



Plastics, prawns, and patterns: Microplastic loadings in *Nephrops norvegicus* and surrounding habitat in the North East Atlantic



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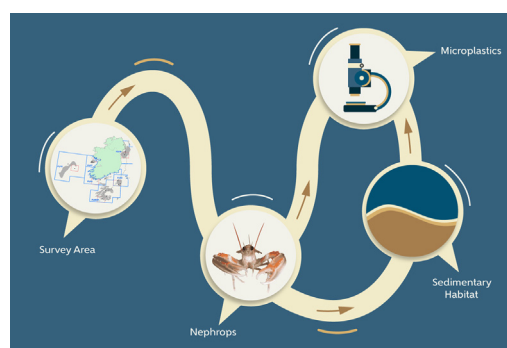
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HIGHLIGHTS

- *Nephrops norvegicus* in combination with habitat as a potential monitoring tool for microplastics
- Low microplastic levels were recorded in *N. norvegicus*, indicating that microplastics do not bioaccumulate.
- Microplastic contamination was assessed in *Nephrops norvegicus* and sediment from six primary fishing grounds.
- Microplastic types and colours from organisms were similar to those retrieved from the surrounding sediment.
- Mean abundance of microplastics recorded in *N. norvegicus* ($n = 600$) was 2.20 ± 2.47 items per individual.

GRAPHICAL ABSTRACT



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ABSTRACT

The presence of microplastics (MPs), a contaminant of emerging concern, has attracted increasing attention in commercially important seafood species such as *Nephrops norvegicus*. This species lend themselves well as bioindicators of environmental contamination owing to their availability, spatial and depth distribution, interactions with seafloor sediment and position in the ecosystem and food chain. This study assesses the abundance of MPs in *N. norvegicus* and in benthic sediments across six functional units in the North East Atlantic. Assessment of the relationship between MP abundance in *N. norvegicus*, their biological parameters and their surrounding environment was examined. Despite the lack of statistical significance, MP abundances, size, shape, and polymer type recorded in *N. norvegicus* mirrored those found in the surrounding environment samples. The three main polymers identified in both organisms and sediment were polystyrene, polyamide (nylons), and polypropylene. The level of MP contamination in *N. norvegicus* could be related to local sources, with relatively low abundances recorded in this study for the North East Atlantic in comparison to other regional studies. Furthermore, larger organisms contained a lower abundance of MPs, demonstrating no accumulation of MPs in *N. norvegicus*. Based on the results of this study, data on MP ingestion could be used to study trends in the amount and composition of litter ingested by marine animals towards fulfilling requirements of descriptor 10 of the Marine Strategy Framework Directive.

1. Introduction

The global production of plastic has increased exponentially since the inception of the plastics industry in the 1950's. Up until 2017, a total of 9.2 billion tonnes had already been produced (Plastic Atlas, 2021), with Europe's production alone reaching almost 55 million metric tonnes in

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2020 (Tiseo, 2022). Plastic is an important material in modern society (Patrício Silva et al., 2020) which has substantially improved our quality of life (Plastics Europe, 2020). The amount of plastic waste produced, which has been rising over time, is expected to more than double by 2050 (Geyer et al., 2017; Lebreton and Andrady, 2019). Furthermore, during the COVID-19 pandemic, the use of single-use plastics increased and therefore these predictions will likely be exacerbated (Benson et al., 2021; Patrício Silva et al., 2021). The release or incorrect disposal of these materials into the environment will likely have negative impacts (Stefatos et al., 1999; Gregory, 2009; Plastics Europe, 2020).

The abundance of plastic pollution has led to a large accumulation of secondary microplastics (MPs) within the marine environment (Isobe et al., 2019), resulting from both degradation and fragmentation of these larger plastics (Kershaw, 2015). MPs are introduced into the marine environment from a variety of different sources and pathways (Rochman et al., 2019). MP sources from land include the agricultural sector (Rehm et al., 2021), tourism (Retama et al., 2016), personal care products (Fendall and Sewell, 2009), domestic waste (Siegfried et al., 2017), and transport (Evangelidou et al., 2020) but can also originate from marine sources such as fisheries (Deshpande et al., 2020) and shipping (Ng and Obbard, 2006). This accumulation of MPs and its expected increase in the marine environment demonstrate a need to monitor the environment to assess the future socio-economic and environmental impacts.

