separation, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Pond 4 (inlet)	Sediment	2	0.6 kg (approx.)	Density separation, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy
Pond 5 (inlet and outlet)	Sediment	4	1.2 kg (approx.)	Density separation, Vacuum filtration, Digestion, Microscopy, Raman spectroscopy

6.3.3. Water sampling

A bulk water sampling procedure was performed collecting samples of (i) raw wastewater influent, (ii) sludge pond effluent and (iii) treated water effluent (Figure 6.3). The raw wastewater influent and sludge pond effluent samples were collected using a peristaltic pump with silicone tubing to take duplicate samples of 2.5 L of respective samples. Samples were collected and stored in glass Duran bottles and returned to the laboratory in the Centre for Freshwater and Environmental Studies in DkIT, for processing. Two 100 L samples of the treated water effluent were collected from the discharge pipe of the fifth vegetated pond (pond 5) using a bright pink plastic bucket. The use of buckets for surface water sampling has similarly been used by Miller et al., (2017) and more recently Osorio et al., (2021). The collection of samples using buckets instead of nets ensures that smaller microplastics (generally < 300μm) are retained for analysis.

6.3.4. Sediment sampling

On one occasion in January 2022, surface sediment samples were collected from the five vegetated ponds, which will be referred to as P1 (outlet), P2, P3, P4 (inlet) and P5 in the results section (Figure 6.3). Sediment samples were collected from the inlet and outlet of each pond; however, due to access issues (high vegetation growth and risks posed to personal safety) no samples could be collected from the inlet of P1 or the outlet of P4. Thus, the following sediment sampling locations were P1 (outlet), P2 (inlet and outlet), P3 (inlet and outlet), P4 (inlet) and P5 (inlet and outlet). Sediments were collected (top 5 cm) in duplicate from each sampling point using an Ekman Grab Sampler, with approximately 300 g of wet sediment taken from each point. In total 16 samples were collected and sealed in aluminum foil trays, taken back to the laboratory for further analysis.

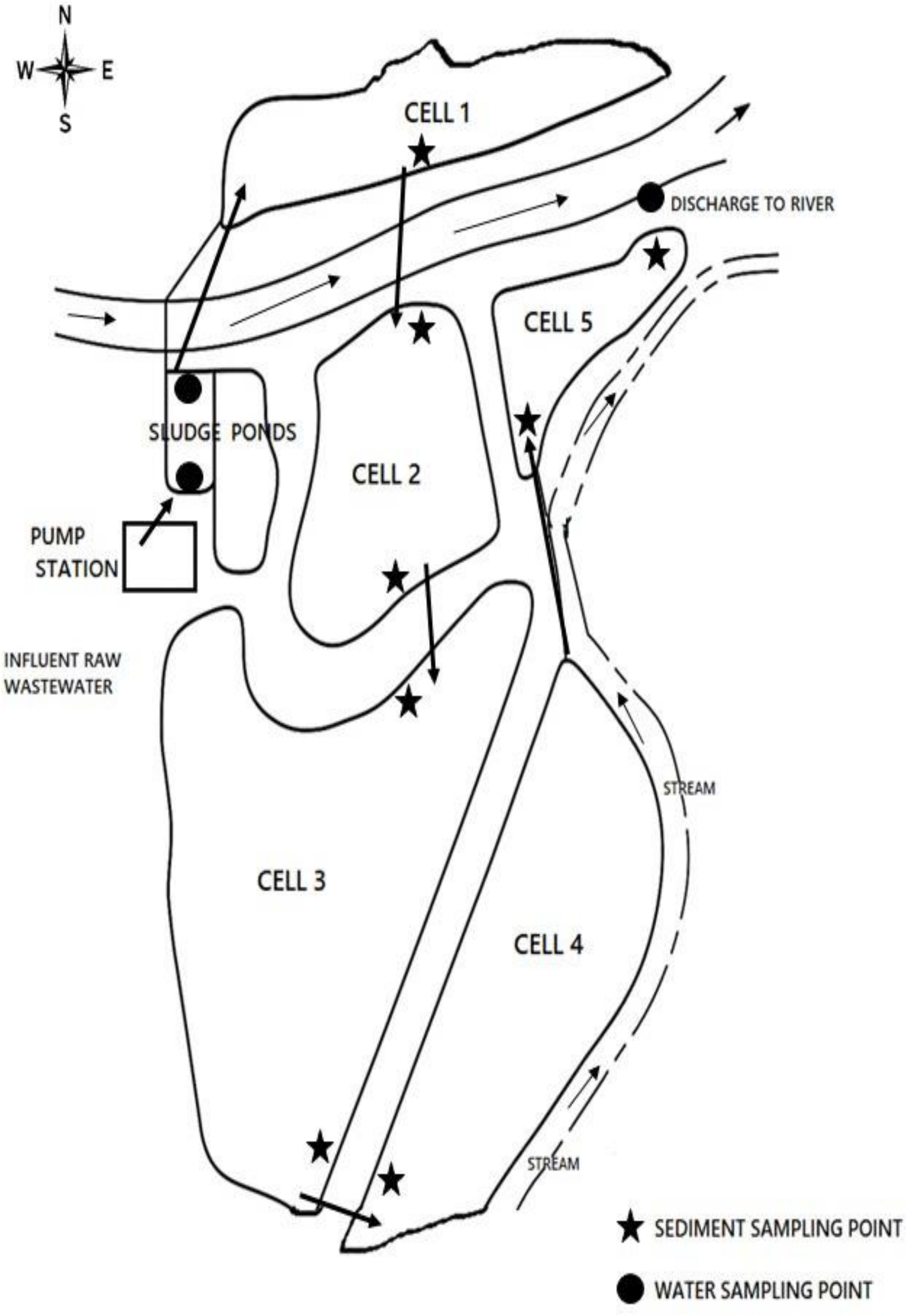

Figure 6.3: Schematic diagram illustrating the locations for water and sediment sampling in the ICW.

Figure 6.4: Two photographs of sample collection points: (a) the raw wastewater influent (b) the treated water effluent.

6.3.5. Water sample processing

The treated water effluent samples were processed on-site on the day of sampling. This involved pouring the total sample volume through a series of stacked stainless-steel sieves (mesh sizes: 50 µm, 100 µm, 500 µm, 1 mm and 5 mm) (Al-Azzawi et al., 2022). On the day of sampling, two sets of sieves were cleaned thoroughly in the laboratory ahead of time to prevent microplastic contamination of samples, and the sieves were all covered using aluminum foil. Sample processing took place as quick as possible to prevent airborne contamination. Once samples were poured through the set of the stacked sieves, all residues that were retained on their meshes were collected using pre-filtered Milli-Q water by rinsing the sieves and collecting all potential microplastic items into sealed glass jars. These were then brought back to the laboratory for the next stage of the analysis. The raw wastewater influent samples and the sludge pond samples were collected and brought back from the site to the laboratory in glass Duran bottles, where they were later poured through a series of cleaned stainless steel sieves of the same mesh size dimensions as previous samples. Once this was completed, using Milli-Q water the glass Duran bottles were rinsed down to release potential microplastics that may have stuck to the inside of the bottles. Each sieve was rinsed down using Milli-Q water to collect all resides and potential microplastic items into sealed glass jars for the next part of the analysis.

6.3.6. Sediment sample processing

Sediment samples were sorted to remove any plant debris or stones that may have been present. No macroinvertebrates were collected from the samples. Sediments were then sieved using a 5 mm and a 2 mm stainless steel sieve, stored in aluminum trays and dried in the oven at 40 °C for 36 hours or until they reached a constant weight.

6.3.7. Microplastic extraction from water

Residues collected in glass jars were further processed using vacuum filtration. All potential microplastics were retained on MN Filter GF-3 Glass Microfibre Circles (pore size: 0.47 µm) filter papers. Post-vacuum filtration, filter papers were left to dry and when dry, organic matter on filter papers were digested using a modified method by Hong et al., (2021): under a fume hood, filter papers were placed on clock-glasses using a tweezers and set on a heating plate at 50 °C. Hydrogen peroxide (H_2O_2) at 30 % was added dropwise (1-1.5 ml) until digestion was completed. This took between 1 to 2 hours to complete per filter paper. All filter papers were left to dry at room temperature in a glass desiccator before being stored in labelled low-density polyethylene (LDPE) clear plastic petri dishes until further analyses. The H_2O_2 (30 %) was pre-filtered to eliminate the risk of sample microplastic contamination.

6.3.8. Microplastic extraction from sediment

Dried sediment samples were analysed for microplastics using a similar method to the one used on soil samples from chapter 4. The modified density separation wet extraction technique was implemented (Corradini et al., 2019, 2020). For each sediment sample taken, subsamples were analysed for microplastics in triplicate by transferring 3 g of dried sediment to a 50 ml glass centrifuge tube. After this, 20 ml of Milli-Q water (p = 1.00 g cm^{-3}) was added to the sediment, and then centrifuged at 5000 rpm for 15 min. Post-centrifugation, samples were allowed settle for a further 15 min. Supernatants were slowly decanted into glass beakers, carefully avoiding disruption of sediment precipitate on the bottom of the glass centrifuge tube. Twenty ml of saturated 5 M NaCl solution (p = 1.20 g cm^{-3}) was added to the tube, centrifuged at 5000 rpm for 15 min and left settle for 15 min, followed by the collection of supernatants. Twenty ml of 5 M Zn_2Cl_2 (p = 1.55 g cm^{-3}) was added to the precipitate in each tube, and the same steps were repeated, however, density separation of microplastics was performed twice with Zn_2Cl_2 for each subsample. All supernatants were collected and vacuum filtered using MN Filter GF-3 Glass Microfibre Circles (pore size: 0.47 µm) and left to dry at room temperature in a glass desiccator before being stored in labelled LDPE clear plastic petri dishes until further analyses. Filter papers with a heavy presence of organic matter were digested using the method by Hong et al., (2021). Under a fume hood, filter papers were placed on clock-glasses using a tweezers and set on a heating plate at 50 °C. Hydrogen peroxide (H_2O_2) at 30 % was added dropwise (1-1.5 ml) until digestion was completed which took approximately 1-2 hours per filter paper.

6.3.9. Microplastic visual identification and characterisation

Microplastics were examined under a stereomicroscope (Olympus SZXY), counted and characterised using microplastic identification protocols reported by (Hidalgo-Ruz et al., 2012). Microplastics were identified and grouped based on their shape (fibre, film bead, or fragment), size and colour. Several measures were adopted to test if suspected microplastics were made from plastic. First, the hardness test was performed to assess the texture and stiffness of suspected particles using a tweezers and a needle (Lusher et al., 2020). In theory, applying pressure to algae, dead animal/plant biomass and/or the mineral fractions of soils will tend to break or crumble them under pressure; however, microplastics will resist the pressure and should not physically change. In addition to this, the hot needle test was performed if uncertainty remained or if microplasitcs were highly weathered. A steel thin-needle was heated to approximately 200 - 300 °C using a soldering iron and put close to a suspected microplastic particle or fibre. If the particle or fibre melted or curved, it would indicate it might likely be composed of plastic (Battaglia et al., 2020; De Witte et al., 2014).

6.3.10. Raman spectroscopy

For the water samples, representative random samples of suspected microplastic particles (n = 240) were picked with the aid of a thin-tipped tweezers from the original counts (n = 1909), mounted to a glass slide using double sticky tape, and analysed Raman Spectroscopy (Horiba LabRAM II, Horiba Jobin-Yvon, France). This reflected 12.6 % of the total number of suspected microplastics from the water samples. For the sediment samples, a number of suspected microplastics (n = 63) were picked with the aid of a thin-tipped tweezers from the original counts (n = 234), mounted to a glass slide using double sticky tape, and later subject to Raman Spectroscopy (Horiba LabRAM II, Horiba Jobin-Yvon, France). This reflected 27 % of the total number of suspected microplastics from the sediment samples. Other studies have used polymer identification methods on up to 10 % and 20 % (Huang et al., 2020; Horton et al., 2017) of the visually identified microplastics, however another study only used 5 % of the total number of suspected microplastics (Jiang et al., 2020). The percentage examined typically depends on a variety of factors including the number of microplastics found in samples, the number of samples, and can sometimes also depend on time, logistics and money constraints. The Raman Spectrometer had a 600-groove mm^{-1} diffraction grating, a confocal optical system, a Peltier-cooled CCD detector and an Olympus BX41 microscope (O'Briain et al., 2020; Loughlin et al., 2021). Spectra were obtained at a range of 100-3500 cm^{-1} using 532 nm laser. All spectra were compared to a spectral reference library (KnowItAll, Bio-Rad) and an in-house extension library was used which contained known virgin polymer type spectra (Marques Mendes et al., 2021). The websites 'Open Specy' and 'PublicSpectra' were also used to identify polymers. The Raman spectrophotometer was used in the Ryan Institute in the University of Galway under the authorisation of both Dr. Liam Morrison and Dr. Ana Marques Mendes.

6.3.11. Quality assurance and contamination prevention

To ensure reliability of the microplastic extraction method, spiking tests were performed using the water and sediments. Positive controls were adopted to validate the microplastic extraction method. For the water samples (n = 6), 2 L of water was spiked with prepared microplastics (n = 24) including white PP spheres (n = 6) (size: 1.55 ± 0.05 mm) red PE spheres (n = 6) (0.5 - 0.6 mm) purchased from Cospheric LLC ©. Transparent PC fragments were prepared by cutting smaller fragments (n = 6) (2.2 ± 0.4 mm) of plastic from a PC plastic petri dish using a scalpel blade, scissors and tweezers. Pink polyester fibres were removed from a 100 % polyester fleece and cut into smaller fibres (n = 6) (2.7 ± 0.9 mm) using a scissors. The same types and dimensions of microplastics were utilised for the spiking of sediment samples (n = 4), 3 g of sediment was used for each test. Average recovery rates were 100 % for the water spiking tests, and 97. 5 % for the sediment samples (see Appendix)

To monitor airborne contamination, settling plates (filter papers) were left out (n = 40) on days of processing and analyses and examined for suspected microplastic contamination. Only three microplastics were detected on the settling plates, indicating minimial airborne contamination (See Appendix). The average number of microplastics per filter paper was less than one, which was considered negligible in comparison to the quantities recovered from each sample matrix. As a result, no correction was applied.

In order to minimise microplastic contamination, measures were implemented at all stages of sample collection and analyses. All work conducted on samples was carried out in a 'clean room' which was specifically used for microplastic work exclusively by a lone operator. The entrance to the room had a sticky mat in place to catch any dust or potential microplastics trapped on footwear and transferred into the room. Before commencing any work in the room, floors were hoovered, and all workspaces were cleaned. A © Dyson model hoover was used as it contained a High Efficiency Particulate Air (HEPA) Filter specifically designed to remove airborne particles such as microplastics. All materials used were made from glass or steel, other than the tubing on the peristaltic pump and the filter paper petri-dishes which were made from PC. The wearing of synthetic clothing during field and lab-work was minimised. Only 100 % cotton lab coats and latex gloves were used during laboratory analyses. Before and between all steps, the equipment was triple rinsed using pre-filtered MilliQ water (0.22 µm). All materials including glassware, samples, tweezers, etc., that were not in use were covered with aluminum foil to avoid airborne contamination in between steps. All solutions used for microplastic extractions (Milli-Q water, saturated NaCl and Zn_2Cl_2 and H_2O_2) were pre-filtered before use with samples (Whatman MN Filter GF-3 Glass Microfibre Circles (pore size: 0.47 µm).

6.3.12. Statistics and data analyses

Microplastic concentrations are reported in MP items L^{-1} for water samples and microplastics in sediments are reported as MP items kg^{-1} of dried sediment. Descriptive statistics including the range, mean, median and standard deviation for all sites were calculated on Minitab® 21.3 (64-bit). A Shapiro-Wilk test and a QQ-plot were used to test the normality of the data. The water and sediment sample data followed a normal distribution ($p > 0.005$). A Two-way ANOVA was carried out to examine the differences between the concentration of microplastics, sample type/location and water pressure/weather.

The removal efficiency of microplastics (Warren et al., 2024) from the ICW system was calculated as follows.

$$\textit{Removal efficiency (\%)} = 1 - \frac{Concentration.outlet}{Concentration.inlet} \text{ x } 100$$

The estimated daily influx of microplastics into the ICW was calculated by multiplying the average microplastic concentration in the raw wastewater influent by the volume of water entering the system on each sampling day. Likewise, to estimate the potential discharge of microplastics from the ICW into the environment, the average microplastic concentration in the treated effluent was multiplied by the volume of water exiting the system on each corresponding day.

For sediment samples, a One-way ANOVA was employed. If significant differences were found, a post-hoc test was performed, which included a Tukey test. All data were recorded using Microsoft Excel. Graphs were produced using Microsoft Excel and Minitab® 21.3 (64-bit) and statistical tests were performed using Minitab® 21.3 (64-bit).

6.4. Results

6.4.1. Microplastic abundance in water samples, and the removal efficiency of the ICW

Microplastics were found in all water samples analysed, ranging from 0.9 – 83 MP items L^{-1}. Microplastic concentrations ranged from 29 – 83 MP items L^{-1} in raw wastewater influent, 34 – 71 MP items L^{-1} in the sludge pond effluent, and 0.9 – 1.9 MP items L^{-1} in treated water. The mean (± SD) number of microplastics in the raw wastewater influent was 43 ± 26 MPs items L^{-1}, 53 ± 8 MPs items L^{-1} in the sludge pond effluent and 1.3 ± 0.4 MPs items L^{-1} in the treated water ($p = 0.004$) (Figure 6.5). On days with reduced and increased inlet flow rates, there were 30 ± 1, and 57 ± 37 MP items L^{-1}, in raw wastewater influent, respectively. Higher concentrations of microplastics were detected in the sludge pond effluent on days with low and high inlet flow rates, with 43 ± 13 and 64 ± 10 MP items L^{-1} found, respectively. Treated water samples contained 1.4 ± 0.7 and 1.2 ± 0.4 MP items L^{-1}, under lower and higher inflow rate, respectively. The mean concentration of microplastics in the sludge effluent was higher than the raw wastewater influent, however; the difference was not significant. The different inlet flow rates (dry/wet weather) did not significantly affect the concentration of microplastics found in samples ($p = 0.123$).

On both low and high inflow pressure days, the ICW demonstrated a high level of efficacy in removing microplastics from the treated water, significantly reducing the number of microplastics entering the adjacent mountain river. Overall, the ICW achieved a 96.6 % removal rate of microplastics from raw wastewater influent to treated effluent. Using recorded flow rates and microplastic concentrations, the potential daily influx and discharge of microplastics were estimated. On low-pressure (dry) days, the estimated number of microplastics entering the ICW reached up to 3.82 $x10^6$ MP items per day, while on high-pressure (wet) days, this increased to 8.51 $x10^6$ MP items per day (Table 6.4). The high removal efficiency of the ICW, meant fewer microplastics exited the system. The potential daily discharges into the mountain river were calculated to be as follows: on dry days up to 2.09 $x10^5$ MP items per day and on wet days, up to 3.75 $x10^5$ MP items per day, highlighting the ICW's strong capacity to retain microplastics under varying inflow conditions (Table 6.5).

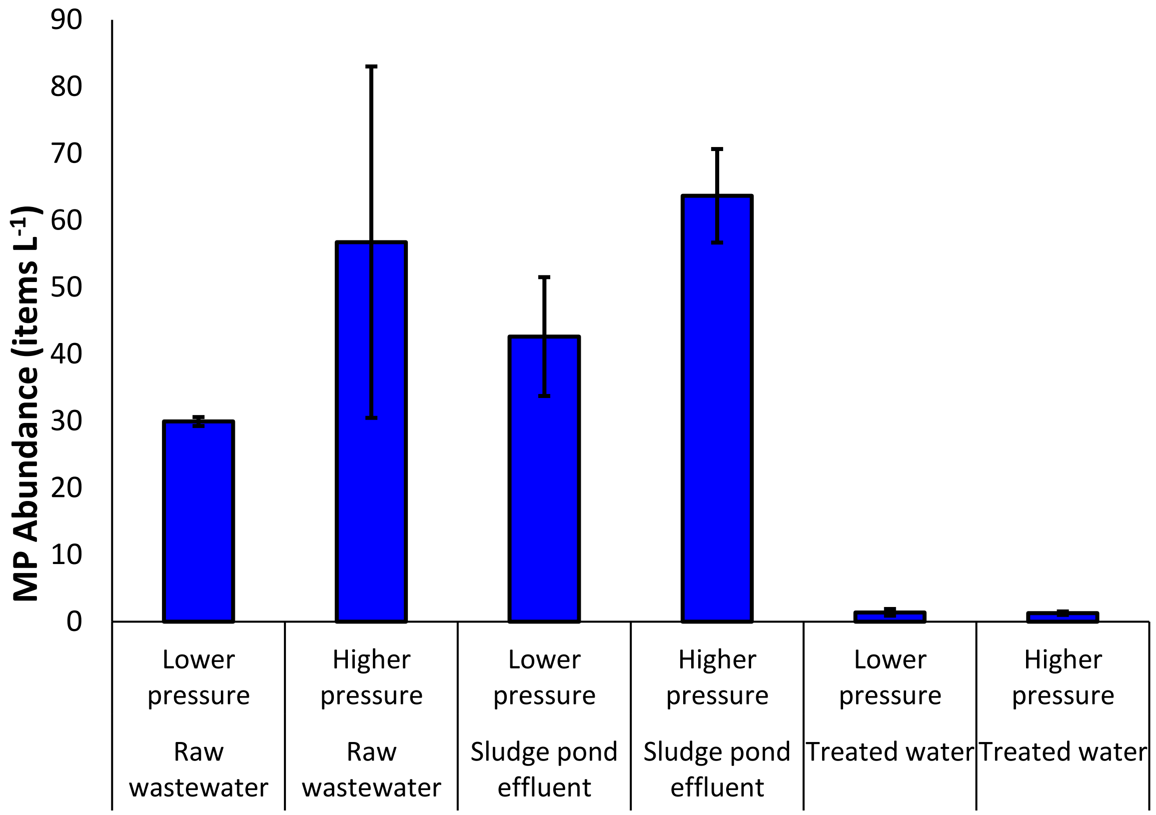

Figure 6.5: The abundance of microplastics (MP items L^{-1}) found in raw wastewater, sludge pond effluent, and treated water under contrasting inflow pressures.

Table 6.4: The calculation results of daily influxes of microplastics into the ICW system, with relevant experimental data.

Sampling day	Mean daily inflow of raw wastewater (L^{-1} per day)	Mean influent (MP items L^{-1})	Mean influx (MP items per day)
Lower inflow pressure	127621	30	3828632
Higher inflow pressure	149443	57	8518268

Table 6.5 The potential discharges of microplastics released into the environment (mountain river water) via treated water, with relevant experimental data.

Sampling day	Mean daily outflow of treated water (L^{-1} per day)	Mean treated water (MP items L^{-1})	Mean discharge (MP items per day)
Lower inflow pressure	149500	1.4	209300
Higher inflow pressure	312503	1.2	375003

6.4.2. Microplastic shapes water samples under contrasting inlet flow rates.

In this study, fibres, fragments and films were detected in the water samples analysed. The number of microplastics from each shape category, with respect to sample type and inflow conditions (weather) are shown as mean (± SD) counts per litre of water in Table 6.6. Most microplastics consisted of fibres, followed by fragments, and films ($p \leq 0.001$). Overall, fibres made up 79.5 % of the microplastics found and fragments and films accounted for 19 % and 1.5 %, respectively. Slightly more fragments were found on days with higher inflow pressure in comparison to lower inflow pressure (Figure 6.6). The number of fragment-shaped microplastics found in raw wastewater, sludge pond effluent and treated water effluent increased by 7 %, 9% and 16 % on days with higher inflow pressure rates in comparison to lower. Films were also found in samples, however, to a much lesser extent than fibres and fragments. There was a slight increase in the number of films identified in samples under a higher inflow pressure.

Table 6.6: The abundance (mean ± SD MP items L^{-1}) of microplastic shapes found in raw wastewater influent, sludge pond effluent and treated water effluent under contrasting inflow pressures.

		Shape (Mean ± SD MP items L^{-1})			
Sample	**Inflow pressure**	**Fibre**	**Fragment**	**Film**	**Bead**
Raw wastewater influent	Lower	24.8 ± 1.0	4.8 ± 0.5	0.3 ± 0.4	0
Raw wastewater influent	Higher	42.6 ± 27.4	12.4 ± 8.2	1.7 ± 1.5	0
Sludge pond effluent	Lower	63.9 ± 32.5	7.4 ± 4.5	0.1 ± 0.1	0

Sludge pond effluent	Higher	47.2 ± 7.8	15.2 ± 0.2	1.2 ± 1.6	0
Treated water	Lower	1.2 ± 0.6	0.1 ± 0.0	0.015 ± 0.0	0
Treated water	Higher	0.9 ± 0.3	0.27 ± 0.0	0.02 ± 0.0	0

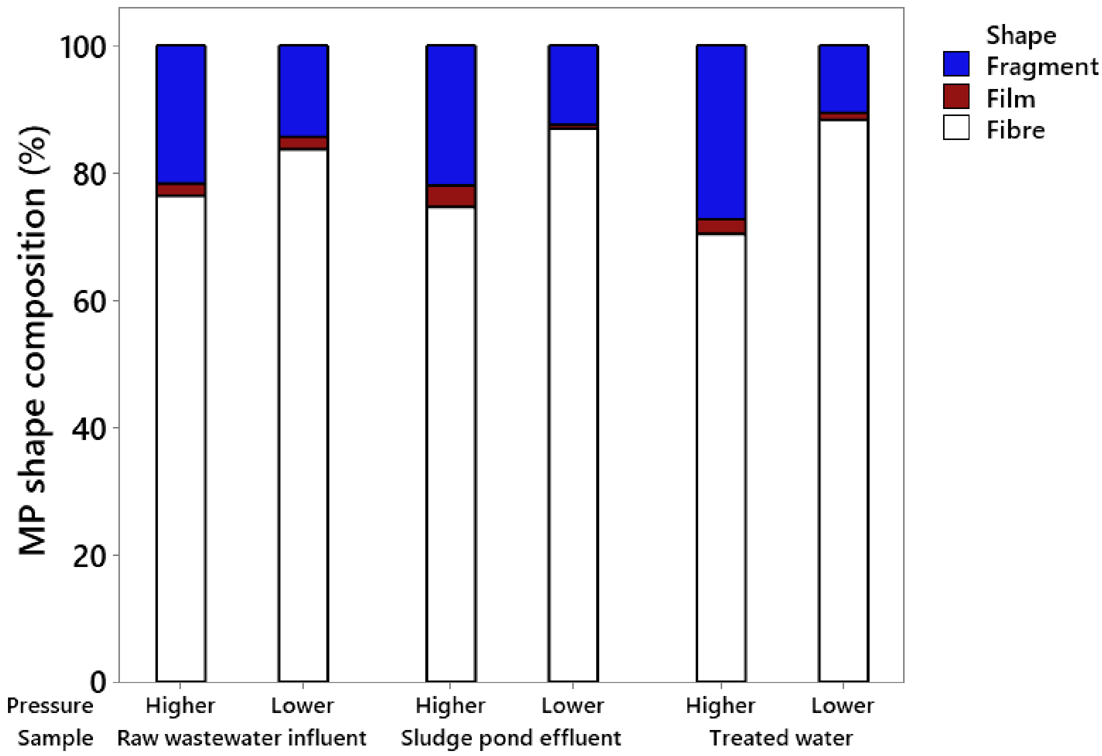

Figure 6.6: The shape composition of microplastics found in raw wastewater, sludge pond effluent, and treated water under contrasting inflow rates.

6.4.3. Microplastic sizes in water samples under contrasting inflow pressures

Microplastics were categorised according to their size distribution in the following classes: 0 – 499 µm, 500 – 999 µm and 1000 – 5000 µm. Overall, most microplastics were between 0 – 499 µm (42 %), which was followed by microplastics sized 1000 – 5000 µm (37 %) and microplastics sized 500 - 999 µm (21 %) (Figure 6.7). A higher number of 'larger' microplastics (1000 – 5000 µm) were found in the raw wastewater influent (44 %) in comparison to the sludge pond effluent (31 %) and the treated water effluent (35 %). Thus, smaller sized microplastics (< 1 mm) were more often found in the latter samples, making up 69 % and 65 % of the total number of microplastics found, in the sludge pond effluent and the treated water effluent, respectively**.**

Table 6.7: The abundance (mean ± SD MP items L^{-1}) of microplastic sizes found in raw wastewater influent, sludge pond effluent and treated water effluent under contrasting inflow pressures.