MPs are ubiquitous and have been identified in every ecosystem explored to date, including intertidal and subtidal sediments (Wang et al., 2019; Alvarez-Zeferino et al., 2020), seawater (Frias et al., 2020), the Arctic (Kanhai et al., 2020) and the Antarctic (Jiang et al., 2020) regions. MPs have even been recorded from the top of Mount Everest (Napper et al., 2020) and in the Marianna Trench, a single use plastic bag was identified at a depth of ca. 10,900 m (Chiba et al., 2018).

MPs are considered potentially hazardous due to their physical and chemical composition and persistent nature, having the ability to affect both aquatic habitats and organisms (Rochman et al., 2013; Jambeck et al., 2015). MPs have been ingested by many organisms such as fish (Lusher et al., 2015), seabirds (Acampora et al., 2016), gastropod molluscs (Doyle et al., 2019) and decapod crustaceans (Hara et al., 2020; Cau et al., 2019). Marine biota and human exposure to MPs are considered key research topics in recent years (Hossain et al., 2020).

Bioindicators can be used to assess environmental health (Holt and Miller, 2011). Mussels (*Mytilus* sp.) have been acknowledged as a key bioindicator species under the Mussel Watch Programme (Beyer et al., 2017) and as a potential bioindicator for MP contamination in the environment (Li et al., 2019). MPs are documented in many marine organisms, and more recently there is an increasing number of studies with a sufficient baseline data for suitable representation of MP loadings at the metapopulation/population level, as suggested by Hermsen et al. (2018).

Nephrops norvegicus (Linnaeus 1758) is a decapod crustacean commonly referred to as the Dublin Bay Prawn or the Norway Lobster found living in muddy bottom environments in deep waters (Welden et al., 2015; Cau et al., 2020). In Europe, *N. norvegicus* are considered to be of high economic value, for example within the Irish fishing industry the 2018 landings were estimated to be worth more than €56 million (Marine Institute, 2020a). Despite this, few studies focus on the ingestion of MPs in this commercial species in Ireland (Hara et al., 2020).

N. norvegicus are opportunistic feeders with a diet mainly composed of molluscs, echinoderms, polychaetes and crustaceans (Murray and Cowie, 2011; Welden et al., 2015) with consumption of non-food materials also recorded (Parslow-Williams et al., 2002). They have the capability to ingest solid particles of up to 20 mm in length and 4 mm in width (Yonge, 1924). This non-selective feeding behaviour, and possibly burrowing habits are potential reasons for the presence of MPs in *N. norvegicus* (Murray and Cowie, 2011; Andrades et al., 2019).

There are previous studies that identified MPs in *N. norvegicus*, for example Martinelli et al. (2021) looked at relatively low number of the species, Cau et al. (2020) looked at a localised area, Welden and Cowie (2016a) didn't include a digestive process. This study is novel in that it takes a

more comprehensive methodological approach exploring two environmental matrices covering an extensive geographical area incorporating key *N. norvegicus* Irish fishing grounds in the North East Atlantic. *N. norvegicus* are known to feed close to their burrows, illustrating the potential for MP contamination of wild caught organisms from their surrounding environment (Cau et al., 2019). A study investigating MP ingestion of *N. norvegicus* from three locations in the North Atlantic Ocean (North Sea, North Minch and the Clyde Sea) recorded a large variation in the presence of MPs within the organisms (28.7%–84.1%), suggesting a possible link between the MPs available in surrounding habitat and the amount of MPs ingested by organisms (Welden, 2015). Furthermore, Welden and Cowie (2016b) discovered that ingestion of polypropylene fibres may negatively affect the growth and nutritional state of *N. norvegicus*, with prolonged exposure over time potentially leading to secondary effects such as mortality and decreased fecundity, with contradictory results from Devriese et al. (2017) illustrating that MP ingestion did not affect nutritional state of *N. norvegicus* during 3 weeks of exposure.

The European Food Safety Authority (EFSA) highlighted the need for assessing and monitoring MPs as a seafood contaminant and the potential effects it may have on human health (EFSA, 2016). A 2019 report which assessed European's awareness of food safety topics highlighted that 48% of respondents were aware of MPs in food, illustrating an increasing public concern for plastic contamination and food safety (EFSA, 2019). There is currently no legislation in place regulating MPs as potential contaminants of seafood (Rainieri and Barranco, 2019).