		Size (Mean ± SD MP items L^{-1})		
Sample	**Inflow pressure**	**0 - 499 µm**	**500 - 999 µm**	**1000 - 5000 µm**
Raw wastewater influent	Lower	9.1 ± 1.8	7.2 ± 0.2	13.5 ± 3.0
Raw wastewater influent	Higher	19.0 ± 9.8	11.5 ± 6.6	26.1 ± 20.7
Sludge pond effluent	Lower	22.3 ± 9.8	8.2 ± 5.6	12.1 ± 16.8
Sludge pond effluent	Higher	23.2 ± 3.2	16.0 ± 1.0	24.4 ± 2.9
Treated water	Lower	0.9 ± 0.7	0.2 ± 0.0	0.2 ± 0.3
Treated water	Higher	0.4 ± 0.1	0.2 ± 0.0	0.5 ± 0.1

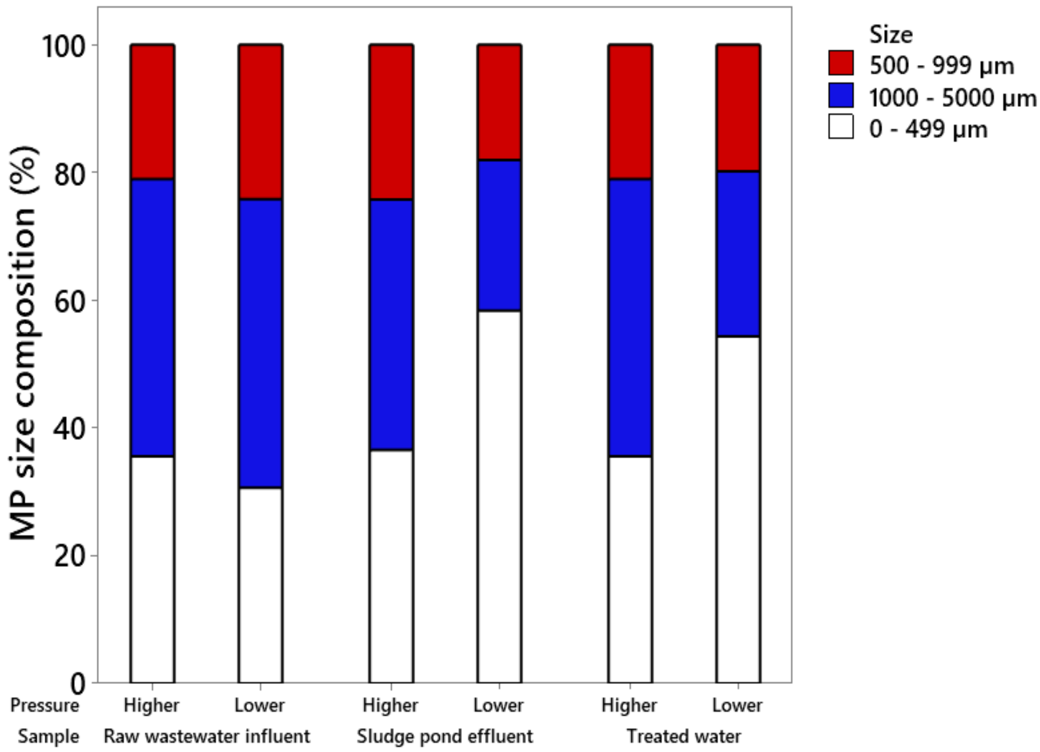

Figure 6.7: The size composition of microplastics found in raw wastewater, sludge pond effluent, and treated water under contrasting inflow pressures.

6.4.4. Microplastic colours in water samples under contrasting inflow pressures

Seven subsets of microplastic colours were identified in water samples, which included the following: transparent, white, black, blue, green, red and yellow. Among all samples, white microplastics were the most abundant colour (7.4 ± 7.4 MP items L^{-1}) followed by black (6.8 ± 6.5 MP items L^{-1}), blue (6.3 ± 1.8 MP items L^{-1}), transparent (4.5 ± 4.5 MP items L^{-1}), green (3.4 ± 4.8 MP items L^{-1}), red (2.2 ± 2.2 MP items L^{-1}), and yellow (1.7 ± 2.7 MP items L^{-1}) (Table 6.8). More transparent and white microplastics were present in raw wastewater influent in comparison to both sludge pond effluent and treated water. Increased numbers of dark microplastics such as black and blue were found. A higher number of darker microplastics made up of black and blue colours were found in the sludge pond effluent samples. In the raw wastewater influent, black and blue microplastics made up 31.95 % and 37.97 % of the colours found on days with higher pressure inflow rates, and this increased to 46.86 % and 41.43 % in the sludge pond effluent (Figure 6.8). Across all samples, green, red and yellow microplastics were found to a lesser extent. The amount of green and red microplastics consistently increased in samples under higher inflow pressures.

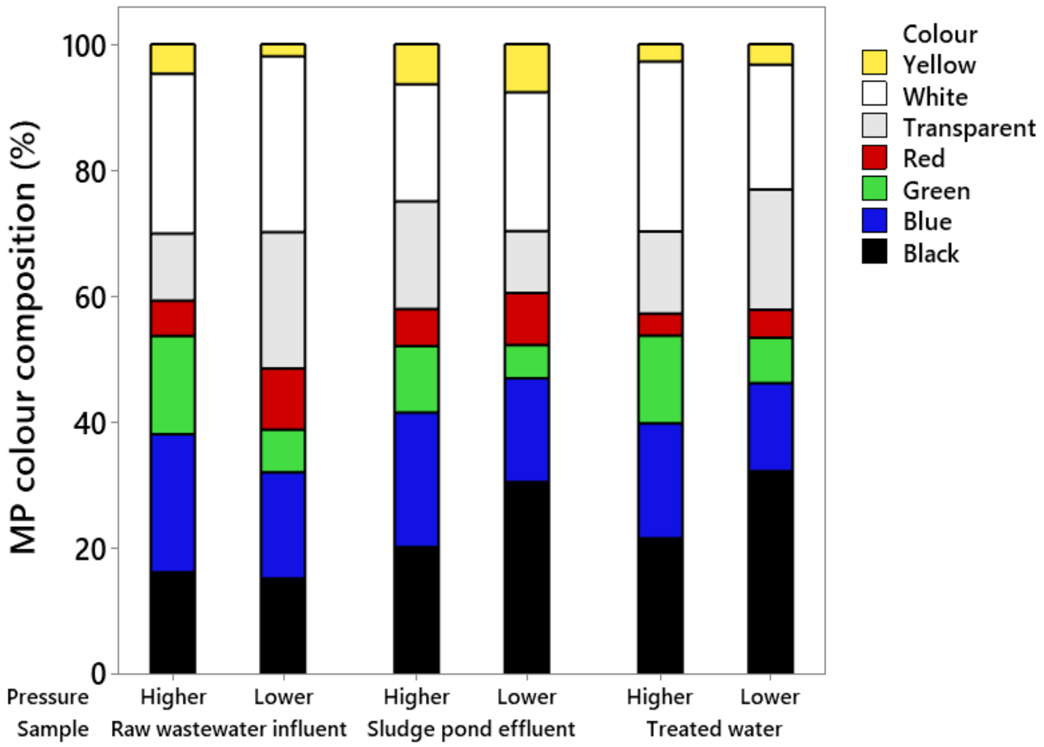

Figure 6.8: The colour composition of microplastics found in raw wastewater, sludge pond effluent, and treated water under contrasting inflow pressures.

Table 6.8: The abundance (mean ± SD MP items L^{-1}) of microplastic colours found in raw wastewater influent, sludge pond effluent and treated water effluent under contrasting inflow pressures.

		Colour (Mean ± SD MP items L^{-1})						
Sample	**Inflow pressure**	**Black**	**Blue**	**Green**	**Red**	**Transparent**	**White**	**Yellow**
Raw wastewater influent	Lower	4.5 ± 0.4	5.1 ± 3.5	2.0 ± 1.4	2.9 ± 0.1	6.41 ± 4.5	8.41 ± 2.8	0.6 ± 0.8
Raw wastewater influent	Higher	9.4 ± 7.0	12.4 ± 7.9	9.9 ± 9.1	3.2 ± 9.1	5.1 ± 1.2	14.0 ± 8.2	2.5 ± 1.2
Sludge pond effluent	Lower	12.8 ± 2.7	7.1 ± 2.6	2.3 ± 0.9	3.9 ± 3.5	4.0 ± 0.0	8.6 ± 2.5	3.8 ± 5.2
Sludge pond effluent	Higher	13.2 ± 8.6	13.0 ± 5.7	6.3 ± 4.4	3.4 ± 2.6	11.1 ± 5.3	12.7 ± 13.0	3.6 ± 4.3
Treated water	Lower	0.4 ± 0.30	0.2 ± 0.10	0.09 ± 0.03	0.04 ± 0.03	0.2 ± 0.06	0.3 ± 0.20	0.03 ± 0.01
Treated water	Higher	0.2 ± 0.06	0.2 ± 0.04	0.1 ± 0.101	0.02 ± 0.03	0.01 ± 0.01	0.06 ± 0.09	0.007 ± 0.01

6.4.5. Microplastic polymers in water samples under contrasting inflow pressures

A subsample of suspected microplastic particles (N = 240) from water samples were analysed using Raman spectroscopy. Of these, 63 % (N= 151) were positively identified as microplastic polymers. The remaining particles were classified as minerals, cellulose, or contained pigments (dyes) that interfered with polymer identification, though they are still considered anthropogenic in origin. Only the particles with confirmed polymer matches were included in the final microplastic number.

In the water samples, nine polymers were detected: nylon, PA, PE, PES, PET, PP, PU, PVA and PVC. The number of polymers detected in order of abundance are as follows; PE (37) > PU (25) > PES (23) > PP > (20) > nylon (16) > PET (12) > PA (10) > PVC (7) > PVA (1) (Table 6.9). Irrespective of weather, most microplastics in the raw wastewater influent were made up of PE (27 %), followed by nylon (18 %), PES (16 %), PP (14 %) and PU (14 %) PET (7 %) and PVC (4%). No PVA or PA polymers were detected in the raw wastewater influent samples. The sludge pond effluent contained the most diverse set of polymers, and again; the most common polymer was PE (29 %), followed by PU (17 %), PES (15 %), PP (12 %), PVC (8 %), PA (7 %), PET (5 %), nylon (5 %) and PVA (2 %). In the treated water samples, PU polymers were slightly more abundant (21 %) than PE polymers (18 %), followed by PES (15 %) and PP (15 %), followed by PA (11 %), nylon (10 %) and PET (10 %). No PVA or PVC polymers were detected in the treated water samples (Figure 6.10).

Overall, PE was the most abundant polymer found across all samples, with slightly more PE found in in raw wastewater samples and the sludge pond effluent in comparison to the treated water. PU microplastics were found in every sample, with slightly more found in the sludge pond effluent and the treated water effluent and less in the raw wastewater influent. PP and PES were found in every type of sample. PET polymers were found in all samples, but mostly on days with higher pressure inflow rates. PA was detected in the sludge pond effluent and the treated water effluent, with slightly more in treated water and on days with higher inflow pressur.. More nylon microplastics were found in the raw wastewater influent samples in comparison to the sludge pond effluent and the treated water effluent. PVC was in the raw wastewater influent and the sludge pond effluent, and none in the treated water effluent. Only one PVA polymer was identified across all samples.

Table 6.9: The number of polymers identified in water samples under contrasting inflow pressures. Total polymers found (N = 151).

		Polymer type								
Sample	**Inflow pressure**	**Nylon**	**PA**	**PE**	**PES**	**PET**	**PP**	**PU**	**PVA**	**PVC**
Raw wastewater influent	Lower	4	0	4	3	3	4	4	0	0
Raw wastewater influent	Higher	4	0	8	4	0	2	2	0	2
Sludge pond effluent	Lower	1	3	8	4	3	2	4	1	2
Sludge pond effluent	Higher	2	1	9	5	0	5	6	0	3
Treated water	Lower	3	5	4	4	6	4	4	0	0
Treated water	Higher	2	1	4	3	0	3	5	0	0
Total		16	10	37	23	12	20	25	1	7

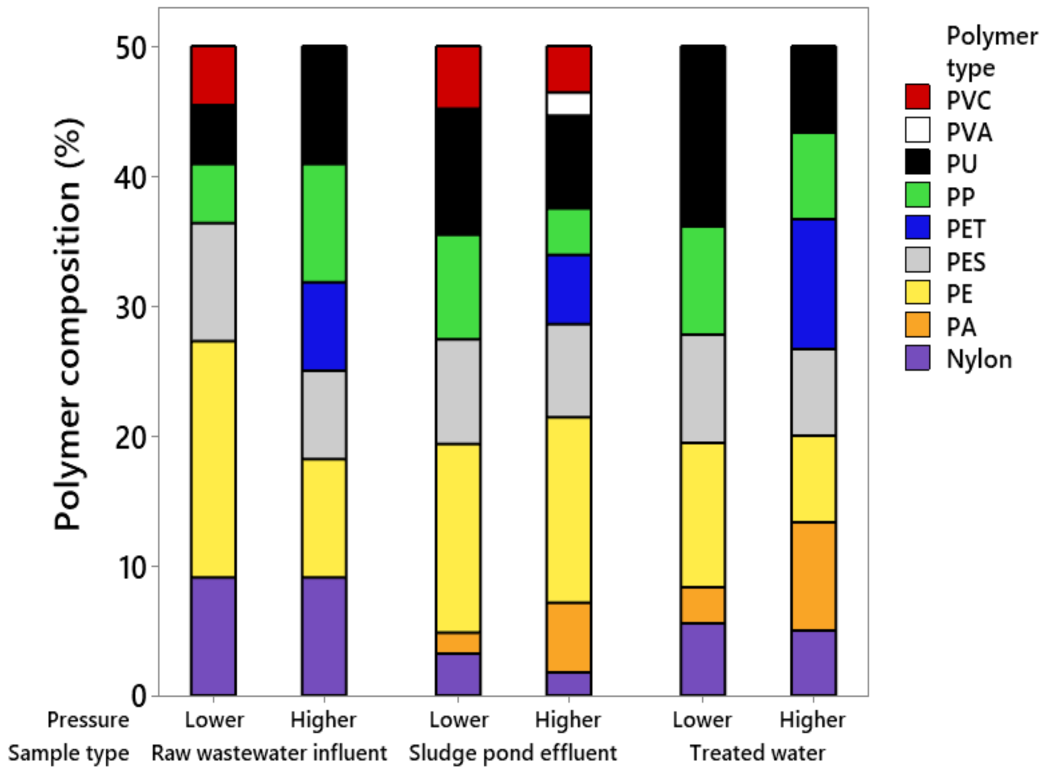

Figure 6.9: The polymer composition of microplastics found in water samples under contrasting inflow pressures (N = 151).

6.4.6. Microplastic abundance in the pond sediments of the ICW

Microplastics were found in all sediment samples collected from the five main sequential vegetated ponds in the ICW, other than in one replicate from the fifth pond, which contained no microplastics. The results indicated that sediments had higher contamination of microplastics, by number of items per volume, than the water samples analysed. Across all five ponds sampled, the microplastic concentrations ranged from 0 – 2096 MP items kg^{-1} dry sediment (Figure 6.11). The microplastic abundance from the first (P1 outlet) to the last pond (P5) showed a decreasing trend. The mean (± SD) abundance of microplastics in sediments in P1 was 2232 ± 193 MP items kg^{-1}, in P2 was 1686 ± 91 MP items kg^{-1}, in P3 was 1298 ± 202 MP items kg^{-1}, and in P4 was 1093 ± 129 MP items kg^{-1}. The lowest concentration of microplastics was found in P5, which was 683 ± 229 MP items kg^{-1}. The number of microplastics in sediments from P1 (outlet) to P5 decreased by 69 %. There was a significant difference in the abundance of microplastics found in sediments across the ponds sampled ($p < 0.001$). P1 (outlet) had significantly higher concentrations of microplastics than all the other sampled pond sediments. The concentrations of microplastics in P2 and P3 were not significantly different, nor were the concentrations between P3 and P4 (inlet), and P4 (inlet) and P5.

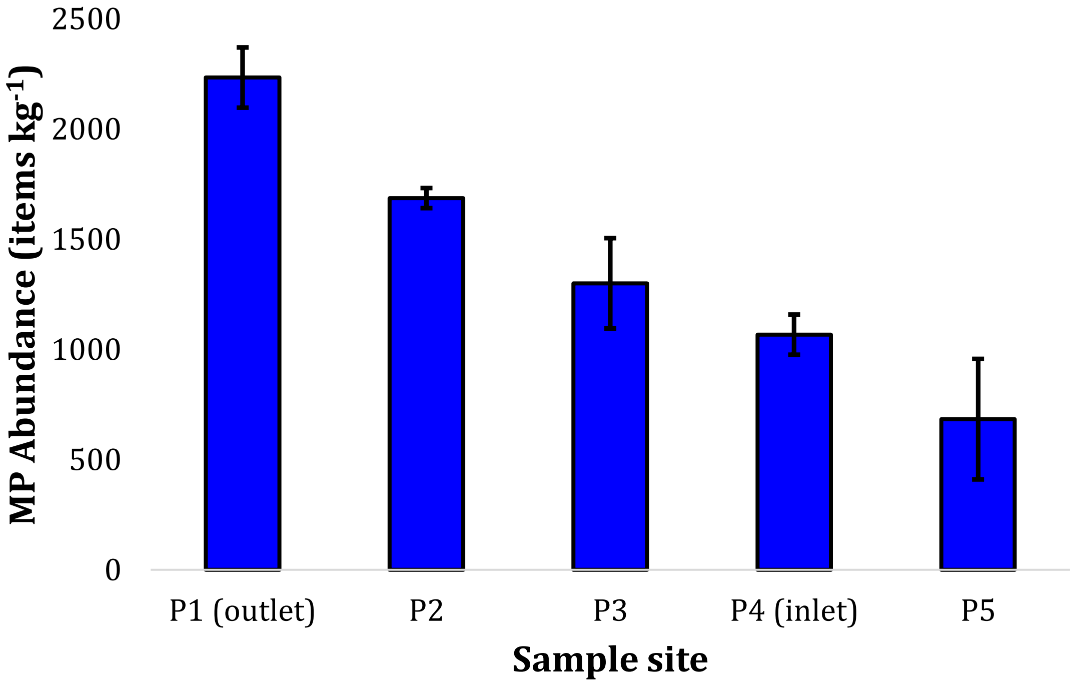

Figure 6:10: The abundance of microplastics (MP items kg^{-1}) found in sediments in the five ponds of the ICW.

6.4.7. Microplastic shapes in pond sediments

Like the water samples analysed, the shapes of microplastics recovered from pond sediments included fibres, fragments, and films. However, bead-shaped microplastics were also detected in pond sediments, but only in the first pond with a concentration of up to 167 ± 79 MP items kg^{-1} (Table 6.10). Moreover, the distribution of microplastic shape categories in sediments differed from the water samples, with sediments containing more fragments and films than were present in the water samples. Overall, fibres were the most abundant shape-type identified in sediment samples, with the highest proportion of fibres found in P4 (78 %) and the lowest found in P1 (51 %) (Figure 6.12), this represented 994 ± 550 MP items kg^{-1} in P4 and 1056 ± 236 MP items kg^{-1} in P1 (outlet), respectively. Fragment-shaped microplastics were the second most commonly identified type of microplastic found in samples. Fragments made up 16 to 34 % of the number of MPs across various ponds. The abundance of microplastic fragments in sediments showed a decreasing trend from P2 (583 ± 319 MP items kg^{-1}) to P5 (83.3 ± 166.7 MP items kg^{-1}). Microfilms were the third dominant category across all ponds. Of these, P5 contained the highest proportion of films in comparison to the other ponds, representing 20 % of the microplastics found in sediments in P5. However, in direct counts, P1 (outlet) contained slightly higher concentrations of film microplastics (277.8 ± 78.6 MP items kg^{-1}) in comparison to P5 which contained (250 ± 319 MP items kg^{-1}).

Table 6.10: The abundance of microplastic shapes (mean ± SD MP items kg^{-1}) found in pond sediments.

	Shape (Mean ± SD MP items kg^{-1} dried sediment)			
Sample site	**Bead**	**Fibre**	**Film**	**Fragment**
P1 (outlet)	166.7 ± 78.6	1056 ± 236.1	277.8 ± 78.6	555 ± 78.6
P2	0	917 ± 419.0	166.7 ± 192.4	583 ± 319.0
P3	0	750 ± 144.3	166.7 ± 166.7	166.7 ± 166.7
P4 (inlet)	0	994 ± 550.7	55.6 ± 78.6	111 ± 157.0
P5	0	333 ± 272.1	250 ± 319.0	83.3 ± 166.7

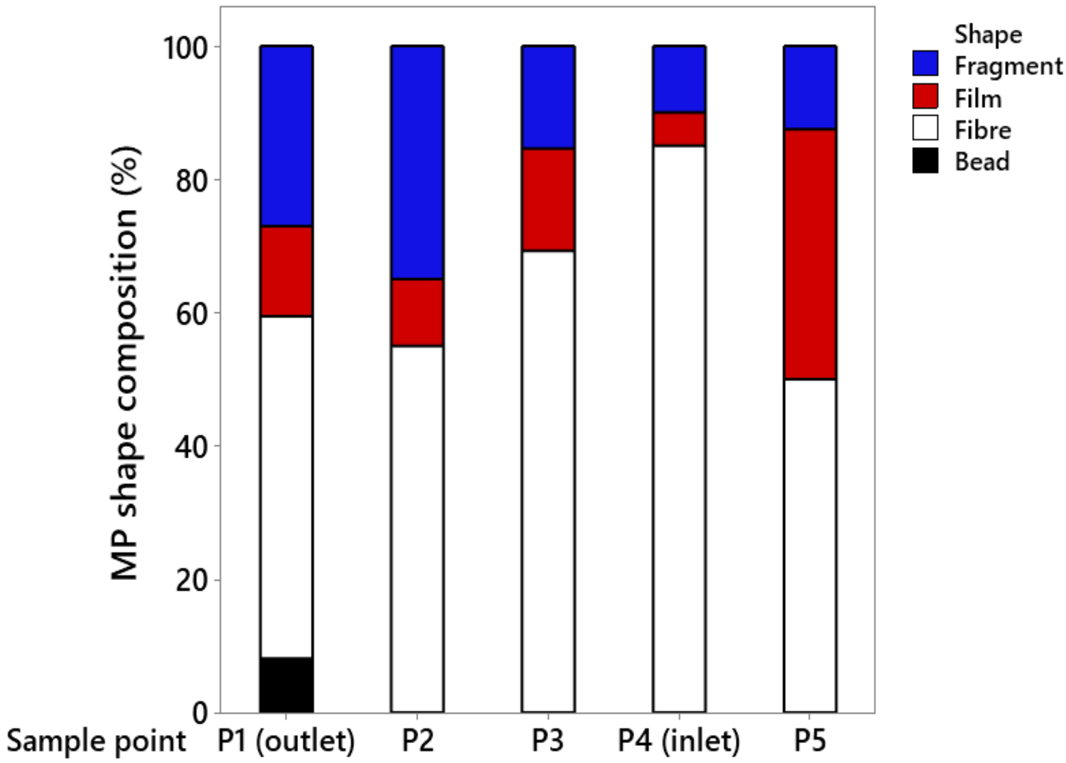

Figure 6.11: The shape composition of microplastics found in sediments in the five ponds of the ICW.

6.4.8. Microplastic sizes in pond sediments

Microplastic sizes were characterised for the microplastics found in all sediment samples, in the following classes: 0 – 499 µm, 500 – 999 µm and 1000 – 5000 µm. In P1 (outlet), an almost equal number of larger and smaller sized microplastics were found in sediments with 51 % of microplastics larger than 1 mm and 49 % microplastics under 1 mm in size (Figure 6.13). This represented up to 333 ± 157 MP items kg^{-1} of microplastics sized 0 - 499 µm, 667 ± 157 MP items kg^{-1} of microplastics sized 500 - 999 µm, and 1056 ± 236 MP items kg^{-1} of microplastics sized 1000 - 5000 µm. The percentage of microplastics found in sediments that were larger than 1 mm showed a decreasing trend from P1 to P5. Therefore, the further microplastics were transported through the ICW; the number of smaller sized microplastics increased by percentage of total.

Table 6.11: The abundance of microplastic sizes (mean ± SD MP items kg^{-1}) found in pond sediments.

Size (Mean ± SD MP items kg^{-1} dried sediment)			
Sample site	**0 - 499 µm**	**500 - 999 µm**	**1000 - 5000 µm**
P1 (outlet)	333 ± 157	667 ± 157	1056 ± 236
P2	305.6 ± 139.8	666.7 ± 90.7	583.3 ± 55.6
P3	667 ± 222	222.2 ± 90.7	388.9 ± 111.1
P4 (inlet)	500 ± 236	222.2 ± 0	222.2 ± 0
P5	305.6 ± 106.4	277.8 ± 143.4	166.7 ± 64.1

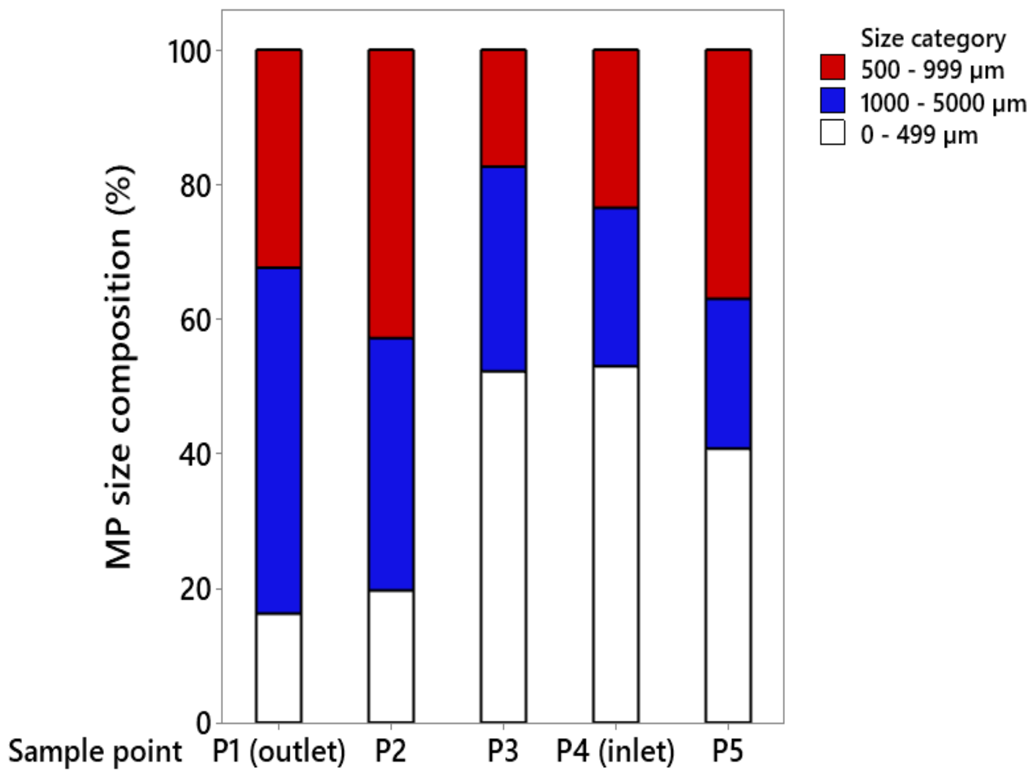

Figure 6.12: The size composition of microplastics found in sediments in the five ponds of the ICW.