The primary aim of this study was to assess the abundance and characteristics of MPs in *N. norvegicus* and their associated benthic habitat in six functional units in the North East Atlantic. The authors hypothesised that MP abundance in organisms and benthic sediment varies between Functional Units (FU's), with higher MP abundances expected with increasing proximity to shore. In determining a baseline this research further explored if MP abundances were correlated with sex, size, moult stage and presence of the parasitic dinoflagellate *Hematodinium* spp. Furthermore, this study assessed whether *N. norvegicus* would also be suitable as a bioindicator for MPs. The results may inform policy makers and potential future monitoring in respect of the Marine Strategy Framework Directive (EC, 2008).

2. Materials and methods

2.1. Study area

Areas of suitable seafloor around Europe comprising habitat for *N. norvegicus* have been designated into specific fishing grounds, referred to as functional units (FUs) each with a designated number. FU's around Ireland fall within the International Council for the Exploration of the Sea (ICES) Subarea 27.7 (Irish Sea, West of Ireland, Porcupine Bank, Eastern and Western English Channel, Bristol Channel, Celtic Sea North and South, and Southwest of Ireland - East and West) (ICES, 2012). Samples of wild *N. norvegicus* populations were collected from the six primary fishing grounds, namely: (i) Irish Sea West (FU15) (ii) Porcupine bank (FU16) (iii) Aran prawn ground (FU17) (iv) SW and SE coast (FU19) (v) Labadie, Jones, and Cockburn (FU20–21) (vi) Smalls (FU22) (Fig. 1). These areas are defined as primary fishing grounds for *N. norvegicus* by the Irish Marine Institute owing to reviews of fishing activity, with stocks surveyed annually Under-Water Television Surveys (UWTV) (Marine Institute, 2020b).

2.2. *Nephrops norvegicus*

2.2.1. Collection

All *N. norvegicus* samples were provided by the Irish Marine Institute and were obtained from commercial fishing vessels between March and October 2020. Sample collection was carried out within the six pre-established prawn grounds using standard commercial fishing gear, caught in compliance with EU fishing regulations. The individuals collected were

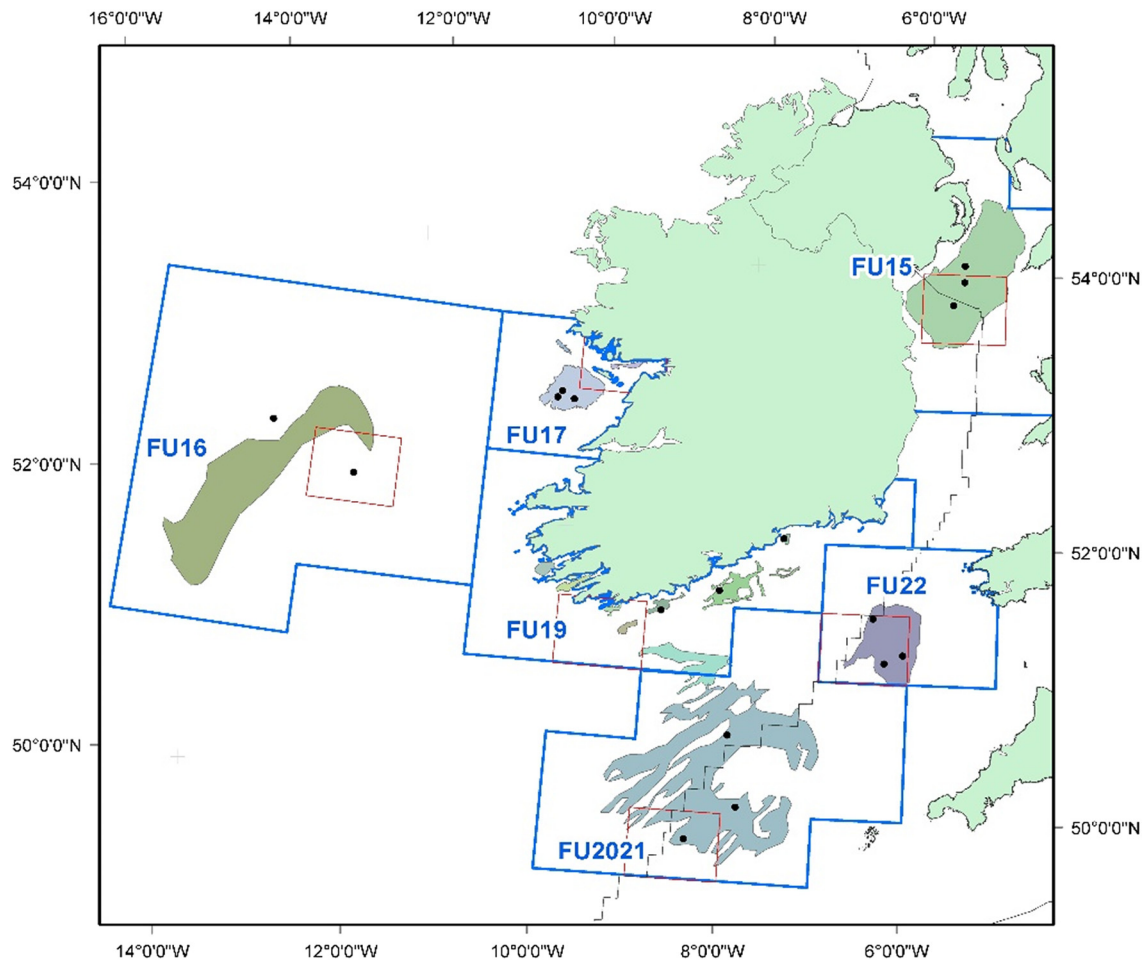


Fig. 1. Designated Functional Unit extensions are delimited in blue: (i) Irish Sea West (FU15) (ii) Porcupine Bank (FU16) (iii) Aran Prawn Ground (FU17) (iv) SW and SE coast (FU19) (v) Labadie, Jones, and Cockburn (FU20–21) (vi) Smalls (FU22). Shaded areas correspond to suitable habitat and UTVW FU survey grounds; ICES Statistical Rectangles outlined in red represent sampling sites for *N. norvegicus* and black dots represent benthic sediment sampling sites.

representative of a commercial catch. *N. norvegicus* samples were subsequently frozen at $-20\text{ }^{\circ}\text{C}$ (Hermsen et al., 2018) until further processing.

2.2.2. Laboratory analysis

Organisms were defrosted at room temperature and the exterior rinsed using ultra-pure water (ELGA PURELAB Option-R 7 BP water purification system, 18 M Ω , 0.2 μm POU filter). The sex, total length (TL), physical damage, carapace hardness, moult stage and presence of the dinoflagellate parasite *Hematodinium* spp. were recorded for each specimen, prior to dissection. TL was measured from the tip of the rostrum to the posterior edge of the telson, sex was determined by the structure of the sexual pleopods (Farmer, 1974) and females were also identifiable by the presence of external eggs (Eiriksson, 2014).

Carapace condition and moult stage of each organism was determined based on the methodology by Milligan et al. (2009). Carapace condition was determined by the hardness of the individual's cephalothorax, divided into three categories, namely, (a) Hard: "if there was no noticeable give in the exoskeleton when squeezed behind the eyes"; (b) Soft: "if the squeezing caused clear distortion"; and (c) Jelly: "when the entire exoskeleton was very soft and gave no resistance to pressure" (Milligan et al., 2009). The moult stage of each organism was based off the same categories, identifying intermoult stage organisms to be hard; late intermoult organisms with removed calcium from the exoskeleton or newly moulted stage but are no longer jelly to be soft, and very recently moulted organisms to be jelly (Milligan et al., 2009; Murray and Cowie, 2011).

The physical damage observed on the external body of the *N. norvegicus* was based on a damage index proposed by Ridgway et al. (2006) which categorises the structural damage caused to the specimen on claws, limbs, eyes, and soft tissue into three categories (a) no damage, (b) lightly damaged, and (c) heavily damaged.