6.4.9. Microplastic colours in pond sediments

Five subsets of colours identified within the sediment samples. The main difference in colours found in water samples compared to sediment samples is the absence of both yellow and white microplastics, which were unidentifiable in sediment samples. The two most commonly detected colours were black and transparent microplastics. Every pond contained black microplastics, the distributions ranged from 37 % to 44 %. The highest percentage of black microplastics were found in P1 (outlet) and the lowest in P5. In terms of abundance, this represented 889 ± 157 MP items kg^{-1} in P1 (outlet) and 250 ± 190 MP items kg^{-1} in P5 (Table 6.12). The highest proportion of transparent microplastics relative to the total, were found in P5, representing 59 % of the total number of microplastics found (Figure 6.14). Although significantly higher concentrations of microplastics were found overall in P1 (outlet), almost the same number of transparent microplastics were found in P1 (outlet) and P5, representing 444 ± 157 MP items kg^{-1} and 444.4 ± 181 MP items kg^{-1}, respectively. Overall, blue microplastics represented the third most commonly identified colour category. The first two ponds (P1 (outlet), P2) contained slightly more blue microplastics in comparison to the last two ponds (P4 (inlet), P5). After P2, the abundance of blue coloured microplastics showed a decreasing trend in the following ponds. Green and red coloured microplastics were found in the sediments of P1 (outlet), P2, P3 and P4 (inlet). However, no green or red coloured microplastics were identified in the sediments of P5. Green and red coloured microplastics comprised of 22 %, 19 %, 18 % and 12 % of the total microplastics in P1 (outlet), P3, P2, and P4 (inlet), respectively.

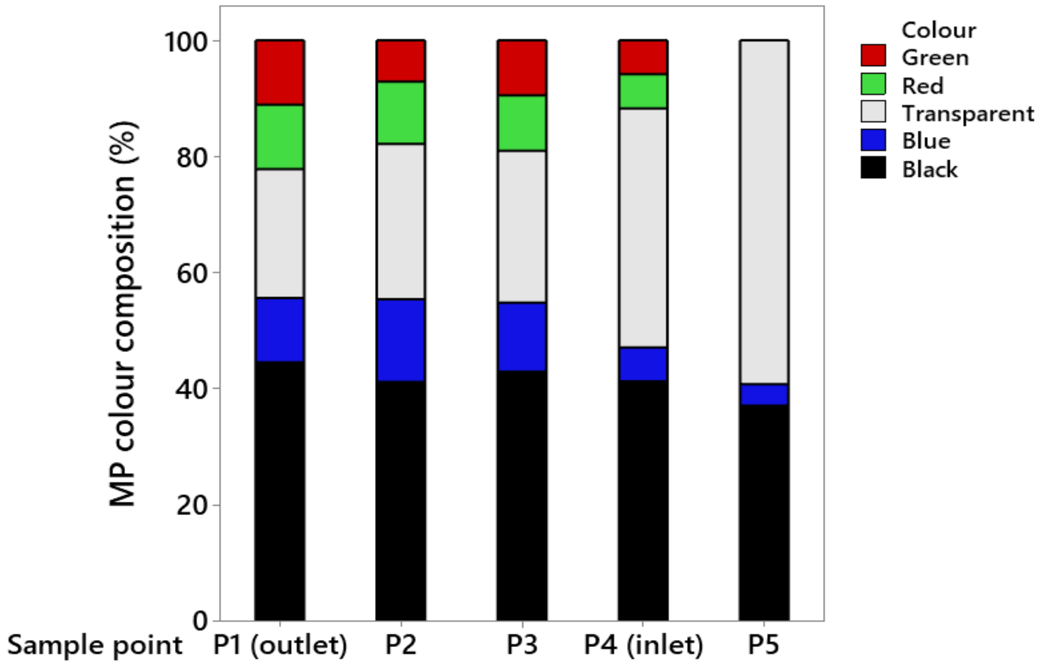

Figure 6.13: The colour composition of microplastics found in sediments in the five ponds of the ICW.

Table 6.12: The abundance of microplastic colours (mean ± SD MP items kg^{-1}) found in pond sediments.

	Colour (Mean ± SD MP items kg^{-1} dried sediment)				
Sample	**Black**	**Blue**	**Green**	**Red**	**Transparent**
P1 (outlet)	889 ± 157	222 ± 157	222 ± 0	222 ± 157	444 ± 157
P2	500 ± 280	222.2 ± 90	83.3 ± 55.6	138.9 ± 106.4	555.5 ± 157.1
P3	611.1 ± 192.4	83.3 ± 55.6	111.1 ± 128.3	83.3 ± 55.6	278 ± 231
P4 (inlet)	389 ± 393	55.6 ± 78.6	55.6 ± 78.6	55.6 ± 78.6	388.9 ± 78.6
P5	250 ± 189.8	27.8 ± 55.6	0	0	444.4 ± 181.4

6.4.10. Microplastic polymers in pond sediments

A subsample (N = 63) of suspected microplastic particles were analysed using Raman spectroscopy from water samples. Of these 82 % resulted in positive matches for microplastic polymers (N = 52). The remaining particles were identified as minerals fragments and various pigments (dyes) which masked a plastic polymer detection but are still anthropogenic compounds. Only positive polymer matches were included in the final numbers on the abundance of microplastics reported in this chapter.

The main polymers found in sediments were nylon (20 %), PA (19 %) and PU (19 %), followed by PES (12 %) PVC (10 %), PP (8 %), PET (7 %), and PVA (5 %) (Figure 6.14). The distribution of polymers found in each pond varied. In P1 (outlet), PVC was the dominant polymer found (28 %), followed by PVA (14 %) and nylon (14 %). In P2, nylon (33 %), PVC (22 %), and PU (22 %). In P3, most microplastics consisted of PA (33 %) and PU (22 %) and in P4 (inlet), PU (30 %) and PES (30 %) were mostly detected. In the final pond, PU, PP, PES, PA all made up almost 20 % each of the total number of polymers detected in P5. Notably, more PVC and PVA polymers were found in pond sediments in comparison to the water samples, but only in P1 (outlet), P2 and P3.

Table 6.13: The number of polymers identified in sediment samples. Total polymers found (N = 52).

	Polymer type							
Pond	**Nylon**	**PA**	**PES**	**PET**	**PP**	**PU**	**PVA**	**PVC**
P1 (outlet)	2	3	0	1	2	0	2	4
P2	3	1	1	0	0	2	0	2
P3	2	3	0	0	1	2	1	0
P4 (inlet)	2	1	3	1	0	3	0	0
P5	1	2	2	1	2	2	0	0
Total	10	10	6	3	5	9	3	6

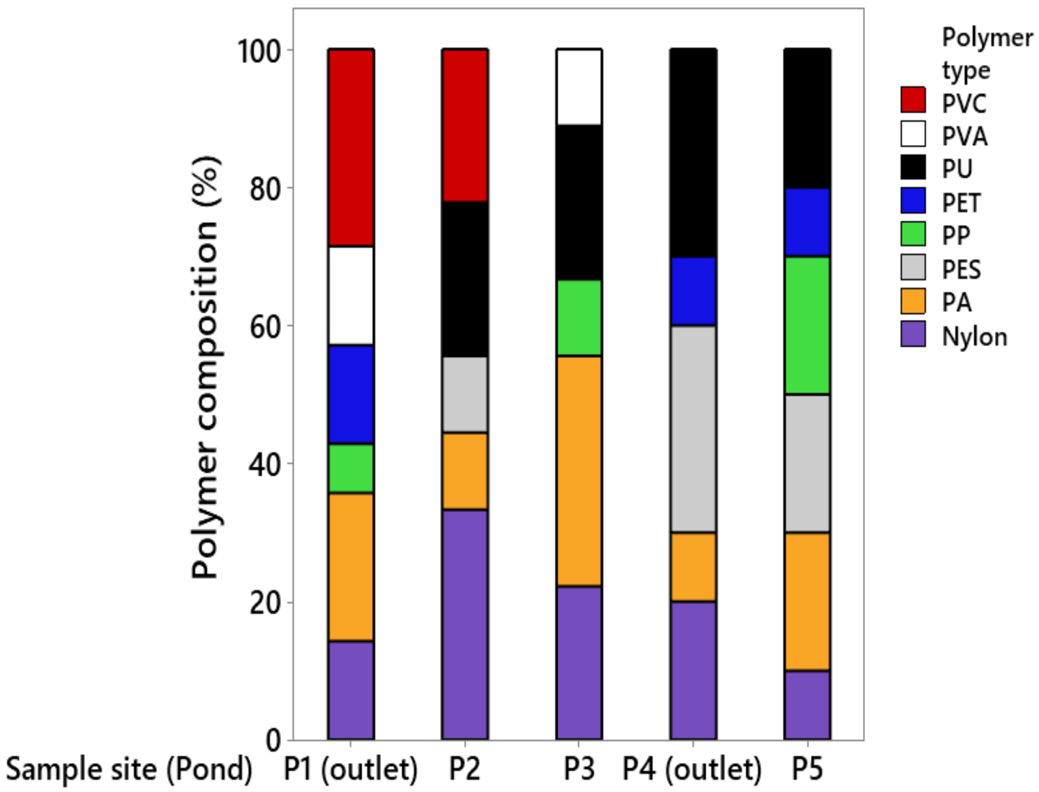

Figure 6.14: The polymer composition of microplastics found sediment samples (N = 52)

6.5. Discussion

6.5.1. Microplastic abundance in water samples

Microplastics were found in all water sample types. Raw wastewater influent contained the highest concentration of microplastics (29 - 83 MP items L^{-1}), and the treated water discharged significantly lower concentrations of microplastics (0.9 – 1.9 MP items L^{-1}) to the mountain river water. Unlike in some other systems (Wang et al., 2020; Zhou et al., 2022), the influent entering the ICW at Castle Leslie was not secondary effluent from a WWTP but instead consisted of untreated domestic wastewater. As a result, high concentrations of microplastics were expected. This is consistent with findings that domestic wastewater is a major pathway through which microplastics enter the environment (Ziajahromi et al., 2017). Domestic wastewater typically carries microplastics from several common household sources such as washed synthetic textiles (Galvão et al., 2020), and personal care and hygiene products (Duis and Coors, 2016). Other sources such as plastic packaging, containers and films used in households can produce secondary microplastics that end up washed down drains (Hamidian et al., 2021). The low concentration of microplastics in the treated water confirms that the ICW system is highly effective in reducing the number (96.6 %) of microplastics from entering the mountain river. These results are similar to other studies on the removal of microplastics from ICWs (Bydalek et al., 2023; Wang et al., 2020; Zhou et al., 2022). However, the abundance of microplastics found in respective influent and effluent waters vary. A recent study on an ICW in Ingoldisthorpe, (UK) assessed sewage derived microplastic abundance and composition from domestic wastewater sources. This ICW had a similar treating capacity of up to 769 individuals, however; the average fibre concentrations in the influent were much lower at 0.01 ± 0.02 fibres L^{-1}. A key distinction between the ICW at Ingoldisthrope and the one in Glaslough is that the influent at Ingoldisthrope consists of effluent from a WWTP, meaning it has already undergone treatment before entering the ICW. As a result, many of the microplastics originally present in the raw wastewater from domestic sources get trapped the different stages of the WWTP, thus do not make their way to the ICW hence the low concentrations reported (Warren et al., 2024). Another study was carried out in Belgium on two small-scale CWs. Similar to this current study; one of the CWs received both domestic wastewater and stormwater and treats the wastewater of 750 individuals. Microplastics in the raw influent ranged from 9.8 to 27.2 MP items L^{-1}. However, the concentrations of microplastics were significantly reduced after the CW treatment, as over 97 % removal of microplastics was achieved (Wang et al., 2024).

Higher concentrations of microplastics were found in water samples analysed on days with higher inflow pressure in comparison to lower inflow pressure, however; the difference was not significant. Other studies have found that rain-wash was a significant contributor to higher levels of microplastics found in CWs (Zhou et al., 2022; Ziajahromi et al., 2020). The potential differences in microplastic abundance may be down to several factors. The inflow of stormwater can increase due to rainfall events, and while this can have the potential to dilute the concentration of microplastics entering the system per volume of water, the overall increased volume can lead to a higher microplastic abundance as stormwater receives microplastics from urban runoff, road runoff and atmospheric deposition (Müller et al., 2020). These can include particles from roads such as tyre wear, degraded larger plastics in the environment, industrial pollutants, paints, and other potential contaminants that may wash off impermeable surfaces i.e., footpaths (Grbić et al., 2020; Monira et al., 2021; Werbowski et al., 2021). Baylan et al., (2024) investigated the removal efficacy of a CW used to treat domestic wastewater in a rural area of Turkey. Similar to the ICW in this study, it is used to treat the municipal wastewater of a population equivalent of 649 individuals. However, the concentration of microplastics in the influent waters was much higher. Across the summer and winter sampling periods, microplastics ranged from 630 – 17,488 MP items L^{-1} of influent water. In the treated water effluent, the abundance of microplastics reduced to 86 – 461 MP items L^{-1}, representing an overall reduction of 87 % of microplastics during summer and 97 % during winter.

6.5.2. Potential removal mechanisms

The removal efficiency of microplastics from the ICW on days with higher pressure inflow rates very slightly higher than it was on lower pressure days. Zhou et al., (2022) reported a dramatic decline in the removal efficiency of microplastics in CW on rainy days. During rain events, the hydraulic loading rate of an ICW increases, and there can be potential knock-on effects to the removal of microplastics from the ICW. Higher flow rates may potentially reduce the contact time between the microplastics and substrate materials, particularly in subsurface flow CWs, thus allowing more time for microplastics to pass through the CW (Zhou et al., 2022). However, on drier days, the reduced flow rates may promote better removal as there would be more time allowed for processes such as filtration, sedimentation and adsorption to occur (Long et al., 2022). One of the factors that may be involved in the higher removal rate of microplastics in the Glaslough ICW on both wet and dry days is the longer hydraulic retention time. This can vary on specific operational or seasonal conditions, however; it lasts approximately 92 days in total, to optimize pollutant/nutrient removal. The high number of vegetated ponds and the shallow flow paths in the ICW naturally support extended hydraulic retention times, which are beneficial for achieving a high removal of contaminants such as phosphorus and suspended solids, reflected in the high-water quality at the site (Dong et al., 2011). Theoretically, this should also be the case for microplastics, the longer retention time may allow more time for microplastics to settle and interact with substrates and vegetation within each of the ponds, before treated water is discharged.

6.5.2.1. Substrate materials and microplastic removal

Microplastic removal may occur through various processes, including sedimentation and retention and sorption by substrates, facilitated by entrapment of microplastics in biofilms, capture and potential uptake by plants, or removal via microbial degradation (to a lesser extent) (Chen et al., 2021; Liu et al., 2023; Su et al., 2022; Xu et al., 2022). The combined action of these processes may enhance the effectiveness of microplastic removal in an ICW. Generally, the substrates are primarily composed of gravel and/or sediment (Liu et al., 2023). The type and size of substrate material in ICWs can play a role in the removal of microplastics from water (Xu et al., 2022). In this study, the substrate material used in the ICW was composed of excavated subsoil containing a mix of coarse and fine-grained particles. Soil samples taken near the inlet of the ICW were classified as sandy gravelly clay, while soils found further along the pathway were identified as sandy silty and silty clay (Dzakpasu, 2014). The sandy gravelly clayey subsoils contain larger, coarse particles compared to the finer textures of silty and clay soils. While these coarser materials typically offer high permeability, they may be less effective in providing the fine filtration benefits associated with smaller-grained substrates. Finer substrate materials in ICWs can enhance microplastic removal due to their greater surface area for adsorption and potential for chemical precipitation (Wang et al., 2024). Research also indicates that ICWs using soil or sediment as the primary substrate (rather than gravel) tend to achieve higher pollutant removal efficiencies from wastewater (Wang et al., 2020).

6.5.2.2. Wetland vegetation and microplastic removal

Microplastic removal can be enhanced by the vegetation present in ICWs. Emergent vegetation in ICWs can slow down the velocity of wastewater in the system which can encourage sedimentation of microplastics (Warren et al., 2024). In this study, the pond basins were shallow and so, emergent vegetation and their litter can form a filtration layer. This vegetative filter can take up a lot of space in the water basin, where suspended microplastics in the water column are likely to be intercepted. The roots, stems and leaves of wetland plants can act as physical barriers, and promote microplastic separation in the system (Liu et al., 2023). The ICW is habitat to species of pond sedges, reeds, cattails, bulrushes, amongst others. Laboratory studies have demonstrated that some wetland plant species, especially those with denser root systems, are effective at capturing and stabilising microplastics. Yin et al., (2021) reported that reeds can effectively intercept PE; PA and PET microplastics by reducing water velocity, meaning larger microplastics had more time to settle out. Vijuksungsith et al., (2024) found that the Common Reed (*Phragmites australis)* could remove more than 50 % of microplastics. The authors suggest that their root system and substrate used influenced the retention of microplastics through physical filtration and biofilm formation on roots. In this study, duckweed was visually observed in abundance across the surface water of ICW ponds. Previous research has identified two duckweed species *Lemna minor* and *Spirodela polyrhiza* as potential vectors for microplastics in aquatic systems. For example, Mateos-Cárdenas et al., (2019) found that *L. minor* can adsorb small PE microplastics (10 – 45 µm) and its dense surface coverage allows it to function as a carrier of microplastics within the water column. Similarly, Dovidat et al., (2020) demonstrated in laboratory conditions that PS microplastics can adhere to the roots, stems and leaves of *S. polyrhiza*.

Given the analytical challenges associated with detecting nanoplastics in environmental samples, this study focused exclusively on identifying and characterizing microplastics within the ICW. However, it is likely that nanoplastics are present in even greater concentrations than the microplastics reported. Research has shown that nanoplastics can be internalized by wetland plants (Tang et al., 2024), with the ability to penetrate plant cells and move within plant tissues. In contrast, microplastics, depending on the size are generally less capable of passing through the plant epidermis and cell walls (Yu et al., 2024). This suggests that plant uptake may represent a potential pathway for the removal of both microplastics and nanoplastics from the water treated in the ICW.

6.5.2.1. Microbes and microplastic removal

Plants in ICWs absorb nutrients from wastewater through assimilation to purify it, whereas plant roots release oxygen and secrete substances providing attachment sites and nutrients to stimulate the growth of microbes (Stottmeister et al., 2003). Microorganisms play crucial roles in ICWs such as the transformation and cycling of carbon, oxygen, nitrogen and phosphorus (Scholz and Lee, 2005). Certain microorganisms can degrade microplastics, however; due to the high molecular weight of many microplastics, they need to be broken down into monomers for uptake and degradation by microbes. This process is complex and takes a significant time to achieve, under the right conditions (Yuan et al., 2020). Microplastics can also provide suitable habitat for microbes, facilitating the formation of biofilms, which may influence the removal of microplastics in ICWs. Recent publications have demonstrated that microplastics can enhance the secretion of extracellular polymeric substances (EPS) by microbes, which can influence the fate, transport and the retention of microplastics in aquatic ecosystems (Ali et al., 2024; He et al., 2022). Moreover, EPS can potentially accelerate microplastic biodegradation by the synergistic actions of multiple exoenzymes (Ge and Lu, 2023; Li et al., 2021). Therefore, biofilm production may promote microplastic capture and retention in water, thereby transporting microplastics to the sediment phase of ICWs.

6.5.3. Microplastics characteristics in water samples

6.5.3.1. Microplastic shapes in water

Fibres accounted for nearly 80 % of the total microplastics identified across all water samples in this study. This finding aligns with expectations, as fibres are commonly reported as the dominant microplastic type in domestic wastewater (Ben-David et al., 2021; EPA, 2023; Mahon et al., 2017). Recent publications report fibres as the most dominanting type of microplastics found in CWs (Long et al., 2022; Baylan et al., 2024; Wang et al., 2024).

Fibre-shaped microplastics in domestic wastewater primarily originate from textile washing, with washing machine effluents releasing between 23,000 to 3.54×10^5 microfibres per kilogram of clothing (Galvão et al., 2020; Acharya et al., 2021; Wang et al., 2024). Variations in fibre release are influenced by fabric type, garment age, water temperature, washing machine deisgn, and detergent use (Jessieleena et al., 2023; Lant et al., 2020). Other sources include flushed synthetic wet wipes, which degrade and release fibres, hindering wastewater treatment processes (Ó Briain et al., 2020; Pantoja Munoz et al., 2018). During one sampling event in this study where higher flow pressures were evident, synthetic wipes and menstrual products were visibly present in the first sludge cell. Additional contributors include nappies which can also release microfibers into the wastewater stream (Alda-Vidal et al., 2020; Munoz et al., 2022).

Fragments made up 19 % of the fraction of microplastics found across the combined water samples analysed. Irrespective of sample type, more fragments found in samples under higher inflow pressure rates, consisting of almost 30 % of the total microplastics found in the treated water samples. The increased number of fragments may be due to smaller fragmented microplastics that enter the ICW through atmospheric fall-out, as demonstrated in other studies (Zhou et al., 2022). Although fragments are sometimes denser and heavier than other microplastic shapes, the impact of weather may support the mobilisation of these microplastics in the environment (Cheung and Not, 2023). Plastic fragments may also be retained on road surfaces and make their way into the combined sewer system that leads to the ICW in Glaslough. Larger plastic items can degrade into microplastics over-time via mechanical stresses such as traffic-induced friction resulting in wear and tear of vehicle tyres (Arias et al., 2022). Other sources can include road and vehicle paints, and construction materials (Burghardt and Pashkevich, 2023). These released microplastics can potentially transport from road dust into stormwater drainage systems, fragmenting further as they travel, ending up in waterbodies (Shafi et al., 2024). Other sources fragments may include secondary microplastics that form through the breakdown of larger plastics such as food containers and packaging, that are discarded on roadsides and run into drainage pipes (Yuan et al., 2023).

6.5.3.2. Microplastic sizes in water

More than 60 % of the microplastics identified in this study were smaller than 1 mm, with approximately 40 % measuring under 500 µm. Notably, a greater number of larger microplastics (over 1 mm) were detected in the raw wastewater influent compared to the sludge pond effluent and treated water samples. As a result, the latter samples contained a higher proportion of smaller microplastics under 1 mm. This suggests that larger microplastics are more likely to be retained in the initial stages of the ICW, while smaller particles are more capable of progressing further through the treatment system. Wetland plants may act as a barrier to larger microplastics transporting further through the ICW system (Helcoski et al., 2020). Moreover, as microplastics transport through the ICW system they could break down and generate smaller microplastics. In theory, microplastics in ICWs should gradually degrade through various natural and mechanical wetland processes based on physicochemical and biological parameters such as temperature fluctuations and microbial activity (Issac and Kandasubramanian, 2021). Higher percentages of smaller microplastics found in the treated water samples may be due to fragmentation of larger microplastics, or perhaps that larger microplastics become entrapped in the wetland vegetation or sediments of the ponds. Smaller microplastics can pose higher risks as they are more easily ingested by many aquatic and terrestrial taxa, size matters and typically smaller microplastics are more likely to be ingested and cause harm to biota (Egbeocha et al., 2018).

6.5.3.3. Microplastic colour in water

Regardless of the type of sample or the inflow pressure rate, white, black, blue and transparent microplastics were most abundant in water samples. However, lighter microplastics were found in the raw wastewater influent in comparison to the sludge pond effluent and the treated water effluent. This meant that a slightly higher fraction of darker microplastics (black and blue) were present in the sludge effluent samples, about 10 % more than the raw wastewater influent. The colour of microplastics can provide insights into the potential sources of microplastics in the system (Kotar et al., 2022). Most single-use or disposable plastics are transparent in nature, and at times, these types of plastics can make their way into sewer systems, either through households or from roadsides (Hahladakis and Iacovidou, 2018; Ngo et al., 2019). Many sanitary towels and synthetic wet wipes are white, and once these products are flushed down the toilet, they begin to release microplastic fibres into the wastewater streams (Ó Briain et al., 2020). Another source of white or transparent microplastics in domestic wastewater may come from dishwasher effluents, and washing machine effluents (Luo et al., 2022).

Zhou et al., (2022) reported that white and transparent microplastics dominated in the influent samples taken from two CWs in China. Long et al., (2022) reported that black and transparent microplastics were most commonly found in influent and effluent samples, and Baylan et al., (2024) detected a significantly higher number of transparent and white microplastics in comparison to black microplastics in influent and effluent samples from a CW. However, the authors noted that in the winter seasons, when rainfall is typically higher, a slight increase in the amount of darker microplastics was evident but overall, the vast majority were transparent and white. One of the biggest sources of coloured microplastics in domestic wastewater originate from washing synthetic textiles, as many clothes items, sheets and bed covers are dyed different colours and can release an extensive number microplastic fibres into wastewater (Kaur and Dandautiya, 2024; Sol et al., 2023). When the ICW experiences higher pressure inflow rates a higher number of coloured and darker microplastics were found which may derive from road-run off, consisting of tyre particles or road paints that are washed into stormwater drainage pipes and make their way to the ICW system. Moreover, studies have shown that microplastics soaked in WWTPs for a long time oxidised and aged to later become either transparent or black (Tang et al., 2020). These two factors, i.e., potential bleaching and oxidation may influence the colour of microplastics found in the water samples in this study.

6.5.3.4. Microplastic polymers in water

The most common types of polymers found in the water samples were PE and PU. However, other polymers such as PP, PES, nylon, PET, and PA were detected, with PVC and PVA to a much lesser extent. As mentioned previously, PE is the most produced plastic on a global scale (Yao et al., 2022); therefore, it is unsurprising that it was found in abundance in the water samples. Some of the potential sources of PE in domestic wastewater and stormwater include sanitary products, and packaging materials that degrade over-time in the environment and drain into stormwater pipes, which make their way into ICWs (Carr et al., 2016; Ó Briain et al., 2020). Moreover, the presence of PE microplastics in domestic wastewater may result from the physical act of dishwashing. The softer side of a dishwashing sponge is typically made from PU plastic; however, the mesh attached to the pad is mostly made of PE or PET. Often while dishwashing, there is a visible reduction in the size and volume of the sponge, which indicates a loss of plastic material through the generation of secondary microplastics (Jessieleena et al., 2023; Lassen et al., 2015; Luo et al., 2022).

Other potential sources of PE microplastics in the water samples may derive from bathing and hygiene activities that include the usage of personal care products (PCPs) such as shower gels, facial cleansers/exfoliators and toothpastes. In addition to eyeliners, nail polishes, sunscreens and other medicinal products (Kukkola et al., 2024; Nawalage and

Bellanthudawa, 2022). Although these types of microplastics are most commonly termed microbeads, they are not always spherical or uniform in shape and are sometimes manufactured in different sizes, shapes and colours (Kukkola et al., 2024). Polyethylene is by far the most common polymer found in PCPs, and other polymers include PP, PET, PU and nylon, all of which were detected in the water samples in the ICW in this study and may potentially derive from PCPs. The United Nations Environment Programme (UNEP) (2017) report that the majority (93 %) of microbeads globally are made using PE, and a study by Bashir et al., (2021) tested 144 PCPs for microplastics and found 76 % contained PE, with most of them comprising of colourless particles. Polyurethane was present in the water samples of the ICW. Potential sources include dish washing pads or sponges which may contribute to microplastic release into sewage (Jessieleena et al., 2023), in addition to PU particles that release from the abrasion of vehicle wheels into stormwater via road run off (Černý and Jančář, 2017; Prenner et al., 2021).

PET microplastics were found in all samples to some degree. Polyethyleneterephthalate or PET resin is a material found in packaging, containers and plastic bottles (PETRA, 2024). Several studies refer to PET as polyester (PES), however; in this study PET and PES were referred to separately because the term polyester is only used when it applies to fibres, particularly fibres from clothes and other synthetic textiles. The presence of PET in samples may have come from the above-mentioned sources, as it is mostly associated with plastic bottles and packaging, however; two thirds of PET produced globally go on to be used in the manufacturing of PES textiles (Smelik, 2023). Moreover, some studies have shown that PET is the main component of glitter products used in artworks and is present in some cosmetics and textiles (Yurtsever, 2019). Several studies report PET as the most abundant polymer found in samples from ICWs. For example, Lu et al., (2022) detected up to 66 % PET in the water samples tested and Warren et al., (2024) reported over 90 % of the confirmed microplastics as PET. It is important to note that the latter study reports them as PET fibres, thus originating mostly from the shedding of synthetic textiles during washing.