Two methods were used to detect the presence of *Hematodinium* spp. for each individual. Firstly, a colour diagnostic method provides a fast assessment of advanced stages of infection, where parasite infestation can be identified by a vivid dull orange colouration of the carapace (Tärnlund, 2000; Stentiford et al., 2001). Secondly, a pleopod method, requires the removal of a pleopod to be examined under a low light stereomicroscope (Olympus SZX7) at 40 \times for presence of dense aggregations of the parasite, appearing as darkened areas. Accumulation of parasitic material was then classified to stage of infection, 0–4, with 0 being uninfected and stages 1–4 patently infected (Field and Appleton, 1995; Tärnlund, 2000).

2.2.3. Microplastic analysis

Digestive tracts, consisting of the foregut, midgut, and hindgut, once removed, were immediately transferred to decontaminated labelled jars. Digestion of the digestive tract was carried out using a 10% potassium hydroxide (KOH) at 40 $^{\circ}\text{C}$ for 48 h, as recommended by Hara et al. (2020). The resulting digestate was filtered using a vacuum pump (VCP130) through 47 mm Whatman $^{\circledR}$ (GF/C) glass microfiber filter paper (1.2 μm particle retention). The filter was then transferred onto a labelled petri dish for visual examination under a stereomicroscope Olympus SZX7. The particles that were identified as possible MPs were transferred

onto blank sterile petri dishes where photographs and measurements were taken for MP colour, size and for polymer characterisation (Kanhai et al., 2017). The MPs were counted, measured, and photographed using Olympus CellSens® software.

Types of MPs recorded were based on the identification schedule of Frias et al. (2019) and size ranges (1 µm to 5 mm) applied based on the definition of Frias and Nash (2019). A Bruker Hyperion 2000 series FT-IR Microscope with a MCT (mercury cadmium-telluride) detector was used to identify the MP Polymers. Sample spectra were collected in transmission mode in 128 scans (minimum), with a spectral resolution of 4 cm⁻¹, in a wavenumber range of 4000–400 cm⁻¹. In addition, background spectra were measured with the same parameters prior to scanning the MP samples (Kanhai et al., 2017). The JPI Oceans BASEMAN project FTIR polymer library was used for polymer identification.

2.3. Sediment

2.3.1. Collection

Sediment samples were provided by the Irish Marine Institute, which were collected between June 2020 and March 2021 from scientific surveys. All benthic sediment sampling occurred in waters at depths between 38 and 630 m. A Day Grab was deployed to collect benthic sediment samples for MP and granulometric analysis. Sediment samples were taken at each of the six primary fishing grounds around Ireland (Fig. 1). Sediment samples were collected at 3 stations from five of the functional units (FU15, 17, 19, 20–21, and 22), while only two stations were achieved for FU16. Two replicate sub-samples were taken from each grab/sampling station ($n = 34$). Furthermore, a single sample for granulometric analysis was collected from each station. All sediment samples were taken from the top 5 cm were placed into decontaminated glass jars with metal lids. All sediment samples were frozen at $-20\text{ }^{\circ}\text{C}$ until processing.

2.3.2. Laboratory analysis

2.3.2.1. Granulometry. Granulometry was used to determine the sediment composition and methodology was carried out as recommended by Pagter et al. (2018). The sediment samples were defrosted and homogenized prior to being placed in the oven to dry at $105\text{ }^{\circ}\text{C}$ for 24 h. Dried sediment (35 g) was weighed out, transferred into a glass beaker with 6% hydrogen peroxide (H₂O₂) (100 mL) was added and left for 12 h to stand in the fume hood. The surplus H₂O₂ was washed out through a 63 µm sieve, and the sample retained in the sieve was washed back into the beaker where 10 mL of 10% sodium hexametaphosphate (Na₆P₆O₁₈) was added and allowed to stand for a further 12 h. The sediment sample was washed again and left to dry for a further 24 h at $105\text{ }^{\circ}\text{C}$. Once dried, an automated column shaker (Endecotts Octagon Digital Sieve Shaker AAR 3915A) with a range of graduated sieves from 2 mm to 63 µm was used to separate sediment. The weight of sediment retained in each sieve was recorded using a Ohaus Adventurer scale. The silt/clay component was recorded based on comparisons of the initial sediment weight and entered into Gradistat® (version 8.0) software to distinguish the sediment composition.