In this study, most fibres consisted of the following three polymers: PES, PA, and nylon, which together make up the highest proportion of polymers detected in this study. Although PA and nylon are often referred to interchangeably, they are not the same thing, and nylons are classified within PAs. The PAs are a group of materials with different properties, thus there is a wider range of uses for PA. In 2021, textile production amounted to 112 million metric tonnes and around 54 % was PES (Smelik, 2023). There has been a continuous growing demand for clothes and global production is predicted to triple by 2050. Currently, there are approximately 90 million metric tonnes of clothes consumed on an annual basis (Huang et al., 2024). The textile and apparel industry are recognised as one of the most polluting industries worldwide and microplastics are one of the main emerging contaminants associated with these industries (Chen et al., 2021; Liu et al., 2021). Polypropylene was the fourth most abundant polymer found in samples, and some level of PP was present in all types of samples, under both lower and higher inflow pressure rates. In recent decades, PP has become one of the most widely used thermoplastic polymers because of its cost-effectiveness and ease of processing. The use of PP has expanded across several industries, with a broad spectrum of applications including water filtration, biomedical products, packaging, clothing, automotive and construction materials (Alsabri et al., 2022; Heidarpour et al., 2011). It is possible that the sources of PP entering the ICW wastewater also derive from synthetic textiles and fabrics. Although PP is used in the production of textiles, it is less durable than PES, and is therefore used on a smaller scale in comparison to PES (Periyasamy and Tehrani-Bagha, 2022). Other sources of PP may include packaging materials and food containers as most lunch boxes are either made from glass, or PP if they are plastic, potentially shedding PP microplastics through handwashing or dishwashing units. Sol et al., (2023) found dishwashers release between 207 and 427 microplastics per load using 3 L of water. However, the authors suggest most microplastics were present in the tap water used, and the dishwashing accessories in the machine. Most microplastics were composed of PP (Sol et al., 2023). A diverse mix of polymers were found in the untreated wastewater, sludge pond effluent and treated water, and while polymers can provide clues to the sources of microplastics in domestic and stormwater effluents, being able to pin-point the direct and indirect sources remain a challenge. It is expected that a significant number of microplastics detected in this study derive from synthetic textiles due to the dominance of fibre-shaped microplastics within samples, the range of colours and sizes found, and high proportions of polymers found that are commonly found used to produce synthetic textiles and fabrics.

6.5.4. Microplastic abundance in sediment samples

In this study, microplastic concentrations in pond sediments were higher than those in water samples, when compared on a per-volume basis. This outcome was expected, as numerous previous publications have demonstrated that microplastics levels in sediments can be several orders of magnitude greater than in various waterbodies (Maes et al., 2017; Scherer et al., 2020). Given that sediments were collected from wastewater treatment ponds, elavated levels were expected due to the ubiquity of microplastics reported in wastewater across existing literature (Iyare et al., 2020; Liu et al., 2021). While most studies on microplastic pollution in CWs and ICWs primarily examine influent and effluent waters (Long et al., 2022; Wang et al., 2020; Zhou et al., 2022), some studies exist reporting microplastic abundance in CW and ICW pond sediments (Baylan et al., 2024; Warren et al., 2024). For example, Warren et al., (2024) report significantly higher sediment concentrations of fibres (8152 ± 7022 per kg^{-1} sediment) and fragments (1938 ± 991) compared to levels observed in this present study. Similarly, Baylan et al., (2024) also found higher number of microplastics in sediment samples, with their lowest concentration (2680 MP items kg^{-1}) exceeding the highest concentration detected in this study (2232 ± 193 MP items kg^{-1}). Lu et al., (2022) also documented 3480 ± 4330 MP items kg^{-1} in sediments collected from the inlet zone of a CW treating wastewater.

The ICW in this study has a higher hydraulic retention time compared to other studies (Baylan et al., 2024), therefore microplastics have more time to settle in the ponds and sink into sediments (Warren et al., 2024). Some microplastic polymers have a greater density than water, which can cause them to sink and accumulate in ICW pond sediments (Kabir et al., 2022). In addition, microplastics may undergo biofouling or biofilms can form on their surfaces, consequently increasing their weight and thereby promoting their ability to sink into and accumulate in pond sediments (Liu et al., 2022). With a higher hydraulic retention time, more time is allowed for these processes to occur. The abundance of microplastics from the first pond (P1 outlet) to the last pond (P5) showed a decreasing trend. The higher concentration of microplastics in the initial ponds of the ICW is consistent with previous findings. Wang et al., (2022) observed that microplastics levels were highest at the inlets of four ICWs along Australia's Gold Coast (ranging from 736 ± 335 to 3480 ± 4330 MP items kg^{-1}) and decreased significantly at the outlets (19 ± 16 and 1060 ± 324 MP items kg^{-1}). This decline is largely attributed to sedimentation, adsorption onto wetland vegetation, and potential aggregation with flocculants, which increase particle weight and promote settling (Leiser et al., 2021; Molazadeh et al., 2023). These processes, along with factors such as polymer type, shape and size, likely contribute to the observed reduction in microplastics from P1 (outlet) to P5 (Razeghi et al., 2021; Xia et al., 2023).

6.5.5. Microplastic characteristics in sediments

6.5.5.1. Microplastic shapes in sediments

Like in water samples, fibres were dominant in pond sediments, followed by fragments, films and beads. Other studies have reported fibres as the most common shapes found in pond sediments (Baylan et al., 2024; Wang et al., 2024), and in other aquatic sediments (Abidli et al., 2017; Alam et al., 2019; Peng et al., 2017; Turner et al., 2019). The greater diversity of microplastic shapes in sediments compared to the water samples, as identified in this study, aligns with findings by Lu et al., (2022). As previously discussed, microplastic fibres in sediments likely originate from various sources, with washing machine effluents being a major contributor. Beads were only detected in the first pond which is an unexpected result suggesting that some bead-type microplastics entering via raw wastewater may be composed of higher-density polymers, causing them to sink and accumulate in sediments over time.

The finding contrasts with literature indicating that most microbeads (which are typically found in PCPs) are PE polymers and should remain buoyant in the pond water (UNEP, 2015; Bashir et al., 2021). Notably, no PE polymers were detected via Raman spectroscopy in this study. However, there is evidence to suggest that not all microbeads are made out of PE, and some are made of PP, PES and PS (Bayo et al., 2017; Gan et al., 2023), polymers which were detected in pond sediments. No microbeads were found in the water samples, which was expected due to Ireland’s Microbeads Prohibition Act (2019). The act sets out legal obligations for anyone who`produces, imports, or sells products containing microbeads in Ireland. Restrictions include placing any cosmetic or cleaning products on the markets that are in excess of the permitted concentration of 0.01 % (w/w). The act also prohibits the disposal of substances containing microbeads into drains that lead to municipal WWTPs, and the direct disposal of microbeads to the aquatic environment (Oirechtas, 2019). Some of the microbeads detected in this study may have entered the pond sediments prior to the law’s enforcement, particularly since the ICW in Glaslough is operating since the early 2000s, and supportin the expectation of higher bean concentrations in sediment. Although microbeads were absent from water samples, their risk of re-entry into the system remains, especially through the use of imported PCPs by local residents.

In this study, a large number of microplastic fibres were detected, many of which are typically lighter than water and thought to remain buoyant (Molazadeh et al., 2023). However, biofilm formation on fibre surfaces can increase their density, causing them to sink and settle in sediments. It remains uncertain whether microplastic shape influences biofilm development or interactions with surrounding contaminants (Rozman et al., 2023). Fragments were also more prevalent in pond sediments than water samples, though still less abundant than fibres overall. Their presence suggests a significant contribution of secondary microplastics resulting from the breakdown of larger plastic items in domestic wastewater or the input of microplastic fragments via stormwater. The decreasing proportion of fragments from P1 (outlet) to P5 indicates that the ICW retains microplastics in the initial treatment stages. Warren et al., (2024) also observed a reduction in fragment abundance with distance from the outlet of the first ICW. An unexpected finding in this study was the increase in both the number and proportion of films in P5 compared to earlier ponds. Bydalek et al., (2023) reported a comparable pattern, where larger microplastic films (>1 mm) were more abundant at the outlet than the inlet. They suggested that the buoyancy of larger films may delay sedimentation, potentially explaining the accumulation in later-stage ponds.

6.5.5.2. Microplastic sizes in sediments

The increasing abundance of smaller particles (0 – 499 µm) in sediments from P1 (outlet) to P5 was a key finding. This indicates that larger microplastics were more prevalent in the early ponds, while smaller particles dominated in the later stages of the ICW. This trend likely reflects the fragmentation of microplastics over time and the greater mobility of smaller particles, allowing them to travel further through the system. A similar pattern was observed in water samples, where treated effluent contained a higher propeortion of smaller microplastics compared to raw influent. These results suggest that smaller microplastics may continue to form and accumulate within the ICW after entry, in addition to potential microplastics from sources such as atmospheric deposition.

Studies have detected microplastic sizes in atmospheric deposition ranging from 0 to 5000 µm, but most commonly, particles < 700 µm are found. The disintegration or fragmentation of microplastics in the ICW may potentially occur via mechanical, chemical, and biological processes, in addition to splitting with exposure to UV radiation (Andrady et al., 2022; Kasmuri et al., 2022). Microplastics in the smaller size ranges have been recorded in sediments across other studies (Baylan et al., 2024; Lu et al., 2022). According to Liu et al., (2023) the roots and main body of wetland plants can act as a filter for microplastics with larger particle sizes, while smaller microplastics are usually captured by the sediments in wastewater ponds. The greatest ecotoxicological risks to biota are associated with smaller sized microplastics, as they are more easily ingested (Wright et al., 2013). Moreover, organic pollutants such as pesticides and pharmaceuticals tend to accumulate on smaller microplastics through adsorption (Kinigopoulou et al., 2022). In addition to size, the age of microplastics can significantly influence their interactions with environmental pollutants. As microplastics age, they can develop oxygen-containing functional groups on their surfaces, which may alter their surface charge, hydrophilicity and polarity. These changes can enhance their ability to attract and bind with other environmental contaminants in sediments (Bhagat et al., 2022; Ren et al., 2021).

6.5.5.3. Microplastic polymers in sediments

A greater diversity of polymers were detected in the water samples compared to sediment samples, which was unexpected. It was surprising that no PE was reported in the sediment samples as PE microplastics are often reported as the most common polymer type found in ICW sediments (Lu et al., 2022), and freshwater sediments (Rodrigues et al., 2018). Typically, the density of microplastics influences their distribution in aquatic environments, with polymer type determining whether particles float or sink. For instance, low-density polymers such as PP (0.87 – 0.93 g cm^{-3}) were recovered from sediment samples in this study. As previously, discussed, buoyant microplastics can sink into sediments through aggregation with flocculants and biofilm attachment (Laursen et al., 2023; Molazadeh et al., 2023). Additionally, the extended water residence time in the treatment facility may enhance microplastic sedimentation.

The sources of polymers found in sediments generally align with those identified in water samples. However, a higher proportion of PVC and PVA microplastics were found in sediment samples. This was anticipated due to their higher densities, 1.38 g cm^{-3} and 1.31 g cm^{-3}, respectively. A potential source of PVC in sediments is the ICW piping network composed of PVC. Other potential sources include packaging films, raincoats and shower curtains (Chappell et al., 2022). Polyvinyl chloride poses particular environmental concern due to tis association with phthalic acid esters (PAEs), a group of chemical compounds that are that are widely employed as plasticisers in PVC plastic products (Panthi et al., 2024). Research shows that exposure of PAEs can have acute and chronic adverse effects on reproduction, and damage to liver, kidney, and other organs of aquatic animals. Meanwhile PAEs can cause death and necrosis in aquatic animal embryos (Zhang et al., 2021). Polyvinyl alcohols are commonly used in water-soluble applications, such as detergent pods for laundry and dishwashers. Although PVA is designed to dissolve in water, it does not fully degrade and may contribute to the formation of PVA microplastics in wastewater (Julinová et al., 2018).

6.5.6. The wider implications of this study

An assessment was conducted to estimate the number of microplastics entering and exiting the ICW on sampling days. The results indicated that on days with lower hydraulic inflow pressure, approximately 3.8 million microplastics entered the ICW daily, while this number rose to 8.5 million on days with higher inflow pressure. In contrast, the estimated number of microplastics discharged into the mountain river was significantly lower ranging from 209, 300 microplastics under low-pressure conditions, to 375, 003 under higher-pressure conditions. These findings highlight the important role of ICWs in reducing microplastic pollution entering natural waterbodies. While the reduced outflow of microplastics benefits the receiving environment, some challenges remain. Treated water still contains microplastics, and over-time, even these smaller quantities can accumulate in the Mountain River Water, potentially posing long=term risks to freshwater organisms.

It is positive that fewer microplastics are entering the river, however; ICWs themselves may act as major reservoirs of microplastics, which could adversely impact the plants and animals inhabiting these systems. Although some smaller microplastics or nanoplastics may be taken up, or assimilated by plants and microbes (Yu et al., 2024) the majority are likely to persist in the pond water and sediments. Wetland animals are at risk of ingesting and accumulating microplastics from both the water column and sediment layers.

Although laboratory controlled studies have examined the effects of microplastics on wetland organisms, there is a need for field-scale research to determine whether these particles are accumulating in ICW biota, and causing ecological harm. Integrated constructed wetlands are often celebrated for their role in enhancing biodiversity, but the retention of pollutants such as microplastics may pose a threat to the very ecosystem they support. Furthermore, the potential uptake and transfer of toxic additives, heavy metals and POPs bound to microplastics within the ICW system warrants further investigation.

As ICWs are increasingly recognised as major sinks of microplastics, it is likely that many of the more than 200 ICW systems operating in Ireland (of which there are over 200 of) (VESI, 2024), have accumulated substantial microplastic loads. However, some ICW systems may be more effective than others at retaining these pollutants. Further research is needed to determine how factors such as wastewater source and system design influence the abundance, characteristics, and removal efficiency of microplastics across different ICW types in Ireland.

Although not directly assessed through this study, the two sludge ponds may contain the highest number of microplastics within the ICW system. These ponds are desludged alternately every five years, with the collected sludge spread along riverbanks to dry and decompose after over time. This practice poses a risk of re-introducing microplastics into both river water and surrounding soils. While the ICW system effectively captures microplastics, desludging may inadvertently return them to the environment. This risk should be considered, and alternative methods developed to reduce the potential release of microplastics.

Currently, there are no large-scale effective remediation strategies for removing microplastics from environmental matrices. Therefore, prevention strategies should be implemented. This could involve introducing new legislation on plastics and microplastics to minimise their release to domestic wastewater. One area of focus could be reducing the widespread use of synthetic materials in clothes and other textiles. However, given that clothes are manufactured on a global scale, and many of the clothes consumed are imported (for example, through online retailers), introducing a ban on synthetic materials would be extremely difficult to implement and could have repercussions across multiple industries. A more practical approach may be to legislate the inclusion of microplastic-retention filters in all washing machines manufactured and sold in Ireland. These filters could prevent microplastics from entering wastewater systems at source. Although some of these products are already on the market, many still lack this feature, continuing to contribute to microplastic pollution in ICWs and the wider environment. Mandating this technology through regulation may be a step to tackle microplastic contamination in the environment.

6.5.7. Limitations of this study

One of the main limitations of the study is that sampling was conducted during four separate campaigns spread across different months, which does not provide a comprehensive picture of seasonal variation or long-term trends in microplastic pollution within the ICW. Microplastic concentrations and removal efficiencies could fluctuate with changes in environmental conditions, local water usage and seasonal weather patterns. Thus, the limited temporal scope jeopardises the study's ability to report on results across different times of the year and various seasonal patterns.

Another key limitation of the study is that, due to inaccessibility constraints, sediment samples could only be collected from either the inlet or outlet of the five sequential vegetated ponds in the ICW. This may have resulted in samples that are not fully representative of the overall concentrations of microplastics within each pond. Specifically, sediments could not be collected from P1 (inlet) or P4 (outlet), due to safety concerns and physical barriers that prevented access. A more robust sampling design would involve collecting samples from multiple locations in each pond, including the middle and combining to form one composite sample to promote higher homogeneity of samples.

The study would also benefit from sediment sampling conducted across temporal or seasonal scales to gain deeper insights into the potential fluctuations in microplastic abundance in pond sediments, as well as the impact of extreme weather events on the resuspension of microplastics from sediments into the water column. Moreover, several pilot sampling campaigns were undertaken before finalizing the study design. During these initial sampling stages, pond water samples were collected, but the high volume and elevated microplastic concentrations made it impossible to accurately count the particles. Further research should include a sampling design for the collection of pond water samples, and an investigation into the potential leakage of microplastics from the ICW into the receiving river water by sampling upstream and downstream of the river would address a key knowledge gap.

6.6. Conclusions

The main conclusions from this study are:

1. The ICW system demonstrated a notably high microplastic removal efficiency, capable of reducing microplastics by up to 96 % from raw domestic wastewater before discharged into the Mountain River. This was facilitated by the design of the ICW, the extended retention time and natural filtration mechanisms by plants and sediments. This suggests that ICWs are a valuable nature-based solution for mitigating the release of microplastics from untreated domestic wastewater into aquatic environments, especially in rural or smaller communities. However, consequently the ICW is considered sink of microplastics, potentially affecting the biota inhabiting the site. The retention of microplastics in the ICW could be causing adverse effects to soils on-site. Further research should be conducted to understand if microplastics are accumulating in soils and biota in the ICW system.

2. The study found that weather conditions made an impact (although not significant) on the concentrations of microplastics in influent waters. On rainy days, the ICW received a larger volume of water, which included urban runoff, and as a result increased the influx of microplastics into the ICW system. Despite this, the ICW maintained its high removal efficiency by preventing significant discharge of microplastics into the mountain river water.

3. The study examined various microplastic characteristics such as shape, size, colour, and polymer type, and found that certain characteristics influence their retention within the ICW system. Fibres were the dominant shape, accouting for almost 80 % of the microplastics detected across water samples. Larger microplastics were more prevalent in influent waters, while smaller particles were more commonly found in downstream ponds; suggesting microplastics play a role in their distribution and retention. These characteristics likely influence the behaviour of microplastics in the ICW, e.g. smaller particles potentially absorbing more easily to substrates or biofilms, and larger fibres potentially more likely retained by vegetation.

6.7. References

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Chapter 7: General Discussion

The overall aim of the research conducted and described in this thesis was to investigate the abundance, sources, and potential impacts of microplastics on selected agricultural production systems and agricultural communities in Ireland. This study has taken a novel holistic approach to addressing microplastic pollution in agricultural soils, by integrating social science, field studies, and controlled experiments - an approach that has never been done before in Ireland. As plastics and microplastics in the environment are a source of anthropogenic pollution, understanding human behaviours and attitudes towards plastic is critical for developing effective strategies to tackle the issue.

Microplastics are prevalent in Irish agricultural soils, as shown by this research, which contributes to an evidence base for microplastics in agricultural soils. Over the last number of decades, the use of agricultural plastics has been steadily increasing due to evolving farming practices and technologies. There has been a growing dependency on agricultural plastics, including plastic mulch films, silage wrap, greenhouse covers, and more. However, there remains a lack of systematic legislation to regulate the production, use, disposal, and recycling of these materials, especially with respect to the contamination of soils.

While most research on microplastic pollution has focused on aquatic ecosystems, this study raises the question of how aquatic ecosystems can be fully protected without addressing the problem on land. Microplastics are removed during wastewater treatment; however, frequently, this sludge which is laden with microplastics is applied to agricultural soils, creating a feedback loop that reintroduces microplastics into aquatic ecosystems. The volume of wastewater sludge produced in Ireland is expected to increase over the next 25 years. Between now and 2040, the amount of sludge produced is predicted to increase by 80 %, and as it stands, over 98 % of the sludge produced by WWTPs in Ireland is applied to agricultural land. This indicates that the problem will become worse unless sufficient action is taken. In this study, biosolid applied agricultural soils were identified as hotspots of microplastic contamination, emphasising the need to reconsider the practice of using biosolids on land.

Currently, there are no monitoring programmes on microplastics in biosolids in Ireland, nor is there any guidance or regulations addressing the risks of microplastics in biosolids to soil agro-ecosystems and the wider environment. Although significant progress has been made in legislating the restriction of primary microplastic beads in consumer products, these represent only a small fraction of the problem. Microfibres are the most common type of microplastic found in the environment and in this study, were abundant in soils, wastewater, and sediment samples. To date, microfibres have never been included in policy to tackle microplastic pollution in the environment. Restricting the use of microfibres in clothing and textile manufacturing may have knock-on effects to economic sustainable development. However, limiting the volume of synthetic fibres in production is necessary, as they are a main source of microplastic found in wastewater treatment sludge and biosolids applied to land.

In Ireland, the Waste Management (Farm Plastics) Regulations 2001 mandate producer responsibility for the recovery and recycling of agricultural plastics. Despite this, some farmers still face challenges in accessing adequate recycling infrastructure, coupled with increasing recycling costs and inconsistent availability of collection services in different parts of the country. Although there is widespread concern among farmers about the volume of plastics used and their associated costs (both financial and environmental) the necessity of plastics and the lack of alternative materials for farming operations highlight the trade-offs that occur between increasing food production and protecting the environment.

The European Green Deal’s Farm to Fork Strategy provides a framework for promoting sustainable agricultural practices to reduce overall environmental pollution, whilst ensuring fair economic returns for farmers. However, it has failed to address plastics and microplastic contamination on farms and in agricultural soils. Immediate action may be less likely due to the importance of economic factors in farmers’ decision-making. Furthermore, the increased plastic usage among Irish farmers may be a consequence of progressive farming initiatives, such as improved biosecurity and technologies that reduce emissions and improve nutrient efficiency. Due to these factors, microplastic contamination in agricultural soils is unlikely to improve without additional focus and effort.

In 2022, the UN formed an intergovernmental committee and passed a resolution titled “End plastic pollution: Towards an international legally binding instrument.” Across the

67 member countries, the vast majority were in favour of developing an international treaty to address plastic pollution, with over 90 % supporting the development of a full life-cycle assessment (LCA) of plastics. In the context of this study, a full LCA of agricultural plastics should be undertaken. This would include defining system boundaries to cover the production, use, and end-of-life stages of agricultural plastics, collecting data on raw material extraction, manufacturing, transportation, usage, recycling, and disposal. The assessment should evaluate their environmental impacts, including soil and water contamination and effects on ecosystem health. It should also address the social and economic implications, balancing the costs of alternatives to agricultural plastics with their environmental benefits. Collaboration between researchers, innovators, and stakeholders, e.g. farmers, is essential for this process.

Since there is no feasible way to remove plastics from soils, microplastics should be incorporated in monitoring schemes within the scope of the new EU Soil Monitoring Law in order to identify microplastic contamination hotspots, and guide intervention strategies. Based on controlled experiments conducted in this research, current levels of microplastic contamination in Irish agricultural soils may not have major negative impacts on the growth of the most dominant grassland species in Ireland. However, this situation may change over time as microplastic pollution in agricultural soil is predicted to increase.

Outside of farming, other terrestrial sustainable practices, such as the nature-based solutions (NbS) provided by ICWs, have provided positive environmental services in relation to wastewater treatment. However, the ICW studied in this research was identified as a microplastic sink, which poses significant challenges, and there are many unevaluated consequences that stem from this issue. The ICW acting as a sink for microplastics represents potential risks to the biota residing on-site. The ICW has also been identified as a source of microplastics. While it effectively removes microplastics from wastewater, the sludge produced by these and similar systems is applied to land as fertiliser, reintroducing microplastics into agricultural soils and subsequently into aquatic ecosystems. Furthermore, although moreresearch is needed, it is recommended that the practice of desludging sedimentation ponds on-site at the ICW be reconsidered, as may contribute to the release of microplastics back into the environment.

Overall, this research emphasises that microplastic pollution is widely evident in terrestrial ecosystems and a comprehensive approach to address the complexity of plastic and microplastic pollution in agricultural systems is required but will not be possible without supporting farmers, and extensive collaboration among experts, industry and other stakeholders. It is important to recognise that agricultural soils are subject to microplastic contamination (and this may only become worse) and acknowledge the limitations of current waste management and legislative frameworks. Action such as assessing consumer behaviours, implementing monitoring schemes, reassessing biosolid application practices, and improving recycling services and infrastructure should be taken. There is currently a major global focus towards sustainable farming and this study proposes integrating soil-focused strategies into broader efforts to combat microplastic pollution.

7.1. Considerations for future studies

Although research on microplastics in soils has advanced in recent years, it remains in its early stages. Therefore, the following areas are proposed for future investigation:

1. The absence of uniform methodologies to detect and quantify microplastics in agricultural soils complicates comparing research studies and implementing policies. The development of standardised protocols for the analysis of microplastics in various soil types is important and must be treated as a priority.

2. Biosolids and PMFs have been identified as sources of microplastics in Irish agricultural soils. However, additional potential agricultural sources of microplastics require investigation. Future studies should assess the presence of microplastics in organic fertilisers such as composts, manures, slurries, as well as inorganic fertilisers such as polymer-coated slow-released fertilisers, to determine the extent to which these inputs contribute to microplastic contamination in Irish soils.

3. The long-term impacts of PMFs bured in soils in Ireland should be addressed, and research and innovation into the economic and practical viability of biobased and biodegradable plastics as alternatives to virgin plastics that are currently used in farming practices. It is important to assess the constituents of biobased plastics, and whether they have harmful effects on the overall health of agricultural soils.

4. Long-term and large-scale field studies are needed to evaluate the effects of microplastics on soil biota, including their influence on soil CEC, pH and microbial activity. Additionally, appropriate ecological risk assessment frameworks should be developed to assess the potential impacts of microplastic contamination in agricultural soils.

5. A food chain analysis of microplastics, from farm to fork, should be undertaken to provide insights into the mechanisms by which microplastics enter the human food chain from agricultural soures. Moreover, the relationship between soil, microplastics, and human health should be investigated, including the role of various polymers in the transport of pollutants, and their potential effects on human health.

7.2. Main conclusions

The main conclusions of this research are as follows:

1. Irish agricultural soils are contaminated with microplastics, with varying concentrations observed across different farming land-use types. Factors contributing to microplastic prevalence include the application of biosolids, PMFs, and potentially through general farming activities. The development of standardised methods for analysing and characterising microplastics in environmental matrices, including agricultural soils is essential. Further studies investigating the lasting impacts of PMFs in soils is warranted, along with reconsideration of biosolids application on agricultural land to prevent microplastic release into the environment.

2. Wastewater and sediment samples from the ICW revelaed high abundance of microplastics, with reduced microplastics exiting the treatment system than entered, indicating that the ICW is a microplastic sink. This presents considerable challenge and there are many unevaluated consequences that stem from this, such as potential risks to biota that inhabit the ICW. Furthermore, the sludge produced by ICWs and similar treatment systems is often applied to agricultural land as fertiliser, thereby reintroducing microplastics into soils and potentially into aquatic ecosystems. This practice should be re-evauated to reduce the transport and accumulation of microplastics in the environment.

3. Microplastics can potentially alter plant growth and soil chemical and biological properties. Through mesocosm experiments, the main effects of microplastics included increased soil pH and varied effects on enzymatic activity. Specific microplastics increased the growth of *L. perenne* and decreased the growth of *T. repens*. Although some of the effects were nominally positive, they still represent deviations from the natural state of agricultural soils. Further research is required to evaluate the overall effects of microplastics in agricultural soils, including risks to soil biota, and microplastic interactions with microbial communities.

4. Addressing microplastic pollution in agricultural soils requires active engagement with stakeholders, particularly those in the agricultural sector. Irish farmers were surveyed regarding their use of agricultural plastics, as well as their awareness and perceptions of plastic and microplastic pollution. While farmers recognised the impotance of plastics for maintaining productivity, many also expressed concern about their environmental

impact. However, challenges such as limited access to recycling facilities and the financial costs associated with recycling agricultural plastics remain major barriers. These issues should be addressed to enable more effective mitigation of plastic pollution in agro-ecosystems.

Appendices

Appendix 1. Copy of Publication: Farmers' attitudes towards agricultural plastics – Management and disposal, awareness and perceptions of the environmental impacts

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Farmers' attitudes towards agricultural plastics – Management and disposal, awareness and perceptions of the environmental impacts

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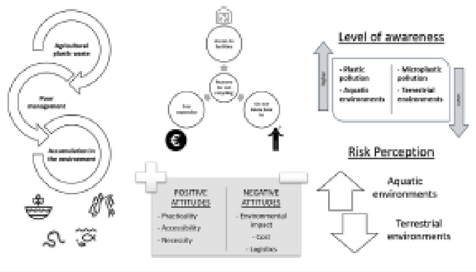
H I G H L I G H T S

- Most farmers are concerned about the amount of plastic waste generated by agriculture.
- Agricultural plastic disposal methods vary by the type of plastic, the cost of recycling and access to facilities.
- Farmers understand that plastics are necessary for tasks on the farm.
- Farmers also associate agricultural plastics with logistical and monetary burdens and their negative impact on the environment.
- Most farmers believe that aquatic environments are under more threat by plastic pollution in comparison to the terrestrial environments.

G R A P H I C A L   A B S T R A C T



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A B S T R A C T

The amount of plastic waste resulting from agricultural practices is increasing and this trend is expected to continue. Although plastics are essential for certain farming tasks, their impact on the environment is becoming a major issue of concern. Mismanaged larger plastics can disintegrate into microplastics and make their way into soils, surface and groundwater sources. Microplastics are extremely persistent and have the potential to facilitate the transfer of contaminants through the environment, potentially affecting terrestrial and aquatic wildlife. A descriptive survey was conducted on a sample of farmers ($n = 430$) in Ireland to assess their attitudes on agricultural plastic waste management and their awareness and perceptions of the impacts of microplastics and plastics on the environment. This study found that most farmers (88.2%) are concerned about the amount of plastic waste generated by farming activities. Agricultural plastic disposal methods vary and recycling rates mostly depend on the type of plastic, the cost of recycling and access to facilities. Most farmers view agricultural plastics negatively due to their impact on the environment but also because of the monetary and logistical burdens associated with them. Farmers were relatively aware of microplastics (57.5%), but overall more farmers felt they knew more about plastic pollution than microplastic pollution and these issues in aquatic systems. This was also evident when it came to their perception of the risks plastics pose on the environment with more farmers believing that aquatic environments are at greater risk than the terrestrial environments. Future research efforts must focus on plastic and microplastic pollutions in soils to inform policy-makers and to create greater public awareness. In addition to this, several developments are needed in a collective effort by governments, policy-makers and other stakeholders to reduce plastic and microplastic problems in agriculture.

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1. Introduction

Over the last five decades, plastics have increasingly become an integral part of our daily lives. Industries such as the automotive, agricultural, electrical and electronics, building and construction, packaging and consumer household appliances all rely on the production of plastics (Plastics Europe, 2021). The practical elements of plastics are well known, as they are clean and convenient packaging materials with high strength and durability, however, the main characteristics that make them desirable are also the main contributing factors to their presence and persistence in the environment (Crawford and Quinn, 2017). In addition to this, over-consumption and inappropriate waste management strategies have led to their ubiquity in both aquatic and terrestrial systems (Yu et al., 2022).

Plastics have created huge opportunities for increased efficiencies in modern agricultural production systems with farmers heavily depending on plastic for mulching crops, wrapping silage, storing feed and fertiliser, greenhouses, polytunnels, and piping (Plastics Europe, 2020) and for many farmers, there are no alternative materials available. Plastics are used to store and transport agricultural products (Razza and Cerutti, 2017), and medicinal and artificial insemination injection products made from plastic are used when treating farm animals (Bas et al., 2011; Rethorst, 2015). As a consequence, globally, the amount of plastic waste coming from agricultural practices is estimated within the range of 2 to 6.5 million tonnes per annum (Brodhagen et al., 2017). Most agricultural plastics are low-value, single use materials that may have encountered a high degree of contamination, therefore they are challenging to collect, recycle and reuse and so, recyclers typically will not accept them in many jurisdictions (Muise et al., 2016). The cost of transportation, recycling and landfilling fees can lead to illegal dumping, burial or burning of wastes on-site, which subsequently has knock-on effects to the environment through the emission of harmful substances into soil, water and air (Muise et al., 2016). The release of pollutants from open emissions can seriously affect local air quality and pose significant risks to human health (Scarascia-Mugnozza et al., 2012). Moreover, agricultural plastic wastes are often dumped along watercourses which have been shown to threaten aquatic life, and wastes may be buried in soils, which may cause significant reductions in soil quality and crop yields (Briassoulis et al., 2013).

Another risk posed by inappropriate agricultural plastic waste management is the generation and accumulation of microplastics in the environment. Microplastics are most typically defined throughout the literature as small plastic particles <5 mm in size (Barnes et al., 2009; Lee et al., 2013). Microplastics persist and accumulate in the environment by direct release or through fragmentation and represent potential threats to terrestrial and aquatic biodiversity and function, and are entering the human food chain (Mercogliano et al., 2020; Cverenkárová et al., 2021; Okeke et al., 2022).

There is currently no effective method to remediate microplastic particles from the natural environment (Deng et al., 2020), and as plastic and microplastic pollution are human-made problems (Kramm and Völker, 2018), understanding the knowledge and perceptions of the public on these topics is vital in order to facilitate public buy-in in terms of tackling the problem first-hand (Henderson and Green, 2020). While many scientific articles have been published regarding the properties of plastic and microplastic particles (Chubarenko et al., 2016; Min et al., 2020), the sources and migration of microplastics in different environments (Auta et al., 2017; Guo et al., 2020; Li et al., 2021), the fate and ecotoxicity of microplastics (Hurley and Nizzetto, 2018; Yu et al., 2020; Du et al., 2021; Rai et al., 2021), and the hazards associated with microplastics and human health (Rahman et al., 2021; Campanale et al., 2020), studies on perceptions and attitudes to plastic and microplastic pollution are largely underexplored and deserve more attention (Deng et al., 2020).

Recently, a report was published on the behaviours and attitudes of the general public towards single-use plastics in Ireland (EPA, 2022). A survey was conducted on a national representation for three consecutive years to track potential evolutions of attitudes and behaviours of plastic consumers over-time. The study found that there is a high concern among the Irish population, and the majority are willing to take steps to reduce plastic usage. Other studies in Portugal, Australia and China have been done to assess societal attitudes towards plastic pollution (Dilkes-Hoffman et al., 2019; Soares et al., 2021; Deng et al., 2020). However, to the best of our knowledge, no studies have addressed the attitudes of farmers towards microplastic and plastic pollution. Farmers and members of the agricultural sector are major users of plastics and in recent times, there is an increased focus on the impacts of farming on the environment. The agricultural sector is recognised as capable of leaking significant amounts of plastics into the environment (Pazienza and De Lucia, 2020). Therefore, the attitudes of farmers towards these issues are important because both plastic and microplastic pollution are evident in the agricultural- and wider-environment and thus, a bottom-up approach to tackling these issues are much needed to develop interventions to reduce plastic release into the environment. Thus, the main objectives of this study were to assess the behaviours and attitudes of Irish farmers towards the usage and disposal of agricultural plastics. The study also aimed to evaluate farmers' awareness of microplastics and their perceptions of the overall impacts of plastics on the environment.

2. Methodology

A descriptive mixed-method survey design was chosen using a self-report structured questionnaire to achieve the main research goals. Descriptive mixed-method designs are widely used in survey research design to integrate quantitative and qualitative findings to strengthen the outcomes of the study. The classic definition of mixed methods research by Greene et al., (1989), is "those that include at least one quantitative method (designed to collect numbers) and one qualitative method (designed to collect words)". Mixed methods involve using quantitative data and qualitative data to gather information that can be used in parallel to complement each other (Zohrabi, 2013; Shorten and Smith, 2017). Descriptive studies using questionnaires can help define the opinions, attitudes and behaviours of data subjects on a given topic (Best, 2003). To achieve this, the questionnaire included 28 questions, using Likert-type, single and multiple choice questions in order to collect quantitative data and open-ended questions for qualitative data. Prior to the administration of the final survey, a pilot-study ($n = 18$) was conducted to increase research quality (Gudmundsdottir and Brock-Utne, 2010) by identifying potential weaknesses in the survey so they can be rectified prior to the implementation of the full study (Malmqvist et al., 2019).

2.1. Participation group

The study group (farmers) was selected with the assumption that they have practical experience with the use of plastics in agriculture because many farming tasks, especially in Ireland, rely heavily on agricultural plastics, no matter what type of enterprise they hold. For this reason, a purposive sampling strategy was adopted. This type of sampling is employed to look for information-rich participants who share certain characteristic(s) related to the research topic (Barglowski, 2018). In order to meet the inclusion criteria of participation, all participants of the study owned or worked full/part-time on a farm in the Republic of Ireland. In addition, all participants had to be over the age of 18 and capable of giving informed consent to take part in the study. In contrast, respondents who did not identify as farm owners or workers and those who were not farming in the Republic of Ireland were excluded from the study. Farmers under the age of 18 or incapable of giving informed consent were also excluded. No groups of farmers from specific farming enterprises alone (i.e. dairy farmers or tillage farmers) or farmers from specific locations of the country were targeted as the aim of the survey was to get a broad representation of the Irish farming community as a whole. Before any data collection commenced, the study underwent a thorough institutional ethical review following the procedures of the Declaration of Helsinki. Active informed consent was obtained from respondents who participated in the survey and full anonymity to all participants of the study is ensured. All precautions and safeguard mechanisms of data security and storage were/are taken by the researchers.

2.2. Survey structure

The survey was designed in five sections. The first section included questions collecting quantitative data on the socio-demographics of participants (age, gender, position on the farm, full-time/part-time, number of years farming, educational level), and questions related to their agricultural production systems (farm type, farm size, farm location). The second section contained questions on the usage and disposal of agricultural plastics, including questions relevant to assessing the current state of agricultural plastic usage in Ireland and what the attitudes of participants were towards these types of plastics. In this section, to collect quantitative data, farmers were asked, "During your time as a farmer, have you noticed an increase in the use of plastics in agriculture?", where they were given the option to select either "Yes", "No" or "I don't know". This was followed by a Likert-type question "How concerned are you about the amount of plastic used in farming activities?" with "Extremely concerned", "Concerned", "Somewhat concerned" and "Not at all concerned" available for choice to participants. To further assess farmers' attitudes towards the usage of agricultural plastics, the following open-ended question was used for the collection of qualitative data "Please list below two words/phrases you associate with 'farm plastics' (either positive or negative)". Respondents were asked to answer multiple-choice questions (to collect quantitative data) around the disposal of several different agricultural plastics. The first question on the disposal of agricultural plastics was phrased "How do you dispose of the following plastics (silage wrap and sheeting, netting, twine, fertiliser and feed bags) used on the farm?" with the following options presented to participants "Pay for disposal", "Pay for recycle", "On-farm disposal", "Reuse on farm" or "Other". Following this, respondents were asked to elaborate further on their mode of agricultural plastic waste disposal in which qualitative data was collected using open-ended questions, "If other, please specify in the space below" and "If plastics are disposed of on the farm, which method is used?". A series of questions were included to capture the reasons behind farmers not recycling their agricultural plastic waste. Again, both quantitative and qualitative data were collected here. A multiple-choice question to collect the former, "In relation to the plastics that you do not recycle, what are your reasons for not recycling these?" The following choices were available to respondents "Too expensive", "Don't know how to", "Lack of facilities", "Contamination (not accepted)", "Not enough generated to recycle" and "Other". Respondents were given the option to provide additional information qualitatively "If other, please specify in the space below". Following this, respondents were asked if they "Agree", "Disagree" or are "Unsure" with the statement "Recycling farm plastics is convenient". Next, to test the knowledge that farmers have on the recyclability of agricultural plastics, the following statement was presented "All farm plastics are recyclable" to which they had the option to respond to it with "Agree", "Disagree" or "Unsure". The final question in this section was a similar style to the previous. Respondents were asked if they "Agree", "Disagree" or are "Unsure" about the statement "The disposal of farm plastics is a big environmental problem". The third section of the survey consisted of questions on biodegradable agricultural plastic use in Ireland, however, for the purpose of this article, data collected on that topic will not be included. The questions included in the fourth section aimed to verify the farmers' awareness of microplastics and plastic pollution in both aquatic and terrestrial systems. This was measured quantitatively using a multiple-choice question "Prior to this survey, had you heard of the term 'microplastic'" with the options "Yes", "No" or "I don't know" available to participants. A Likert-type question was presented to participants "Please indicate to what extent you are aware of the following: "Plastic pollution in the oceans", "Plastic pollution on land (incl. farmlands)", "Microplastic pollution in the oceans" and "Microplastic pollution on land (incl. farmlands)". Respondents had a choice to select either "Very aware", "Aware", "Somewhat aware" or "Not aware" for each of the categories. The final section of the survey sought to determine the perceptions of farmers on the impact of plastics on the environment. Respondents were asked "How serious of a threat do you think plastic pollution poses for each of the following: "Oceans", "Freshwaters (e.g. lakes and rivers)", "Land (incl. farmlands)", "Soils (incl. agricultural soils)", "Marine and freshwater wildlife (e.g. fish, seabirds, etc.)", "Farm animals", "Soil animals (e.g. earthworms etc.)", "Wild plants", "Crops" and "Humans". Again, a Likert-type question was included to collect quantitative data, which presented the following choices available to participants: "Very serious", "Serious", "Somewhat serious" and "Not at all serious".

2.3. Sample size

The minimum sample size of participation for the final administered questionnaire was 384 observations based on Cochran's sample size formula for categorical data, which was previously employed by Bartlett et al. (2001). This considers a 95 % confidence level and a standard level of precision at 0.5. According to figures provided by the Central Statistics Office of Ireland, the sample population (Irish farmers) was 278,600 during the time the survey was developed (CSO, 2022a)

2.4. Data collection

The collection of survey responses was carried out between July and October 2020. The survey was disseminated through an online platform (Microsoft® Forms® 2016). The link to the questionnaire was distributed via email through a variety of social/personal agricultural networks. In addition to this, a link to the survey was shared with social media platforms and farming print media. Hard copies of the survey were made available in order to cater for farmers who were inexperienced with digital platforms or/and those with limited or no online access.

2.5. Data analyses

Data were cleansed using Microsoft® Excel® 2016. Initially, data were cleansed to remove any replicate responses or responses received from outside of the Republic of Ireland. All unintelligible open-ended answers were removed from the data set to ensure data quality. Post-data cleansing, 430 surveys remained for analyses. All quantitative analysis was done using Minitab® 20.3. Open-ended answers were analysed qualitatively using inductive content analysis (Vaismoradi et al., 2013) using Microsoft® Excel® 2016. This was done to investigate the patterns of words and phrases in order to formulate concepts and themes to answer the main research goals of the study. A wordcloud was generated on Wordart.com. Wordclouds are used as a way to display text data in a graphical form and typically visually represent word frequency (Atenstaedt, 2012; DePaolo and Wilkinson, 2014). Words listed by at least two respondents were only included for this analysis (Yeganeh et al., 2020). A top-down approach was used to group words into one of the four categories (negative connotations; positive connotations; neutral; or ambiguous) (Dilkes-Hoffman et al., 2019). The word cloud shows the negative words associated with agricultural plastics in red, positive words in blue and neutral/ambiguous words in green. The size of the words reflects the frequency of occurrence in the data. The positive and negative words collected were later analysed using a bottom-up approach to identify additional groupings. Pearson's chi-squared tests for independence were used to determine whether there were statistically significant differences between the expected and the observed frequencies in categorical variables.

3. Results and discussion

In total, 430 survey responses were taken into account for the final analyses of the study. However, although the majority of respondents completed the full survey, some did not answer all questions but were included in most of the analyses presented here.

3.1. Respondent demographics and agricultural production systems characteristics

The majority of respondents were male (82.9 %) and aged between 25 and 39 years of age (33.6 %) (Table 1). The high percentage of male to females involved in the farm labour workforce are in line with what was expected. Eurostat figures on the breakdown of farmers in the EU showed that on average, women accounted for 35.1 % of the agricultural workforce (Eurostat, 2017). The Census of Agriculture 2020 reports that 26.9 % of the Irish agriculture labour work force are female (CSO, 2022a). A higher number of respondents who took the survey were under 40 years of age, however, most (55.3 %) of the farmers in Ireland are 55 years of age or more and only 5.3 % represent the under 35 years category (CSO, 2018).

The majority of responses came from the Border (33.5 %) region of Ireland, followed by Midland (21.7 %), Mid-East (13.0 %), Western (10.8 %), Mid-West (7.3 %), South-East (6.8 %) and South-West (6.6 %). Responses were received from all the 26 counties of the Republic of Ireland. Most respondents identified as the farm owner/manager (80.9 %), with the rest of respondents identifying as farm workers (19.1 %). The number of respondents who reported as working either full-time or part-time was 44.8 % and 55.2 % respectively, with the highest proportion reporting that they have been working in farming for >20 years (46.1 %). The top three main farming enterprises were beef production (39.5 %) followed by dairy (24.5 %) and mixed grazing livestock (14.2 %). However, in Ireland, beef production systems represent 56.4 % of the farm types, followed by dairy and sheep (CSO, 2022b). Respondents reported they owned or worked on farms between 26 and 50 ha in size the most (29.5 %), followed by farms of 51–75 ha (23.1 %). The average farm size in Ireland is approximately 32.4 ha (CSO, 2022b). The majority of respondents reported they had completed at most a level 5/6 (agricultural cert/green cert) (38.3 %), which followed by level 8+ (honours degree or higher) (21.4 %), and 16.9 % received no formal agricultural training.

Table 1
Respondent demographics and characteristics of their agricultural production systems.

Item	No.	Percentage (%)
Gender (N = 387)		
Male	321	82.95
Female	66	17.05
Age range (N = 360)		0
18–24	72	20.00
25–39	121	33.61
40–49	71	19.72
50–59	56	15.56
60–69	33	9.17
70–79	7	1.94
80+	0	0
Region of Ireland (N = 423)		
Border	142	33.57
Midland	92	21.75
Western	46	10.87
Mid-East	55	13.00
Mid-West	31	7.33
South-East	29	6.86
South-West	28	6.62
Position on farm (N = 404)		
Farm owner/manager	327	80.94
Farm worker	77	19.06
Working (N = 415)		
Full-time	186	44.82
Part-time	229	55.18
Duration farming (N = 423)		
0–5 years	40	9.46
5–10 years	75	17.73
10–15 years	64	15.13
15–20 years	49	11.58
20+ years	195	46.10
Main farming enterprise (N = 427)		
Beef production	169	39.58
Dairy	105	24.59
Mixed crops livestock	22	5.15
Mixed grazing livestock	61	14.29
Sheep	44	10.30
Tillage	20	4.68
Other	6	1.41

3.2. Attitudes towards plastic usage and disposal methods of agricultural plastics in the Irish agricultural sector

3.2.1. Agricultural plastic use is increasing and most farmers are concerned

Respondents were asked if, during their time as a farmer, had they noticed an increase in the use of farm plastics, to which 79.6 % reported 'Yes' they have, 14.3 % answered 'No', leaving 6.1 % who did not know (Fig. 1a). When considering that more than half of the respondents have been farming for over twenty years, it is no surprise that almost 80 % of respondents reported to seeing an increase in the use of plastics in agriculture. Plastics have been used for agricultural practices since the 1950s, and overtime agriculture has become increasingly intensive, resulting in agricultural plastics becoming more available to farmers (Robinson and Sutherland, 2002). While there is no regional data available on the coverage or quantity of agricultural plastic in Ireland, in the European Union around 1.7 million tonnes of agricultural plastics were used in 2018, which is between 3 and 4 % of the total converter demand of European plastic usage. Global forecasts predict that plastics used for greenhouses, mulches and silage films are set to increase from 6.1 million tonnes in 2018 to 9.5 million tonnes in 2030 (FAO, 2021). In this study, the results showed that 88.3 % of respondents

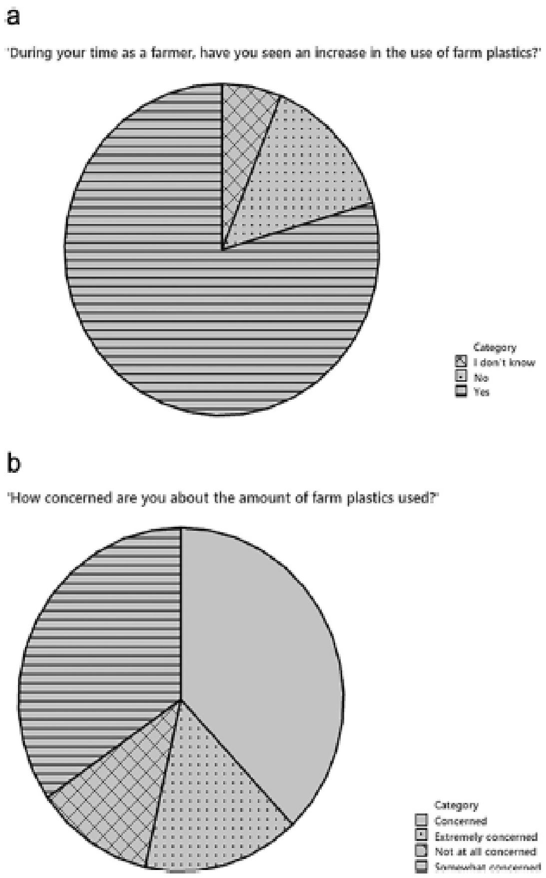

Fig. 1. The total percentage of farmers noticing an increase in the use of farm plastics since they began farming (a). Respondents concern about the amount of plastics used in agriculture (b).

Table 2
Responses to 'Please list two words/phrases you associate with the words farm plastic'.

Main category	Count (number of words)	Percentage (%)
Negative connotations	329	65.8
Positive connotations	117	23.4
Neutral connotations	19	3.8
Ambiguous connotations	35	7.0
Total	500	100

Table 4
Content related to the positive connotations listed.

Additional category (Positive connotations)	Count
Related to the environment (e.g. recyclable, reusable)	17
Related to material properties and function (e.g. clean, durable)	52
Related to accessibility (e.g. available)	4
General positive associations (e.g. convenient, important)	44
Total	117

expressed some level of concern about the amount of plastics used for farming activities, with 15.6 % extremely concerned and 11.8 % who are not at all concerned (Fig. 1b). A decline in concern is evident among younger cohorts, with only 9.8 % of farmers aged under 40 stating they are extremely concerned, in comparison to 25 % of farmers over the age of 50 years. This result ties in well with a recent study carried out by the Irish Environmental Protection Agency (EPA) on the attitudes and behaviours towards single-use plastics in Ireland. Over half of the sample population stated they were 'very concerned', and only 5 % stated that they are not at all concerned with the amount of plastic used as a society. Their results also show that age may impact the attitudes of society in relation to amount of plastic used, with more adults over the age of 65+ feeling more concerned about it (EPA, 2022). In this study, the region ($p = 0.035$) and size ($p = 0.006$) of the farm respondents own or work on was seen to make a statistically significant difference in how concerned they were about the amount of plastics used for farming activities. For example, farmers owning or working on smaller sized farms were more concerned about the amount of plastics used in farming activities, compared to those on bigger farms. In relation to the region the farmers were located; farmers in the Border and Western regions of Ireland were more concerned about the amount of plastics used in agricultural activities. This makes sense because typically farms on the West of the country are smaller than farms in the Midland and Eastern regions.

3.2.2. *Agricultural plastics are necessary for farm tasks, but are bad for the environment and have monetary and labour costs*

Respondents were asked to 'Please list two words/phrases you associate with the words: farm plastics (either positive or negative)'. In total, 349 responses, which amounted to the collection of 500 words, were counted and analysed. Responses mostly fell into the negative connotations category (65.8 %), which is followed with 23.4 % of responses falling into the positive connotations category. The words considered ambiguous and neutral consisted of 7 % and 3.8 % of the responses respectively (Table 2). Additional inductive content analysis was carried out to identify concepts and themes within the negative and positive connotations categories. In relation to the negative words collected, responses were mostly related to the environment (147), followed by general negative associations (95), words related to time and labour constraints (36), cost (32) and accessibility (19) (Table 3). In the positive connotations category, most positive words provided were related to the material properties and function of agricultural plastics (52), followed by general positive associations (44), words related to the environment (17) and accessibility (4) (Table 4). The majority (67.6 %) of respondents listed only negative words, followed by 15.3 % who chose only positive words, and 14.4 % who included both one positive and one negative word in their answers. The remainder of respondents chose only neutral or ambiguous words for their response (Table 5). Perhaps, as expected, some respondents (14.8 %) only negatively associated agricultural plastics with the cost factors, including monetary, time and labour constraints. Some participants stated 'Costly. Difficult to manage when it piles up in springtime, dirty, messy wet.' (P 187), 'The cost of disposal and the amount of room it takes up after being used' (P 214), and 'Expensive to buy and expensive to dispose of' (P 235). However, more farmers (29.4 %) only associated agricultural plastics with the negative impact they have on the environment. Some participants included more general comments such as 'Bad for the environment' (P 19), 'Environmental disaster' (P 125) and 'Long life in the environment' (P 118), but some were more specific. One participant responded with 'Eyesore and environmental issue as it blows across fields and see it on the roads' (P 61), another with 'Blowing everywhere' (P 178), and 'Blowing in the wind' (P 176). Others stated 'Harmful to environment when not disposed of in the correct manner, ending up in sea beds etc.' (P 72) and 'Visible in every ditch in the country (and) will be an issue in the food chain in years to come' (P 124). Moreover, other results show there is a high level of agreement among respondents on the disposal of agricultural plastics presenting a big environmental problem, with 82 % of respondents agreeing with this statement. Age ($p = 0.041$) was seen to have a statistically significant effect on farmers' perception of agricultural plastics presenting a big environmental problem, with younger farmers more in agreement than older farmers. While an overwhelming number of respondents solely focused on their negative associations with agricultural plastics, many respondents expressed conflicted attitudes. For example, 14.4 % of farmers responded with both one positive and one negative association. In some these cases, many farmers believe that agricultural plastics are negative due to the cost factors associated with them and their impacts on the environment. Yet they also acknowledge that plastics are positive and a necessity due to their functionality on the farm. Seven participants commented that agricultural plastics are a 'Necessary evil'. One participant responded with '(A) necessary evil, (but) what else would I cover the silage pit with?' (P 430) and another with 'Good for ensiling grass or maize, however in some cases is not disposed of in an environmentally friendly manner, leading to pollution' (P 107) (Fig. 2).

3.2.3. *Agricultural plastic disposal methods depend on the type of plastic, cost and access to facilities*

Disposal methods vary by the type and composition of the plastic with some being easier to dispose of. Of all the types of agricultural plastics, bale wrap is the type mostly recycled by participants (58.1 %). Education ($p = 0.038$) was seen to make a statistically significant effect on how

Table 3
Content relating to the negative connotations listed.

Additional category (Negative connotations)	Count
Related to the environment (e.g. pollution, waste, non-recyclable)	147
Related to cost (e.g. expensive, extra-cost)	32
Related to time/labour constraints (e.g. extra-work, time-consuming)	36
Related to accessibility (e.g. excessive, no-alternative)	19
General negative association (e.g. dangerous, evil, difficult)	95
Total	329

Table 5
The breakdown of the number of times only negative or positive words were listed; both one negative and one positive word were listed together; and neutral/ambiguous listings.

Type of response	Count (number of respondents)	Percentage (%)
Only negative word(s) listed	237	67.6
Only positive word(s) listed	53	15.3
Both one negative and one positive word listed	50	14.4
Neutral/ambiguous word(s)	9	2.7

Fig. 2. Most commonly listed words respondents associated with the words 'Farm plastics'. The font size of the words reflect the frequency of responses.

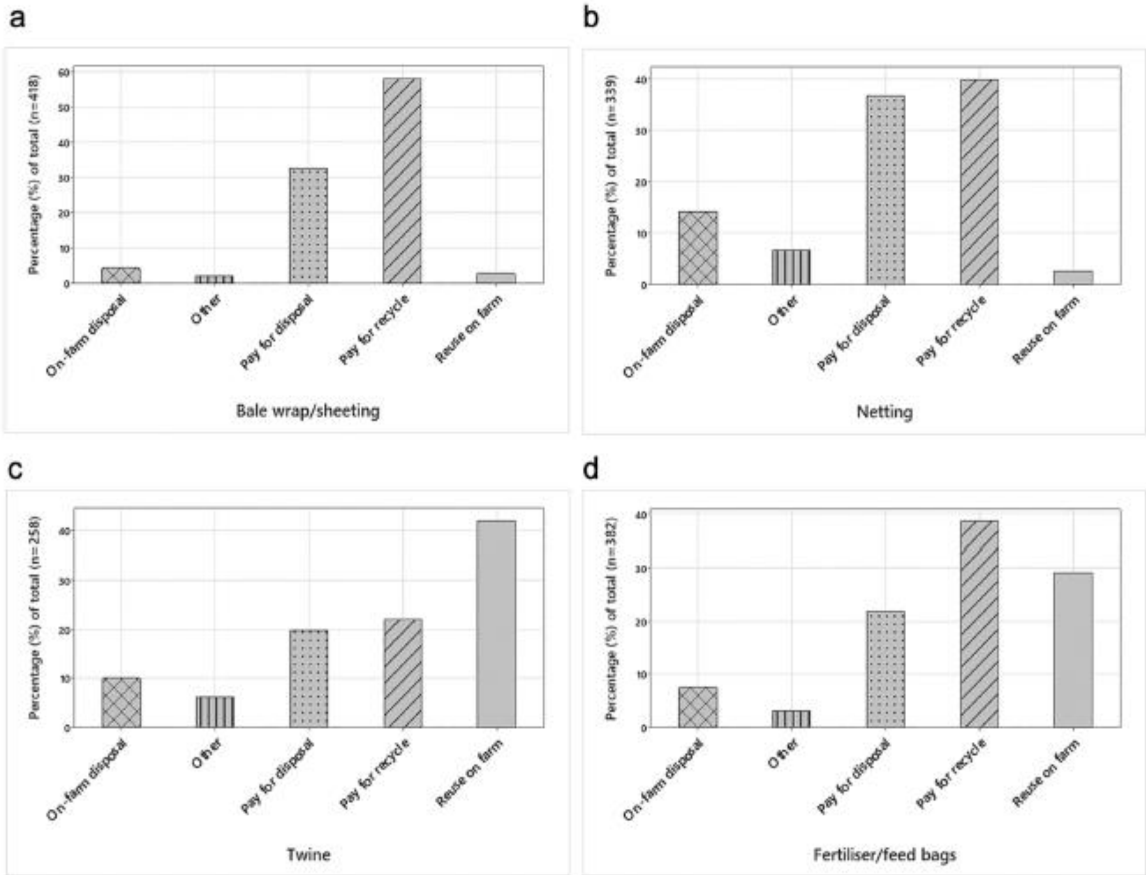

Fig. 3. Farm plastic disposal methods for each type of plastic (a) bale wrap/sheeting, (b) netting, (c) twine and (d) fertiliser/feed bags.

farmers dispose of bale wrap. A higher level of education tends to lead to a higher level of recycling and reusing. Other agricultural plastics widely sent for recycling include netting (41.4 %) and fertiliser and feed bags (38.5 %). The data shows that twine may be considered the most difficult to recycle, with only 20.1 % recycling twine, however, more farmers (41.8 %) find alternative uses for twine on the farm. Some expressed that they find their own means of disposal on the farm or they use 'other' methods, which was mostly the case for netting (20.9 %) and twine (16.2 %) (Fig. 3). This may be because there are minimum weight requirements for acceptance of agricultural plastics at the national farm plastics recycling compliance scheme. The farmer is charged per half tonne for each type of agricultural plastic they choose to recycle, and as netting and twine are typically lightweight plastics, generating enough waste may be challenging on different farms.

Under current Irish legislation, (S.I. No. 396/2017 - Waste Management (Farm Plastics) (Amendment) Regulations 2017) farmers have an obligation to recycle the farm plastic material waste they generate. However, when respondents were asked to elaborate on what types of 'other' or 'on-farm' methods were used, 14 % (n = 63) volunteered information stating they either burn or bury certain agricultural plastic waste (Fig. 4). One respondent stated they 'Recycle wraps and sheeting, (but) pit burn twine and net' (P 307), and another said 'Netting is the only thing disposed of on the farm, it is burned' (P 43). Burning plastic in open-field is illegal under the Waste Management (Prohibition of Waste Disposal by Burning) (Regulations 2009), as it releases toxic gases into the environment, including substances such as dioxins, and furans. Other by-products of burnt plastic (soot and ash) can cause health and environmental impacts through the release of volatile organic compounds, particulate matter, particulate bond heavy metals, and polycyclic aromatic hydrocarbons (PAHs) which travel depending on atmospheric conditions, settling on crops in neighbouring fields and entering waterways, potentially making their way into the food we eat (Kumar et al., 2022). Moreover, while some farmers understand that doing this is 'wrong', disposal methods need to be convenient and cost-effective in order to motivate farmers to manage agricultural plastic waste effectively in an environmentally sound manner.

There seems to be some misconception among some farmers about the recyclability of certain agricultural plastics. One participant responded that they 'Burn or bury netting and fertiliser bags as they can't be recycled' (P 376). The national agricultural plastics recycling compliance scheme allows for the recycling of silage bale wrap and sheeting, netting, twine, fertiliser and feed bags, drums and containers. Other plastics used on the farm such as plastic mulch films and piping are not accepted for recycling. In a separate question, results showed that 41 % of farmers agree that 'All farm plastics are recyclable', but 22 % disagree and 37 % are unsure whether they are. Age (p = 0.040) made a statistically significant difference on the whether farmers think that all farm plastics are recyclable.

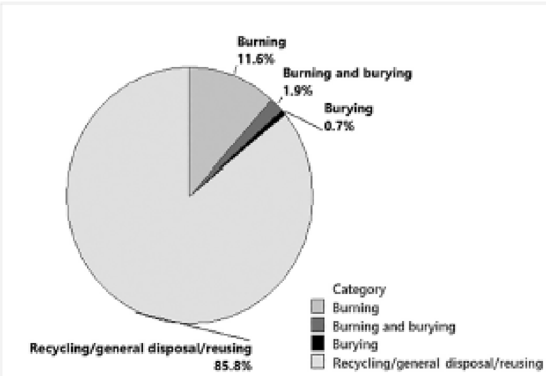

Fig. 4. The percentage of farmers burning and burying certain farm plastic types.

For example, more farmers older than fifty years of age think that all agricultural plastics are recyclable in comparison to farmers under the age of 40. However, a higher number of younger farmers are unsure if they are all recyclable. These results provide evidence that there are gaps in the level of knowledge some farmers have around the recyclability of agricultural plastics in Ireland, which present several implications. First, if farmers are unaware on how to recycle agricultural plastics, it may discourage them trying which may ultimately lead to them choosing a different method of disposal, for example burning or burying on the farm. Second, not all agricultural plastics are recycled in the same way. There is a different method for recycling silage plastics in comparison to recycling fertiliser and feed bags. Prior to the recycling of agricultural plastics, there are certain separation criteria that must be adhered to and if farmers are unaware of this, they may be storing and separating incorrectly, which can cause problems down the line for the collectors and recyclers of agricultural plastics. For example, if a person brought all their agricultural plastic wastes mixed together to the recycling depot, this would slow down the process and/or those plastics may potentially become unfit for recycling and thus not accepted. Farmers incorrectly storing and separating plastics may be charged extra at recycling depots which may discourage them from recycling agricultural plastics thereafter. Due to the hydroscopic nature of certain agricultural plastics, they can retain water, but also, the plastics may be covered in bits of dust, grit and soil, which will result in a heavier weight. To ensure extra charges are not applied, farmers must be aware of the consequences of poor storage conditions. It may be of benefit if the media (such as national/local farming newspapers and radio stations) publish or announce notices regularly on how to store and separate agricultural plastics correctly for recycling all throughout the year. In addition to this, it may be of benefit if notices are displayed in local marts, co-ops, and shops to encourage and educate farmers on how to recycle agricultural plastic waste. It is important to consider that certain groups of farmers may use different sources of information. Läpple (2013) found that organic farmers used advisory services more often than conventional farmers but no differences were found between the two groups in terms of the use of media information. Another strategy to help improve farmers' knowledge and confidence on the recycling of agricultural plastics may be through education and training surrounding agricultural plastic waste stream management and recycling. It may be of benefit if agricultural courses include content on how to manage and recycle agricultural plastics. Moreover, content on the implications of poor agricultural plastic management, such as plastic and microplastic pollution and their potential impacts in agro-ecosystems should also be added to these courses to help improve the current state of knowledge on these topics.

A follow up section was included to try to understand why agricultural plastic waste is not being recycled by some farmers. The most commonly stated primary reasons farmers do not recycle agricultural plastics are due to the following factors: (1) a perception of a lack of facilities available to them, (2) they feel they do not generate enough agricultural plastic waste to recycle at the standard cost, and (3) they do not know how to recycle agricultural plastics. As expected, many farmers (30.9 %) reported twine as the most difficult type of agricultural plastic to generate an adequate amount of waste for acceptance at recycling facilities. Moreover, to a lesser extent, contamination issues were also among the reasons reported as to why farmers do not recycle agricultural plastics, with bale wrap and netting considered the most difficult to keep free from contamination (Fig. 5. (a) (b)). Age (p = 0.030) has a statistically significant effect on why farmers do not recycle agricultural plastics, with older farmers believing there are a lack of recycling facilities available to them, however, younger farmers perceive the cost of recycling to be the main barrier. Typically, in Ireland, in any given jurisdiction of the country, which would serve up to thousands of farmers, there are only a set number of days available each year where farmers can recycle their agricultural plastic waste. Due to transportation and monetary costs, farmers may be less inclined to travel to the recycling depots to avail of the services. Forty one percent of farmers reported that recycling agricultural plastics is inconvenient for them, while 46 % think it is convenient and 13 % were unsure. The size of the farm (p = 0.011)

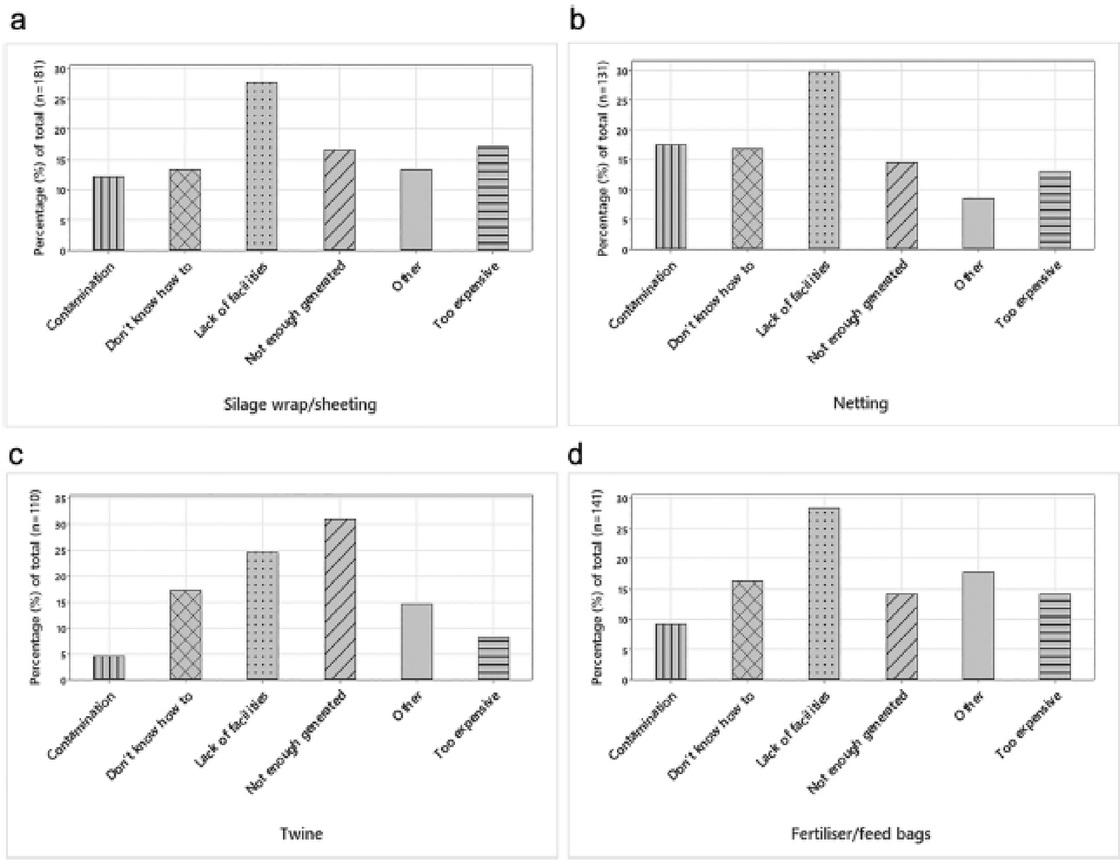

Fig. 5. Reasons farmers are not recycling farm plastics: (a) bale wrap/sheeting, (b) netting, (c) twine and (d) fertiliser/feed bags.

and the region ($p = 0.021$) farmers worked had a statistically significant difference on their attitude towards the convenience of recycling agricultural plastics. Farmers occupying larger farms find it more convenient to recycle agricultural plastics than those who own or work on smaller sized farms. Again, this may be due to the minimum weight cap placed on the amount of plastic that is accepted at recycling centres. Larger sized farms may have more production, therefore generating enough plastic waste to recycle on bigger farms may not be an issue in comparison to smaller farmers that generate less plastic waste. With regards to location, farmers in the Western regions of the country stated that they find recycling agricultural plastics less convenient than farmers in the Midlands and Eastern regions. The size of farms in the Midlands and in the East of Ireland are on a much bigger scale than farms on the West which supports the idea that it is more difficult for farmers working on smaller holdings to recycle agricultural plastics. Another factor, which may affect the convenience of recycling agricultural plastics in the West, is potentially due to poorer roads and public infrastructure in this region of Ireland.

3.3. *Farmers' awareness of plastic and microplastic pollution and their perceptions of the environmental impacts*

Many farmers (57.7 %) reported they had previously heard of the term 'microplastic' prior to taking the survey. As expected, education had a statistically significant difference on the awareness of farmers towards microplastics ($p = 0.016$). In addition, while most respondents are, at some level aware of plastic and microplastic pollution in the oceans and on land, there is greater awareness of plastic pollution and less awareness of microplastic pollution (Fig. 6.). This is no surprise because macroplastics are visible and microplastics are mostly 'invisible'. Furthermore, research articles and media reports on plastic pollution started earlier than those on microplastics. Interestingly, respondents reported that they are more

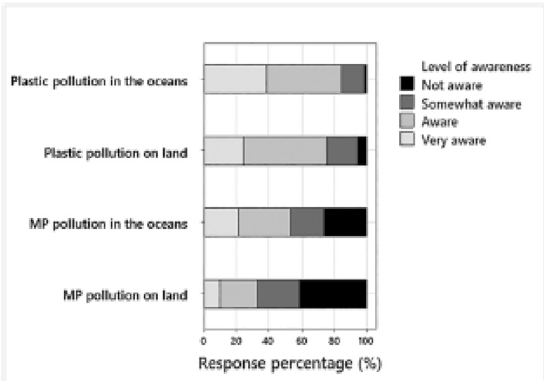

Fig. 6. Respondents' level of awareness of plastic and microplastic pollution in the ocean and on land.

aware of plastic and microplastic pollution issues in the ocean than on land. Although farmers are out working on the land every day, they may be unaware of the issues on land because again, most research articles and media attention has been mainly focused on the aquatic environments (Jenkins et al., 2022). Increased emphasis must be placed on the occurrence and impacts of microplastics on-land and in soils by the media in order to strengthen the publicity and education of relevant knowledge of microplastics in these terrestrial systems; however, this is only possible if adequate scientific research has been done to inform the media. It can thus be suggested that there is a need to increase the number of research studies on the impacts of microplastics and plastic pollution on terrestrial systems.

As farmers are more aware of plastic and microplastic pollution in aquatic environments, this seems to influence their risk perception. Overall, farmers think that plastic pollution threatens aquatic environments more than the terrestrial systems. For example, more respondents perceive plastic pollution as a bigger threat to the oceans, freshwaters, and marine and freshwater animals in comparison to soil, soil animals, crops and wild plants. The majority respondents (over 80 % for every category) do think that the threats plastics pose are, to some extent serious in all of the ten environmental compartments presented (Fig. 7.). However, over half of the respondents perceive these risks are *very serious* in oceans and freshwaters, compared to less than a fifth who interpret the same level of risk towards components of the terrestrial systems such as soil, wild plants and crops. These results relate to findings by Deng et al. (2020) who found that 43.8 % of respondents believed that microplastics mostly accumulate in the oceans, followed by animals and plants (14.2 %), air (13.5 %), rivers and lakes (6.3 %) and soil (4 %). Filho et al. (2020) also found that most (60 %) of their survey respondents considered the problems associated with plastic in the ocean as *extremely serious*, and <40 % of respondents reported the effects of plastics on soils as *extremely serious*, and even less (20 %) consider the effects of plastics on air as *serious*. Another interesting finding to our study is that over 70 % of respondents feel that farm animals are at serious risk from plastic pollution. Some respondents reported that agricultural plastics are 'Dangerous towards livestock' (P 155) and that 'Animals (are) eating the plastics' (P355). Rumen impaction due to foreign bodies such as plastic materials can cause many different problems for animals. The ingestion of these materials can hinder physiological and chemical processes such as fermentation, which can lead to indigestion and microflora disruption. In addition to this, plastic ingestion over time may lead to a build-up of toxins in the animal, which in turn could affect meat and milk quality intended for human consumption (Akraiem and Abd Al-Galil, 2016). Despite findings showing that annual plastic waste released into the environment by land-based sources is estimated as 4 to 23 times higher than from marine based sources, our results show that the perception of

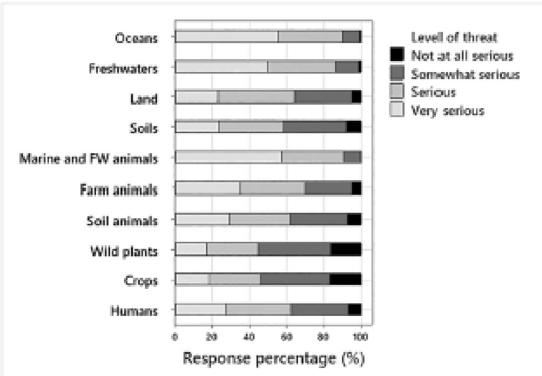

Fig. 7. Respondents' perception of how serious of a threat plastic pollution poses on different components of the environment.

most farmers is that plastic pollution is more damaging to marine and freshwater environments. This reiterates the fact that knowledge of plastic and microplastic abundance and effects in terrestrial systems is still extremely limited (Horton et al., 2017).

3.4. Strategies to combat plastic and microplastic pollution in agricultural soils

At a global level, in the coming decades, decisive changes regarding the implications of plastic pollution urgently need to be undertaken. Currently, there are a lack of specific legislation at a European level on the use of plastics in agriculture. There is also no criteria for sustainable soil management with reference to microplastics contamination in agricultural soils. There are policy developments in the EU coming in 2023 that focus on a new Soil Health Law with the vision that all soils will be 'healthy' by 2050. Two of the eight objectives of the EU Horizon Soil Mission include to "Reduce soil pollution and enhance restoration" and "Improve soil literacy in society" (Correia, 2021). Some of the main findings from our study show that farmers' believe agricultural plastics are a source of pollution; however, they perceive plastics pose a bigger risk to aquatic environments in comparison to the terrestrial, including soils. This indicates that an emphasis on the importance of soils and the pollution of soils by plastics must be delivered to society. Therefore, it is necessary for these new policy developments to include monitoring programmes to assess plastic and microplastic contamination levels in soils and initiatives to promote communication and citizen engagement.

Many farmers' expressed that they use conventional agricultural plastics because there are no alternatives available. There is currently no single EU law in place on biobased, biodegradable or compostable plastics in a comprehensive manner (European Commission, 2022). These materials may offer alternative solutions to the plastic pollution problem, but they also present many challenges. The certifications for these products are limited and unstandardised. Therefore, it is recommended that the new policy frameworks for biobased, biodegradable and compostable plastics should cover the economic and practical viability of these materials for use in agriculture.

3.5. Limitations of the study

Data for this study was collected both online and through hard copies. Collecting survey data online can be advantageous because it is relatively easy to conduct and can be administered via free platforms (Wu et al., 2022). However, there are also several limitations associated with collecting survey data online. The ability to reach certain types of participants can be challenging, and in our case, it was difficult to collect responses from potential participants who were inexperienced with digital platforms and/or those who have limited or no internet access due to living in remote areas. Thus, to capture the attitudes of these individuals, hard copies were made available to retrieve responses. However, the reliability of survey data quality using multiple data collection methods can depend on certain factors. The primary concern is "Are responses to online surveys identical, similar or different to paper copy surveys?". Online survey data collection may result in response bias due to the nature of data collection (Boyer et al., 2001).

Another limitation to the study is the socio-demographic profile of respondents. The number of participants under the age of fifty years represented the majority (73.3 %) of respondents, in comparison to the number of respondents over the age of fifty (26.7 %). However, according to CSO data, 53.3 % of farm holders in Ireland are >55 years old (CSO, 2018). Whilst acknowledging that the population of the study were younger than the average named farmer, the younger generation of farmers will still have a significant impact on the sector for the coming decades and therefore the relevance of the sample population remains high. In addition to this, it is believed that the CSO data is collected based on the person who owns the farm, which is something that is not necessarily the same as the average age of farmers, and this subsequently makes it difficult to quantify the age-dynamic in Irish family farm structures.

4. Conclusions

The main conclusions of this study are:

1. Most farmers are recycling agricultural plastic waste. However, the rate of recycling depends on a wide range of factors including the type of agricultural plastic, cost, accessibility to recycling facilities and their knowledge on what can be recycled and how to recycle it. Initiatives should be put in place to educate farmers on how to recycle farm plastics correctly to help mitigate plastic and microplastic pollution in soils.

2. Farmers acknowledge that they need agricultural plastics to perform tasks on the farm, and that realistically no other material with suffice. However, many farmers view agricultural plastics as a burden due to the logistical and monetary factors associated with them. Farmers are also concerned about the negative impacts that the disposal of agricultural plastics present to the environment. Despite this, awareness and concern towards the environment does not always correspond into positive action. Some farmers openly admitted to the burning and burial of plastic waste on-site, which is not only damaging to the environment, but also is illegal.

3. Farmers are relatively aware of microplastics, but are more aware of plastic pollution than microplastic pollution. In addition to this, farmers feel that aquatic environments are under greater threat than the terrestrial environments are. This demonstrates that farmers understand and care more about the impacts plastics and microplastics have on waterbodies and their counterparts, which might be because most of the research efforts have focused on these systems to date. Further research on the abundance and potential effects of microplastics on soils is needed.

4. Combined efforts by governments, policy makers, and other stakeholders must be undertaken in order to reduce the plastic and microplastic problem. Developments should be made in relation to the policies regarding soil health and this includes the contamination of soil via plastics and the potential impacts plastics have on soil stability and structure. Moreover, governments should set out initiatives to promote citizen engagement to help improve the functionality of agro-ecosystems. Furthermore, new research and innovation into the economic and practical viability of biobased and biodegradable plastics should be addressed to investigate the potential of these materials as alternatives to conventional plastics in agriculture.

CRediT authorship contribution statement

Clodagh D. King: Conceptualisation, Writing, Methodology, Data curation, Data analysis **Siobhan N. Jordan:** Conceptualisation, Reviewing, Editing, Supervision, Funding acquisition **Caroline Gilleran Stephens:** Conceptualisation, Reviewing, Editing, Supervision, Funding acquisition **Joseph P. Lynch:** Conceptualisation, Reviewing, Editing, Supervision.

Data availability

Data will be made available on request.

Declaration of competing interest

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

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Appendix 2. Procedural blanks, airbone contamination and recovery test data from chapter 4.

Appendix 2. Table 1: Reagents tested for microplastics, number of microplastics and microplastic description.

Reagent	Microplastic count	Microplastic description
Milli Q water		
Rep 1	0	
Rep 2	0	
Rep 3	0	
NaCl		
Rep 1	0	
Rep 2	1	Grey fragment (< 200 µm)
Rep 3	0	
Zn_2Cl_2		
Rep 1	1	Black fibre (200-300 µm)
Rep 2	0	
Rep 3	0	

Appendix 2. Table 2: Microplastic airborne contamination (counts and description) during laboratory analysis.

Filter	Location	Microplastic count	Microplastic description
1	Main prep area	0	
2	Fume hood	0	
3	Main prep area	0	
4	Fume hood	0	
5	Fume hood	0	
6	Main prep area	1	Transparent fibre (2340 µm)
7	Fume hood	0	
8	Main prep area	0	
9	Fume hood	0	
10	Main prep area	0	
11	Fume hood	0	
12	Main prep area	0	
13	Fume hood	0	
14	Main prep area	0	
15	Fume hood	0	
16	Main prep area	0	
17	Fume hood	1	Transparent fibre (780 µm)
18	Main prep area	0	
19	Fume hood	0	
20	Main prep area	0	
21	Fume hood	0	
22	Fume hood	0	
23	Main prep area	0	
24	Fume hood	0	
25	Fume hood	0	

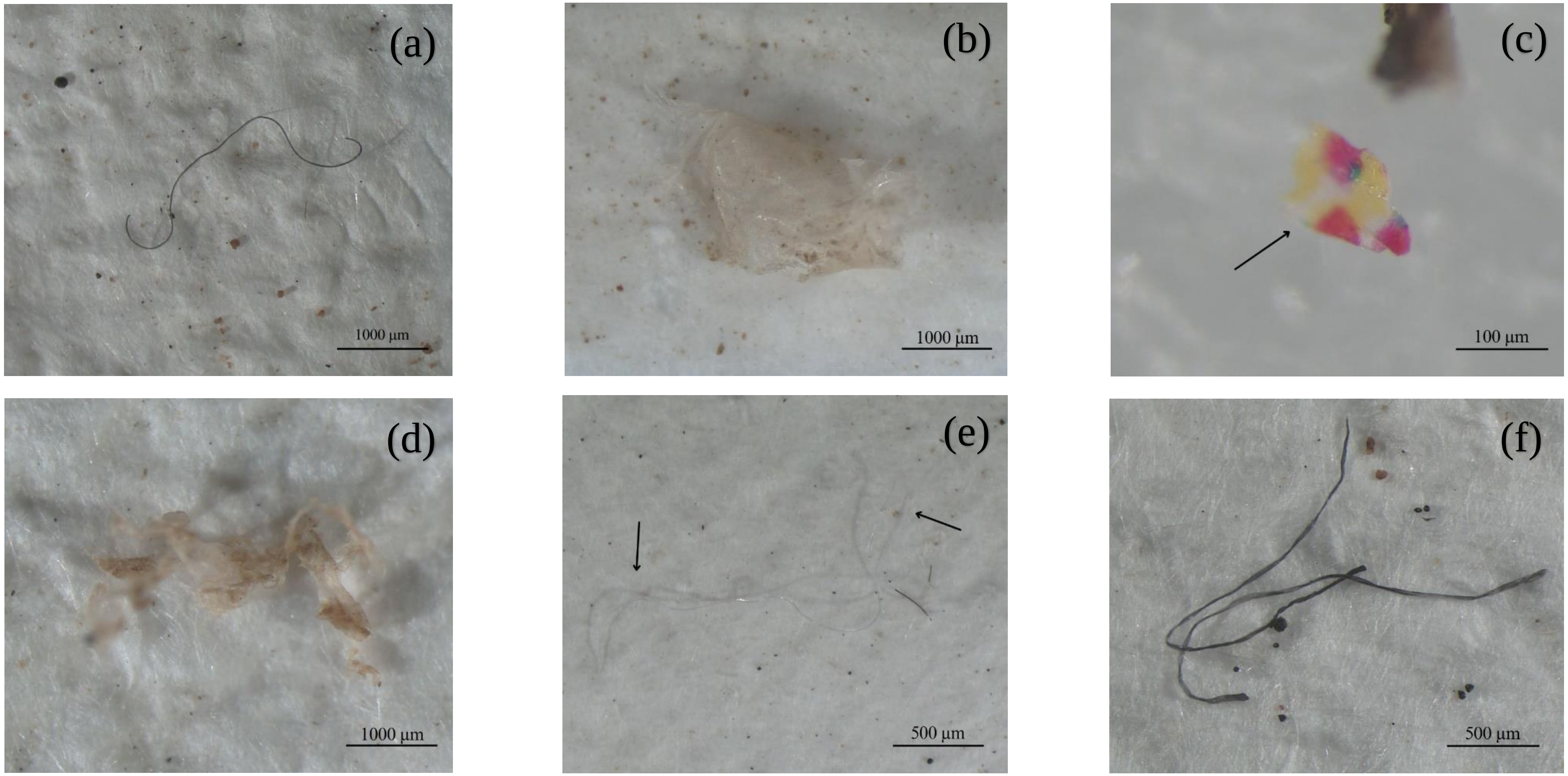
26	Main prep area	0	
27	Main prep area	0	
28	Fume hood	0	
29	Main prep area	0	
30	Fume hood	0	
31	Microscopy area	0	
32	Microscopy area	0	
33	Microscopy area	0	
34	Microscopy area	1	Transparent fibre (3927 µm)
35	Main prep area		
36	Fume hood	0	
37	Main prep area	0	
38	Fume hood	0	
39	Main prep area	0	
40	Fume hood	0	
41	Main prep area	0	
42	Microscopy area	0	
43	Microscopy area	1	Transparent fibre (1439 µm)
44	Main prep area	0	
45	Fume hood	0	
46	Main prep area	0	
47	Fume hood	0	
48	Main prep area	0	
49	Main prep area	1	Transparent fibre (2731 µm)
50	Fume hood	0	
51	Microscopy area	0	
52	Main prep area	0	
53	Microscopy area	0	
54	Microscopy area	0	
55	Microscopy area	0	
56	Microscopy area	0	

57	Microscopy area	2	Black fibre (1442 µm), transparent fibre (928 µm)
58	Microscopy area	0	
59	Microscopy area	0	
60	Microscopy area	0	
61	Microscopy area	0	
62	Microscopy area	1	Blue fibre (984 µm)
63	Main prep area	0	
64	Fume hood	0	
65	Main prep area	0	
66	Main prep area	1	Transparent fibre (3200 µm)
67	Fume hood	0	
68	Main prep area	0	
69	Fume hood	0	
70	Microscopy area	0	
71	Microscopy area	0	
72	Microscopy area	0	
73	Microscopy area	0	
74	Microscopy area	0	
75	Microscopy area	1	Transparent fibre (1327 µm)
Total		**10**	

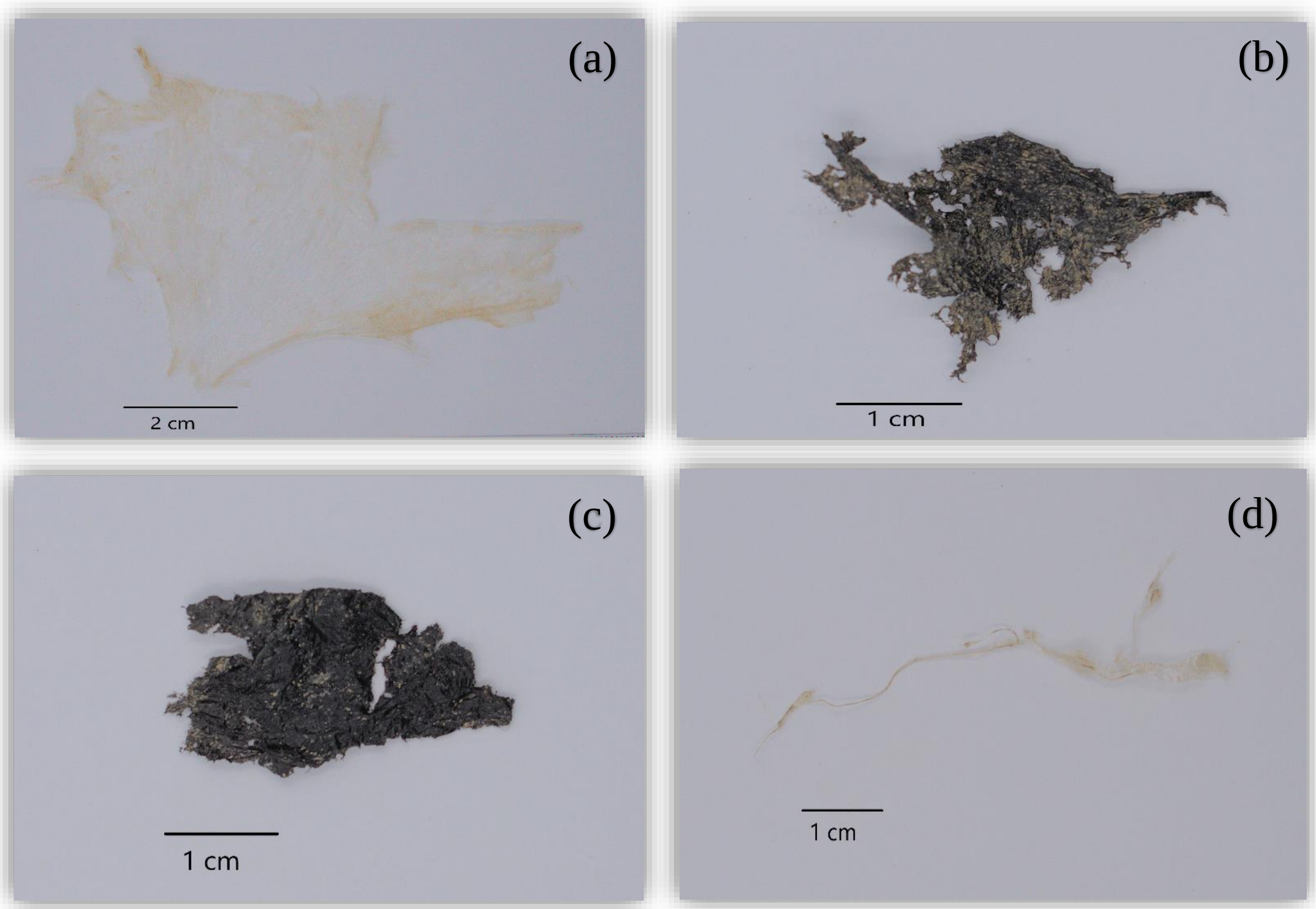
Appendix 2. Table 3: Recovery tests data for validation of density separation method.

Sample	PP	PE	PC	PES
Spike-1				
Rep1	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Spike-2				
Rep 1	6 (100 %)	5 (90 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Spike-3				
Rep 1	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	5 (90 %)	6 (100 %)	6 (100 %)	6 (100 %)

Appendix 3. Sample photos of micro, meso and macroplastics found in agricultural soils

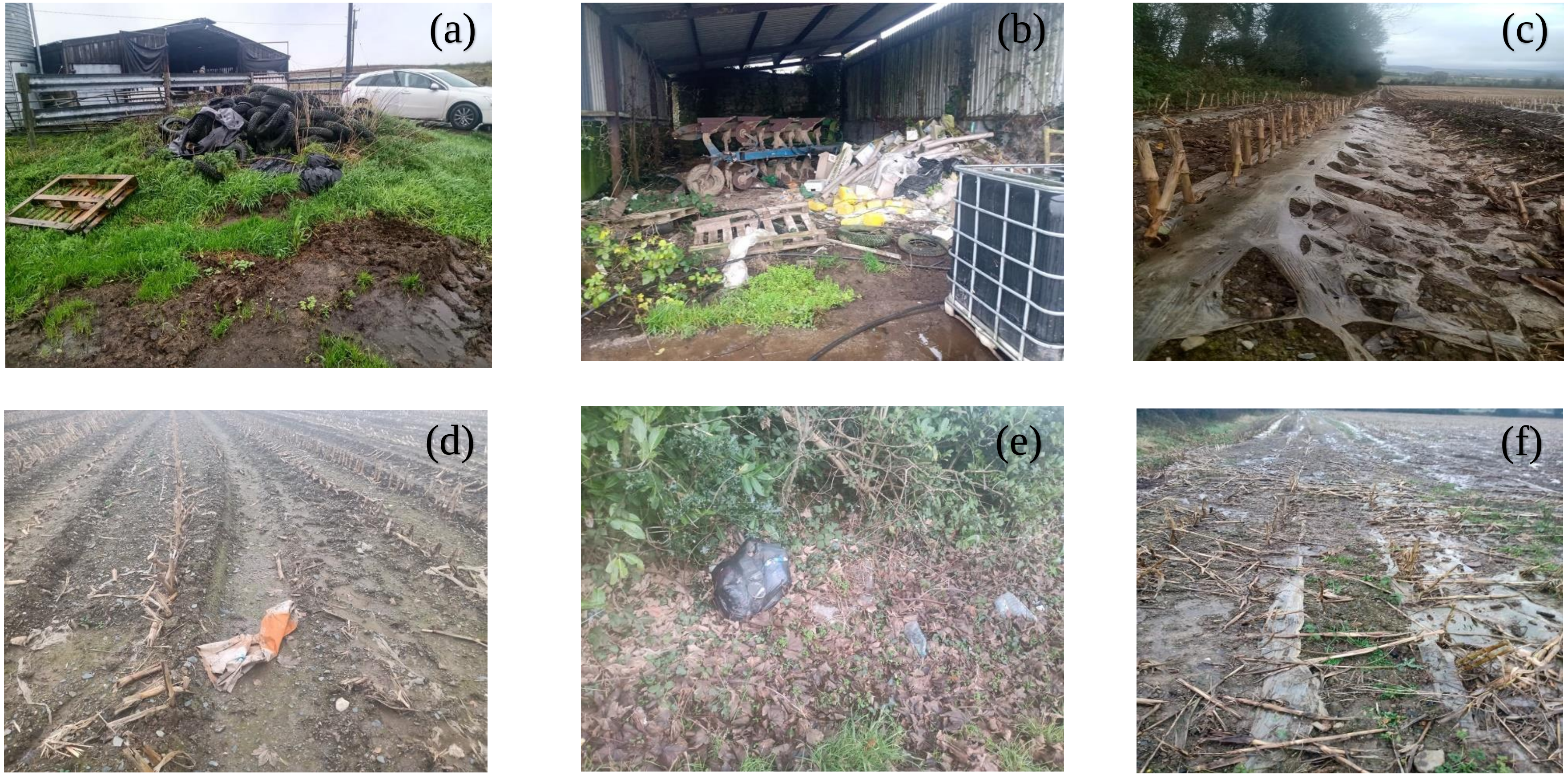


Appendix 3: Figure 1. Microplastics found in agricultural soils sampled. (a) Black fibre (PET) (4000 – 5000 µm) (b) Transparent film (PE) (2000 – 3000 µm) (c) Multicoloured fragment (160 µm) (PP) (d) Transparent film (PE) (4000 – 5000 µm) (e) Transparent fibre (PVC) (2000 – 3000 µm (f) Two black fibres (3000 – 4000 µm) (both PE).

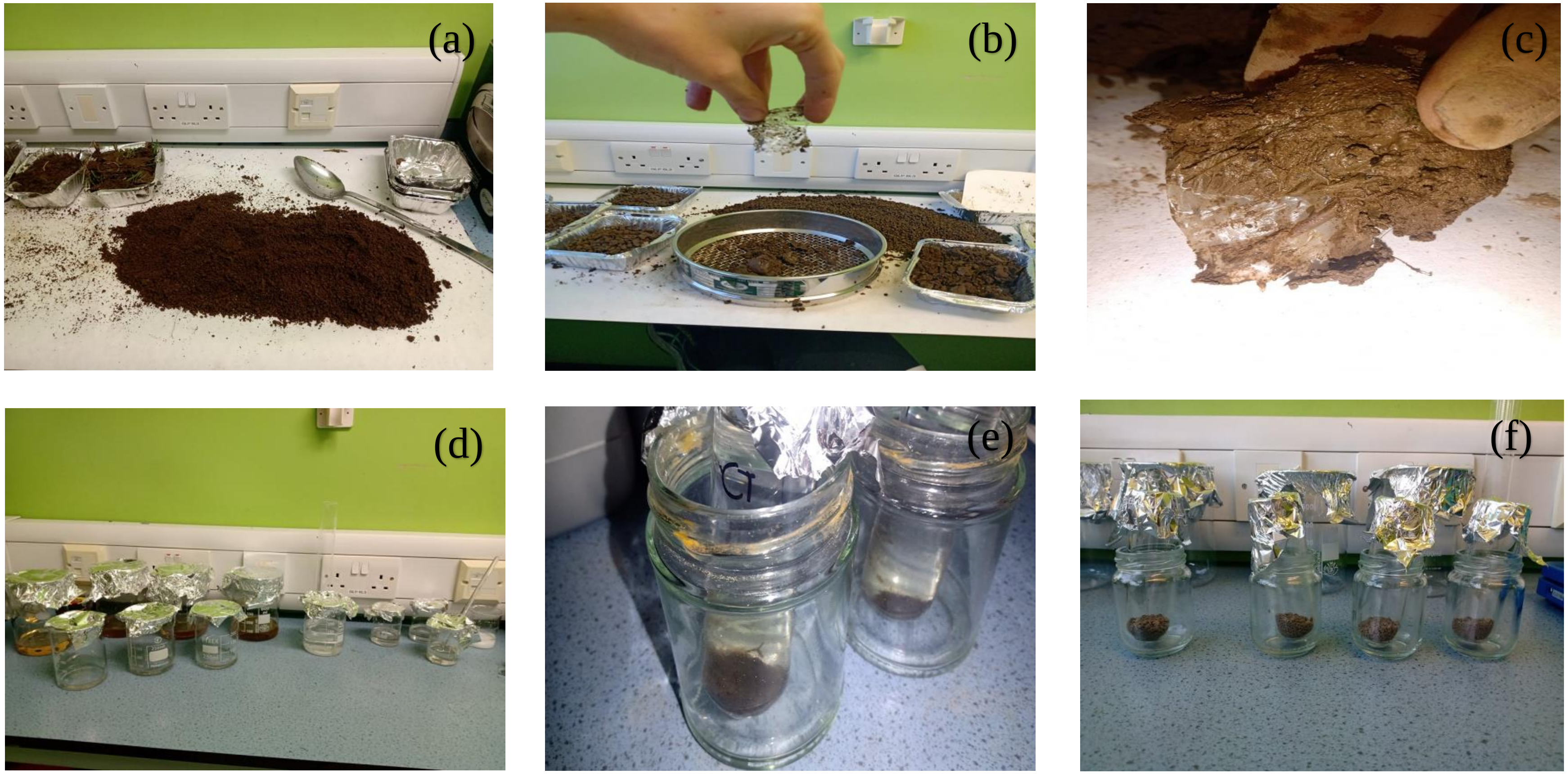


Appendix 3: Figure 2. Meso and macroplastic films found in agricultural soils sampled. (a) Transparent film (PE) (11 cm) (b) Black film (PE) (3.2 mm) (c) Black film (PE) (2.8 cm) (d) Transparent film (PE) (7.2 cm).

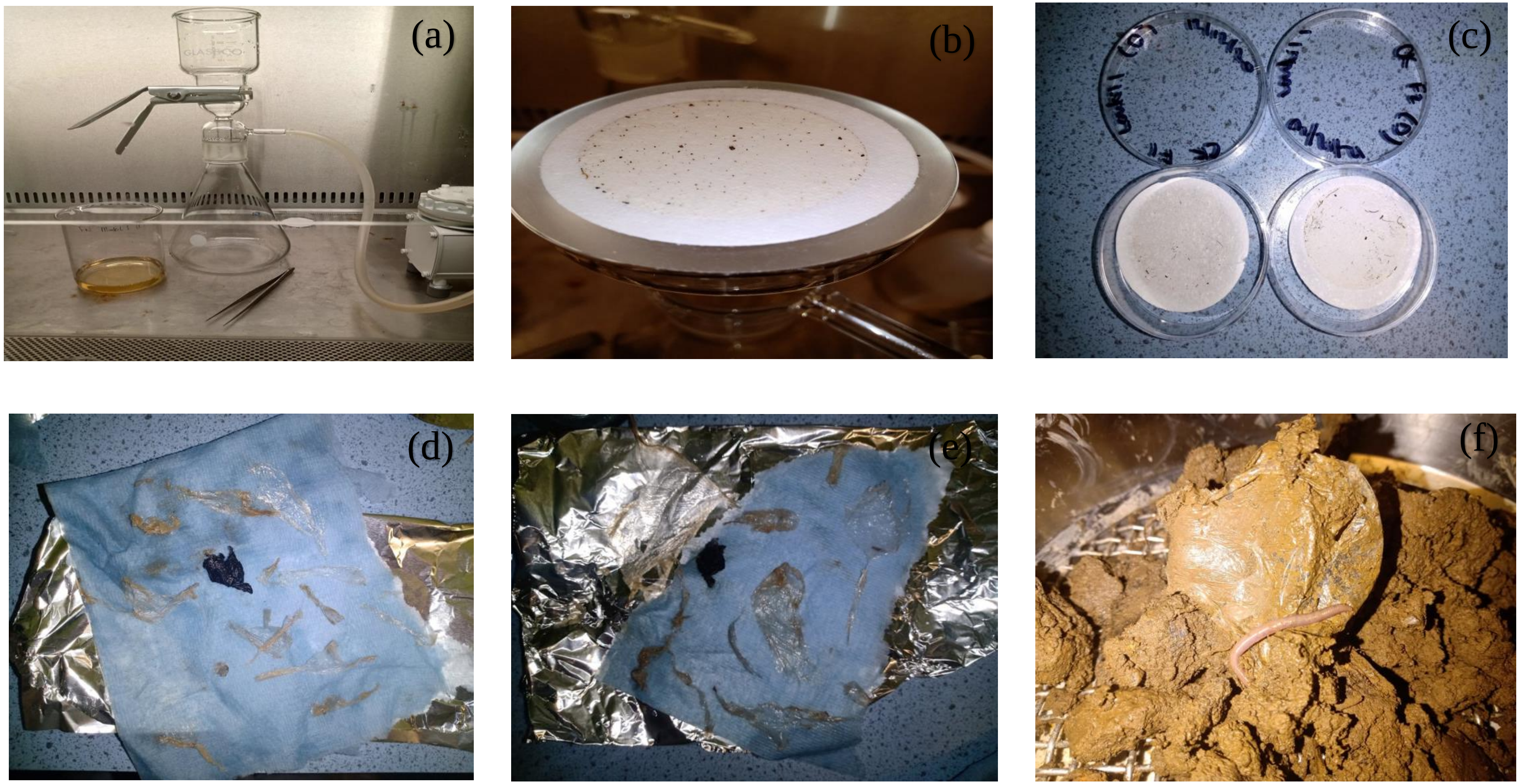
Appendix 4. Field and laboratory photos from chapter 4.



Appendix 4: Figure 1. (a) Agricultural plastic waste, tyres and silage wrap, (b) agricultural plastic waste in farm shed, (c) plastic mulch film remains on field, (d) agricultural plastic waste in field, (e) illegal plastic dumping on side of field, (f) plastic mulch remains on field.

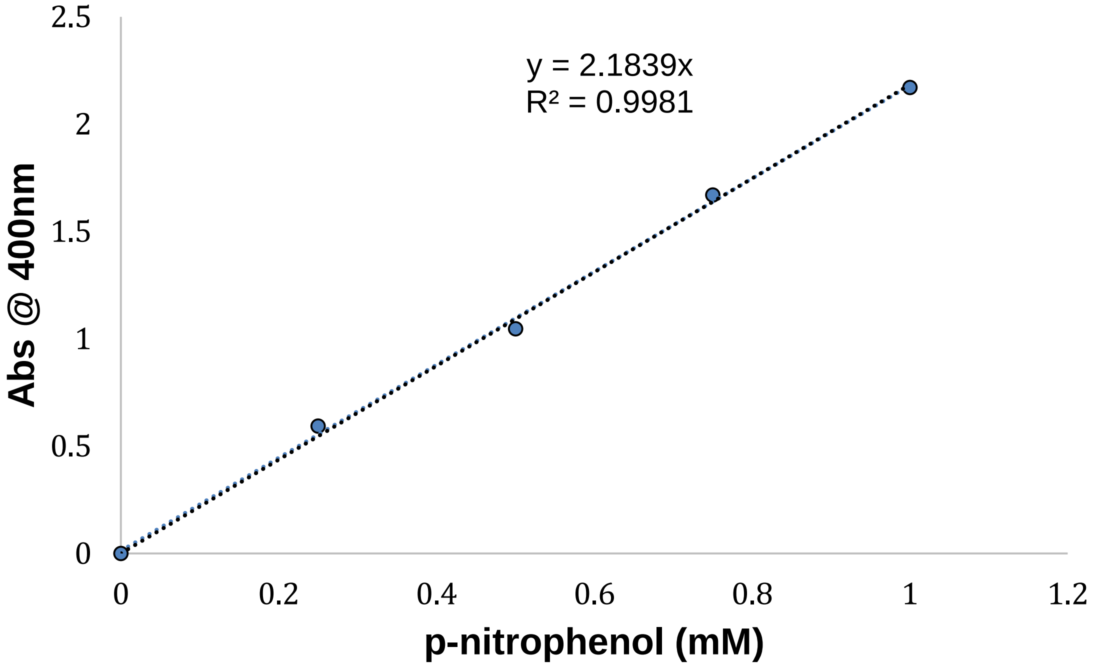


Appendix 4: Figure 2. (a) 5 mm sieved soil, (b) plastic film recovered while sieving soil, (c) plastic mulch macrofilm found in soil, (d) soil sample aliquots pre-density separation, (e) saturated Zn_2Cl_2 solutions with supernatents following density separation (f) soil sample aliquots during density separation.



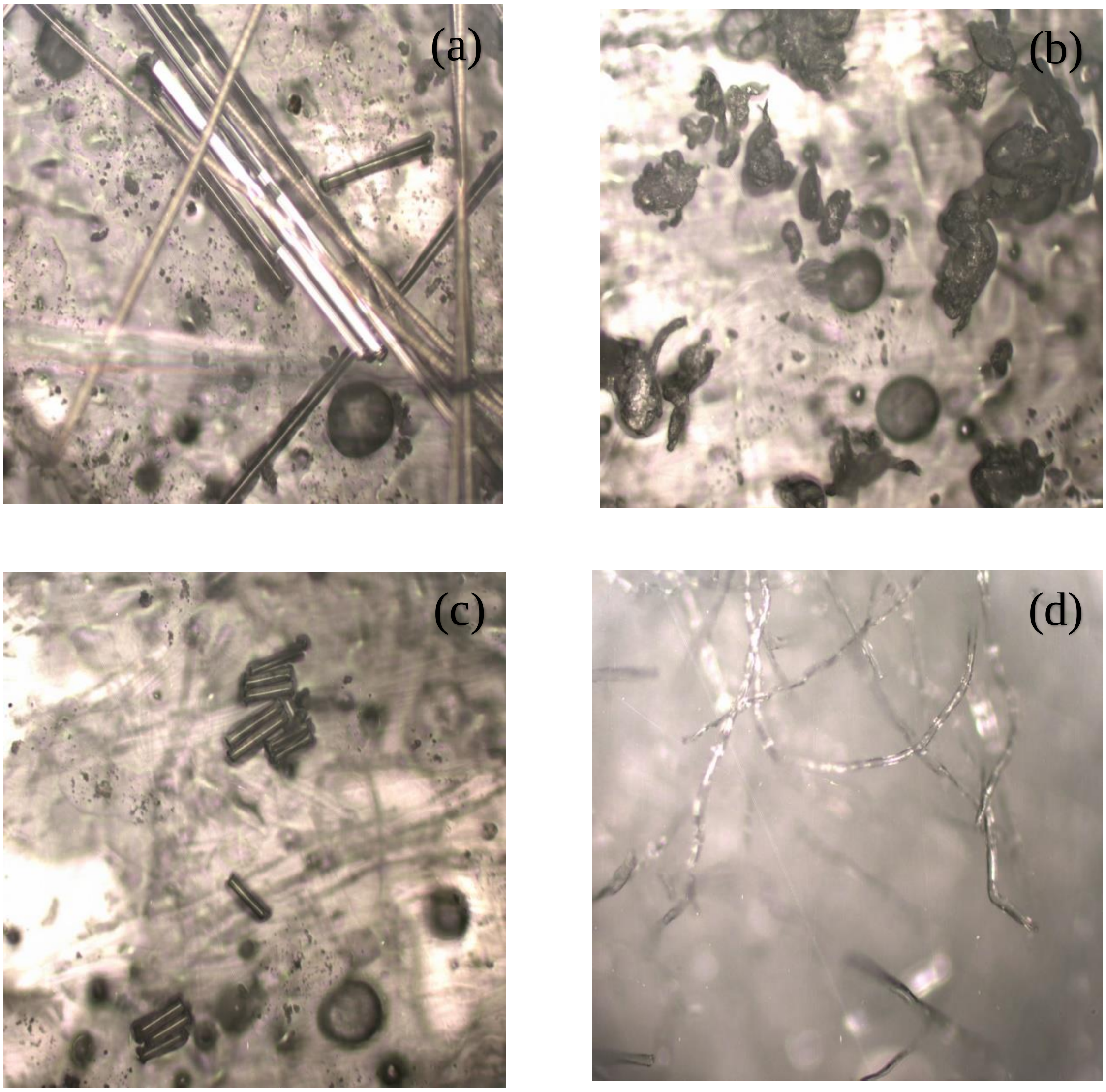
Appendix 4: Figure 2. (a) Vacuum filtration unit (b) filter paper on vacuum filtration unit, (c) two filter papers post-vacuum filtration, (d) meso- and macroplastic films recovered from mulch field, (e) meso- and macroplastic films recovered from mulch field, (f) earthworm recovered from plastic film

Appendix 5. p-Nitrophenol standard curve



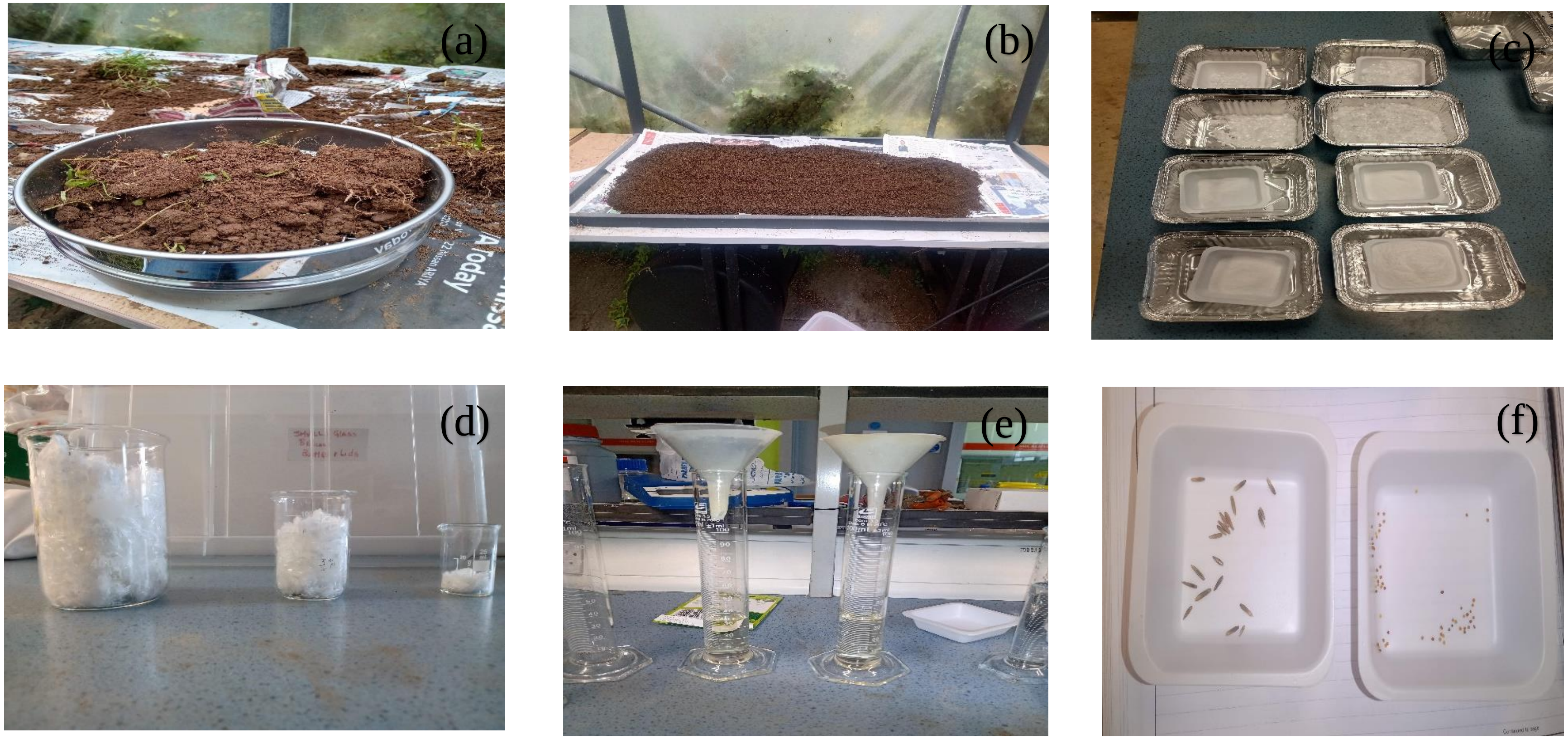
Appendix 5: Figure 1. Standard curve utilised for soil enzyme activity assays.

Appendix 6. Microplastics used in pot trial experiments



Appendix 6: Figure 1. (a) PP fibres (3000 µm), (b) PE fibres (400 µm), (c) PES fibres (250 µm), (d) PES fibres (3000 µm)

Appendix 7. Pot trial experiment photos



Appendix 7: Figure 1. (a) Preparing soil pre-pot trial, (b) air-drying sieved soil pre-pot trial (c) microplastic samples for spiking, (d) quantity of PP (3000 µm) microplastics used for 18 pots (3 different concentrations), (e) soil water holding capacity tests (f) *L. perenne* and *T. repens* seeds.

Appendix 7: Figure 2: (a) PE microfibres added to bulk soil sample pre-mixing (b) PE microfibres in bulk soil sample post-mixing

Appendix 7: Figure 3. (a) and (b) setting pots up with random allocation

Appendix 7: Figure 4. Early plant growth in the first 2-3 weeks of the trial

Appendix 7: Figure 5. Plant growth at Harvest 1

Appendix 7: Figure 6. Plant growth at Harvest 2

Appendix 8. Recovery test and airborne contamination data for chapter 6.

Appendix 8: Table 1. Recovery tests data for the validation of microplastic extraction methods on water and sediments.

Sample	**PP**	**PE**	**PC**	**PES**
Spike-1 (water)				
Rep 1	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Spike-2 (water)				
Rep 1	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Spike-3 (water)				
Rep 1	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Spike-4 (sediment)				
Rep 1	5 (90 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Spike-5 (sediment)				
Rep 1	6 (100 %)	6 (100 %)	6 (100 %)	6 (100 %)
Rep 2	6 (100 %)	5 (90 %)	6 (100 %)	6 (100 %)

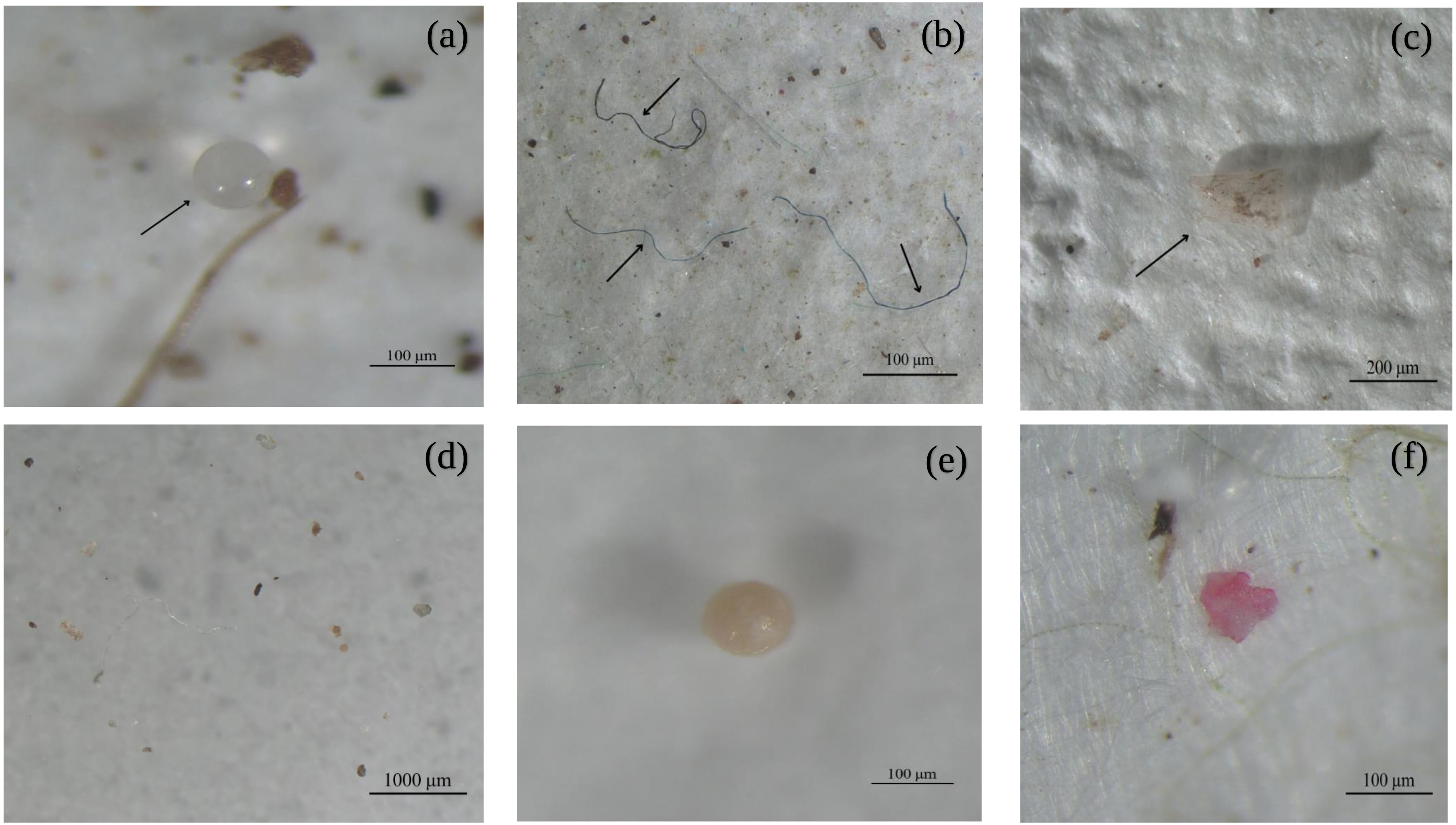
Appendix 8: Table 2. Microplastic airborne contamination (counts and description) during laboratory analysis over 40 days in three areas of the lab.

Filter	Location	Microplastic count	MP description or other
1	Van	0	
2	Van	0	
3	Main prep area	0	
4	Main prep area	0	
5	Fume hood	0	
6	Fume hood	0	Transparent fibre (2340 µm)
7	Main prep area	0	
8	Main prep area	0	
9	Main prep area	0	
10	Main prep area	0	
11	Fume hood	0	
12	Fume hood	0	
13	Fume hood	0	
14	Van	0	
15	Main prep area	0	
16	Main prep area	0	
17	Fume hood	0	
18	Main prep area	0	
19	Main prep area	0	
20	Main prep area	0	
21	Fume hood	0	
22	Fume hood	0	
23	Main prep area	0	
24	Fume hood	0	
25	Van	1	Transparent fibre (3347 µm)
26	Van	0	

27	Main prep area	0	
28	Main prep area	0	
29	Main prep area	0	
30	Fume hood	0	
31	Main prep area	0	
32	Fume hood	0	
33	Microscopy area	0	
34	Microscopy area	1	Black fibre (2039 μm)
35	Microscopy area		
36	Microscopy area	0	
37	Microscopy area	0	
38	Microscopy area	0	
39	Microscopy area	0	
40	Microscopy area	0	

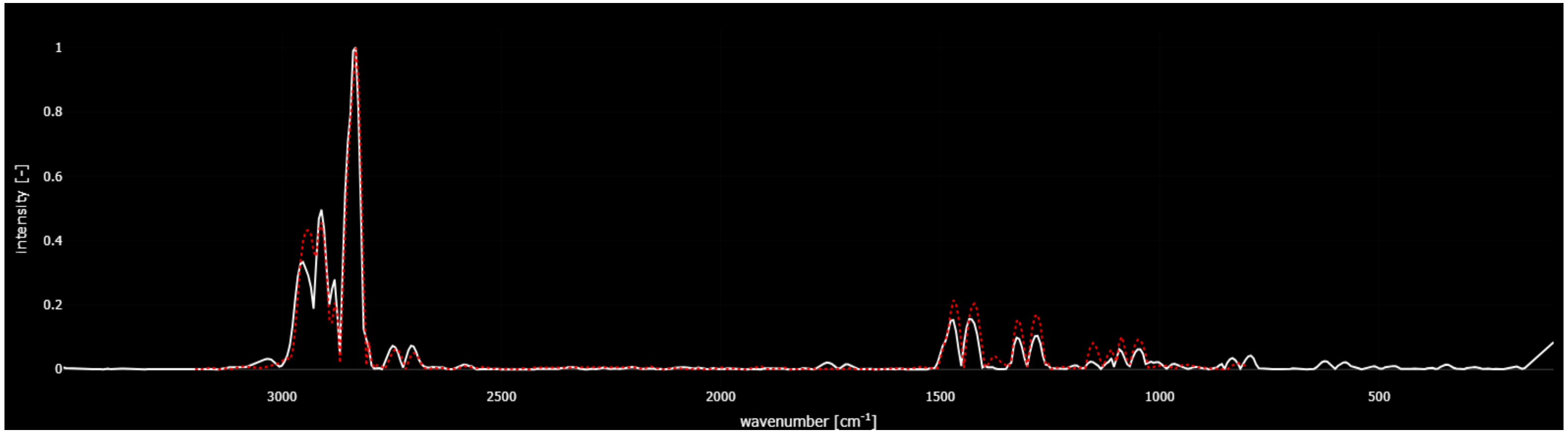
Appendix 9. Sample field and laboratory photos from chapter 6

Appendix 9: Figure 1. From top left (a) raw wastewater influent sampling location with visible wipes and sanitary products, (b) sludge pond effluent sampling location, (c) treated water effluent sampling location, (d) raw wastewater influent sample, (d) residual particles from washed-down sieves used for treated water sampling, (e) sample filter papers containing microplastics pre- and post-digestion.

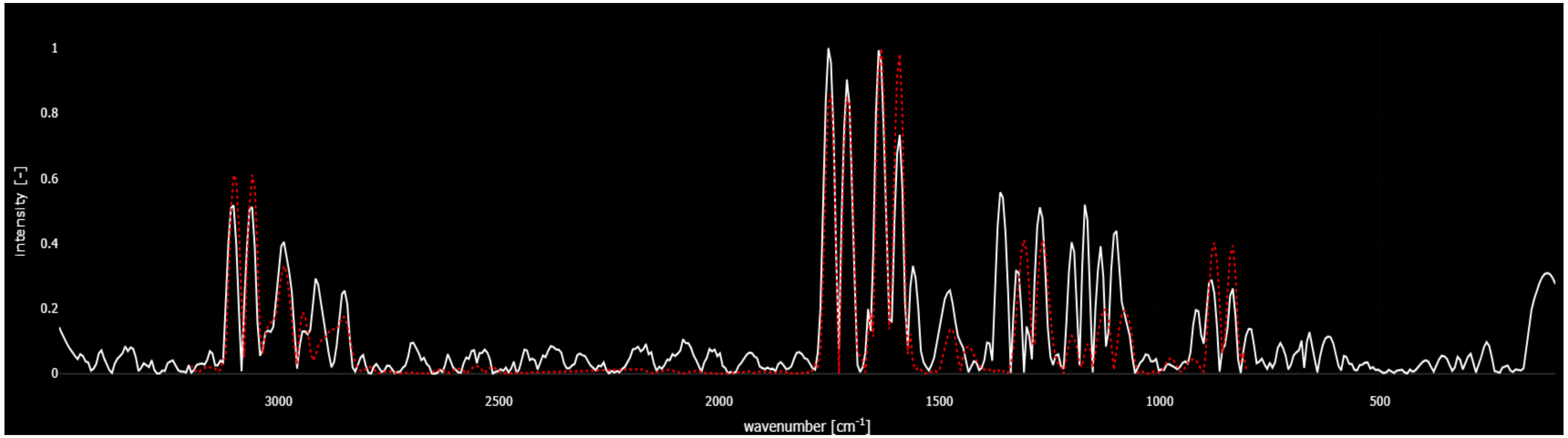


Appendix 9: Figure 2. From top left (a) Microplastics found in water and sediments from the ICW. (a) Microplastic bead (PP) (< 100 µm), (b) Three microplastic fibres (3000 – 5000 µm), (c) Microplastic film (PE) (100 – 200 µm), (d) Multiple microplastic fragments (50 – 200 µm), (e) Microplastic bead (PA) (< 100 µm) and (f) Microplastic fragment (100 – 200 µm)

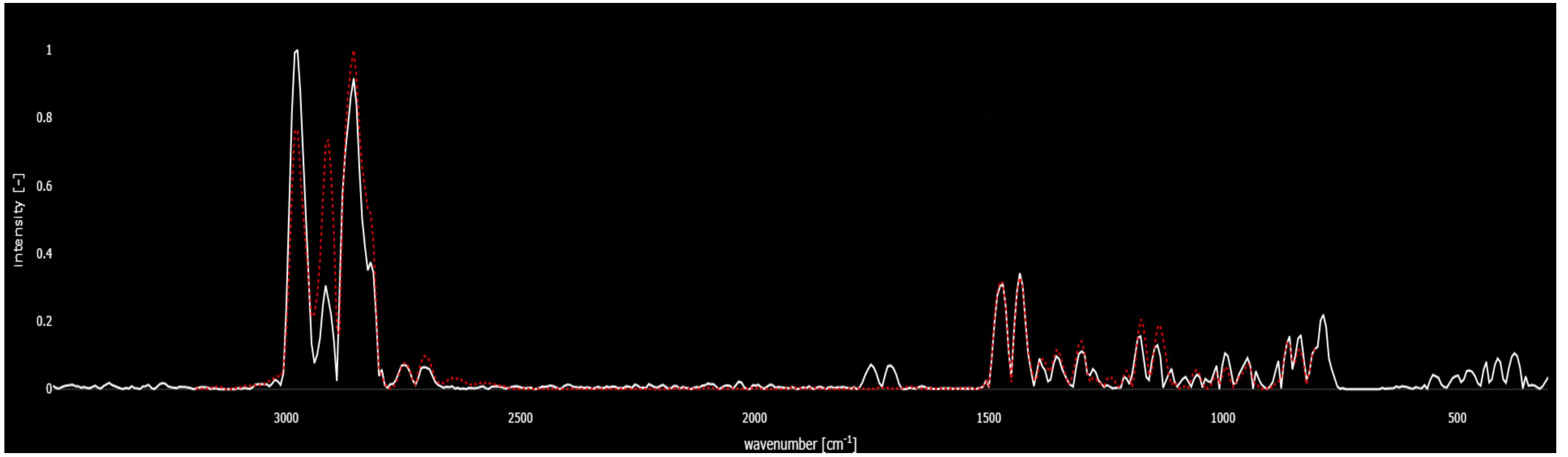
Appendix 10. Raman Spectra



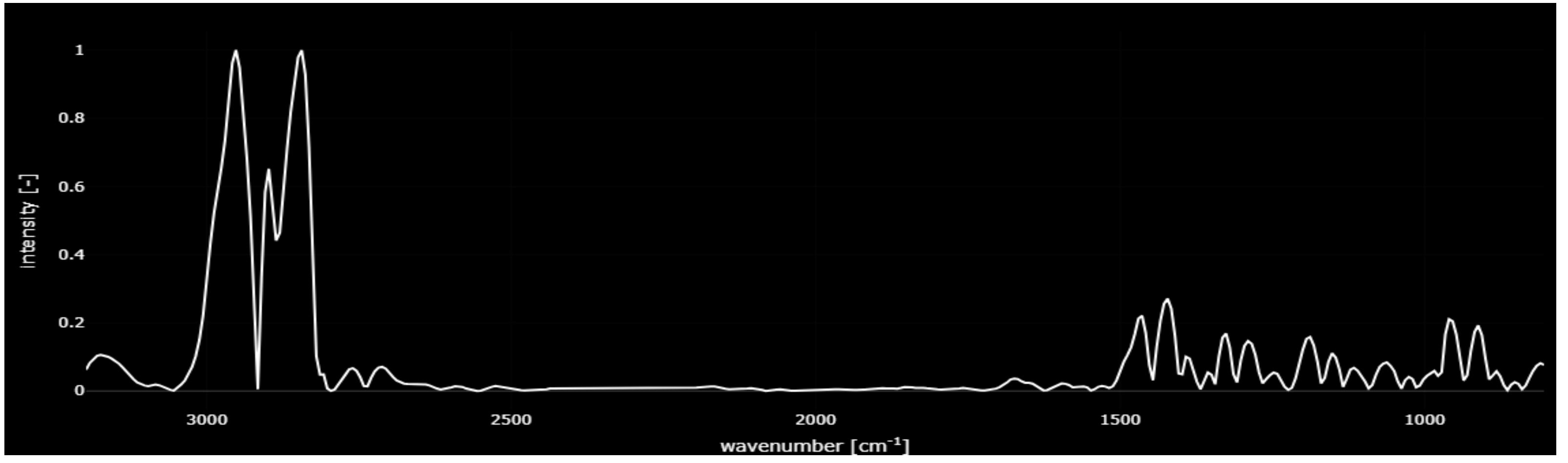
Appendix 10: Figure 1. Raman Spectrum of PE



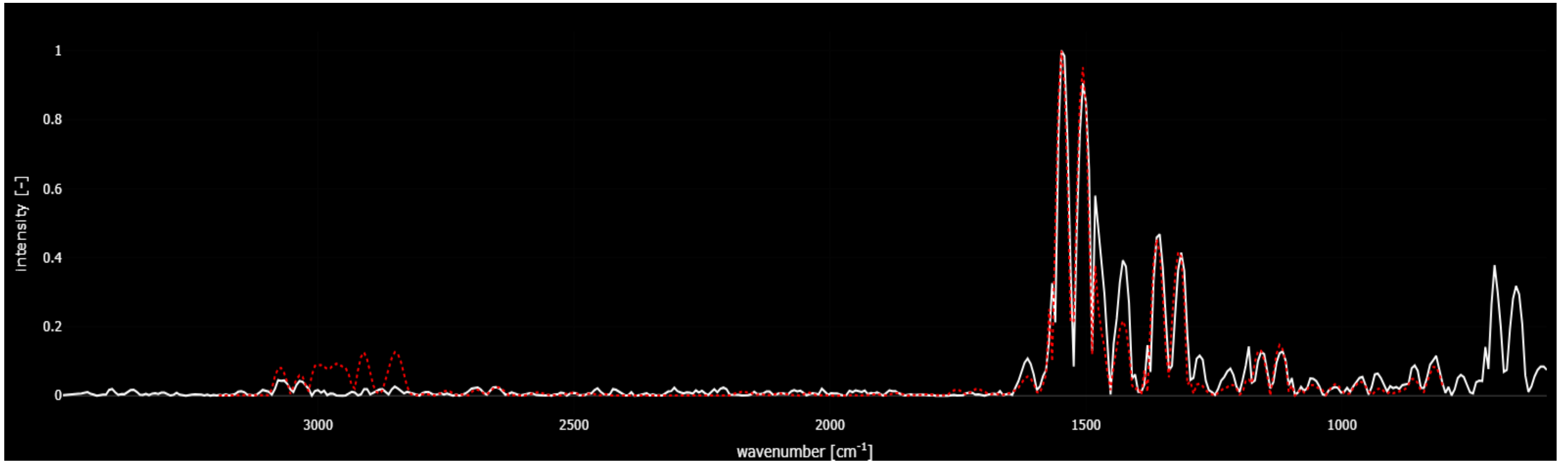
Appendix 10: Figure 2. Raman Spectrum of PES



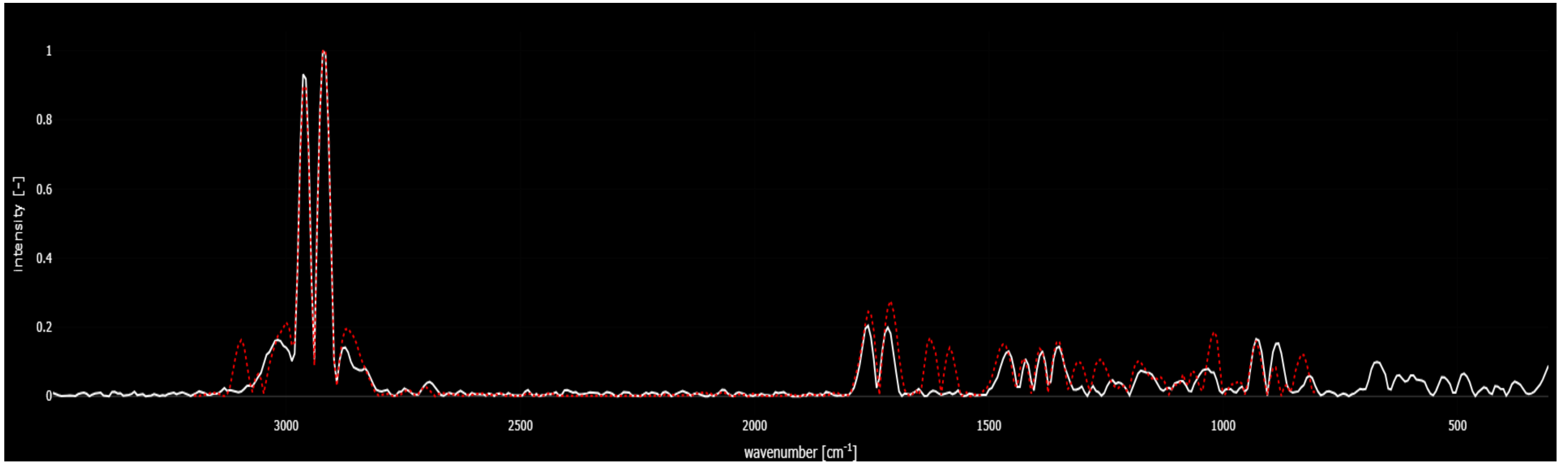
Appendix 10: Figure 3. Raman Spectrum of PP



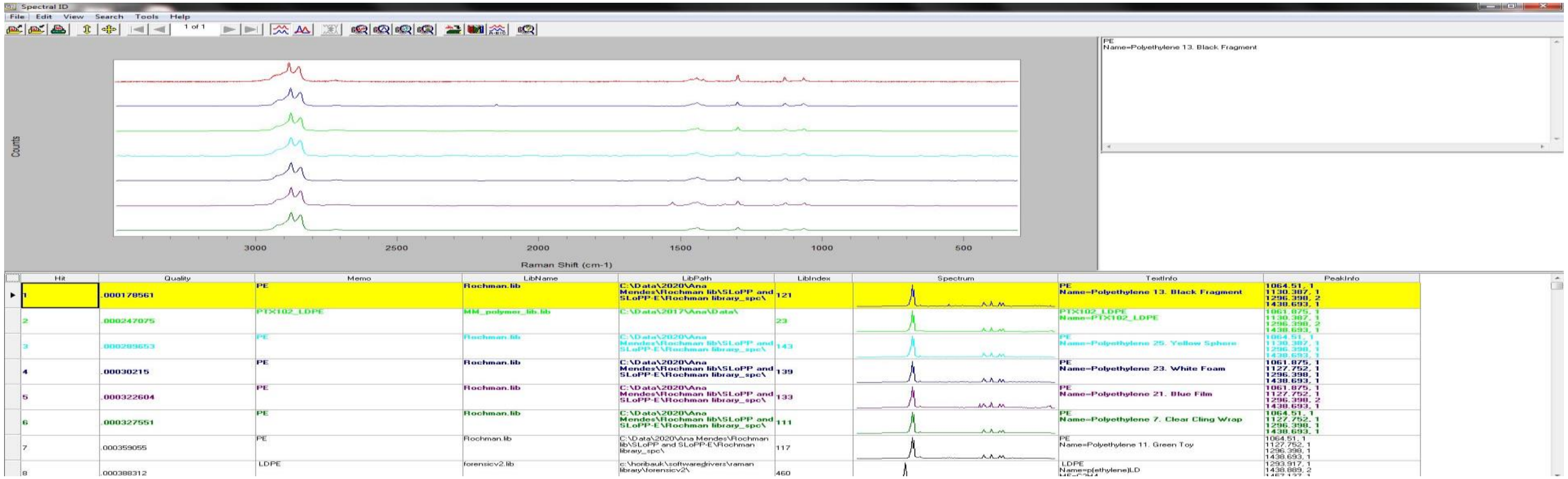
Appendix 10: Figure 4. Raman Spectrum of PET



Appendix 10: Figure 5. Raman Spectrum of PVC

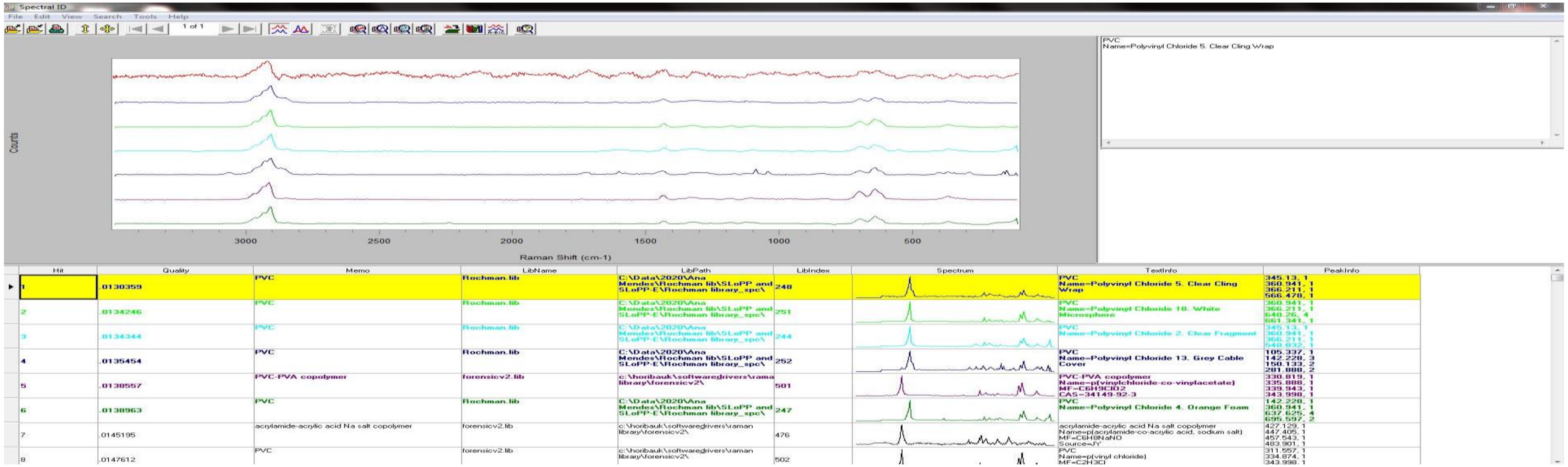


Appendix 10: Figure 6. Raman Spectrum of PVA



Appendix 10: Figure 7. Image of Spectral ID matches of PE using the in-house library extension.

Appendix 10: Figure 8. Image of Spectral ID matches of PE using the in-house library extension.



Appendix 10: Figure 9. Image of Spectral ID matches of PVC using the in-house library extension.

Appendix 10: Figure 10. Image of Spectral ID matches of PVA using the in-house library extension.

Appendix 11. Ethics approval granted for studies of chapter 3 and 4

An Scoil Sláinte & Eolaíochta
School of Health and Science

24th February 2020

Ms. Clodagh King,
Centre for Freshwater & Environmental Studies,
School of Health and Science,
Dundalk Institute of Technology,
Dundalk,
Co. Louth

Re: ***Agcumulate:*** **A study on the accumulation of microplastics in soils and terrestrial ecosystems.**

Dear Clodagh,

The School Ethics Committee reviewed the ethics application for the above study at its meeting dated the 12th February 2020. I acknowledge the amendments which you sent on the 24th February 2020. This application is now approved.

Wishing you the best of luck in your research.

Yours Sincerely,

Edel Healy

____
Dr.Edel Healy
Chair of School of Health & Science Ethics Committee

cc. Dr. Caroline Gilleran Stephens/Dr. Joseph Lynch/Dr. Siobhan Jordan

An Scoil Sláinte & Eolaíochta
School of Health and Science

18th May 2020

Ms. Clodagh King,
Centre for Freshwater & Environmental Studies,
School of Health and Science,
Dundalk Institute of Technology,
Dundalk,
Co. Louth

Re: A study on the accumulation of microplastics in soils and terrestrial ecosystems

Dear Clodagh,

The School Ethics Committee reviewed the ethics application for the above study at its meeting dated the 30th April 2020.. I acknowledge the amendments which you sent on the 15th May 2020. This application is now approved.

Wishing you the best of luck in your research.

Yours Sincerely,

Edel Healy

____

Dr.Edel Healy
Chair of School of Health & Science Ethics Committee

cc. Dr. Caroline Gilleran Stephens/Dr. Joseph Lynch/Dr. Siobhan Jordan