2.3.2.2. Loss on Ignition. Loss on Ignition (LoI) was carried out to estimate the organic matter content within the sediment and methodology was carried out as recommended by Pagter et al. (2018). The sediment samples were defrosted and homogenized prior to being placed in the oven to dry at $105\text{ }^{\circ}\text{C}$ for 24 h. A subsample of the dried sediment was placed into a pestle and mortar and was crushed into a fine powder. Five grams of fine powdered sediment was baked in a furnace at $450\text{ }^{\circ}\text{C}$. After 6 h, the sample was removed from the oven and left in a desiccator to cool. The subsample was reweighed and the difference between the initial weight was recorded.

2.3.3. Microplastic analysis

Sediment was removed from the freezer and washed using ultra-pure water into aluminium trays and dried in an oven at $40\text{ }^{\circ}\text{C}$ for approximately seven days. The dry sediment was weighed and placed into decontaminated

jars. MPs were extracted from the sediment matrix using a density separation method using Sodium Tungstate Dihydrate (Na₂WO₄·2H₂O) solution (41% w/v; 1.4 g/cm³) as recommended by Pagter et al. (2018). Sodium tungstate solution was added to the sediment (3:1 ratio) (Claessens et al., 2013). The mixture was stirred for 5 min with a stainless-steel stirrer, covered with aluminium foil to prevent contamination, and left to settle for 24 h to allow for the settlement of the silt/clay component. Following the settling period, the supernatant containing floating MPs was pipetted off using a glass pipette and vacuum filtered using a vacuum pump (VCP 130) through a 47 mm Whatman® (GF/C) glass microfiber filter paper. Once the supernatant was filtered, the walls of the filtration device were rinsed using sodium tungstate dihydrate solution to avoid dilution of the solution, and to obtain any particles left on the walls of the funnel. The filter was then transferred onto a labelled petri dish for visual examination and sorting of MPs was performed under a stereo microscope connected to a camera with Olympus CellSens® software. This procedure was repeated three times for each sediment sample.

Classification of MP types, sizes and polymer composition followed that for *N. norvegicus* samples (see Section 2.2).

2.4. Contamination control

Cross-contamination was reduced by using a 100% cotton lab coat and nitrile gloves at all times (Pagter et al., 2018). Wearing of synthetic clothing under lab coat was avoided (Hermesen et al., 2018). Decontamination of glassware was carried out using dilute (10%) Nitric Acid (HNO₃), followed by rinsing three times using ultra-pure water and left to dry upside down to avoid accumulation of airborne particles. All surfaces were cleaned prior to use. Air controls were used every day during all stages of processing. Procedural blanks were carried out on ultra-pure water, sodium tungstate and potassium hydroxide to monitor potential contamination. The contamination quality control for the microplastic analysis in biota carried out in this study was assessed according to the criteria set out by Hermesen et al. (2018) and recorded a good score of 17/20.

2.5. Data analysis

All statistical modelling was performed in Minitab version 18 and RStudio version 4.1.1 software. Descriptive statistics and tests for normality were conducted on all data sets to determine whether parametric or non-parametric statistical analyses were appropriate. MP abundances were analysed using a Kruskal Wallis test for analysis of variance, followed by Dunn's test for multiple comparisons. A correlation analysis (Spearman Rank Correlation) was performed to examine the relationship between the abundance of MP and physical characteristics (body weight, total length, condition, moult stage, and sex of the tested samples). A correlation analysis (Spearman Rank Correlation) was also performed to examine the relationship between MP abundances in *N. norvegicus* and in the sediment within FU's. The significance level for all statistical tests was set at $\alpha = 0.05$.

3. Results

The sex of the *N. norvegicus* ($n = 600$) were identified as 52.3% female and 47.7% male. Out of the 600 individuals measured, 96.3% were assessed to be within the size at onset of sexual maturity (SOM), which is estimated to be 23.2 to 27.6 mm Carapace Length (CL) in females and 25.9 to 31 mm CL in males (McQuaid et al., 2006). Total length (TL) ranged between 61.7 and 145.7 mm, with an average of 95.55 ± 14.01 mm. Almost half of the investigated individuals (47.84%) were observed to have a hard carapace condition, which is assumed to be at the intermoult stage. The organisms with a soft carapace condition represented 27.83% of the sample, which is assumed to be at late intermoult, or recent moult and the jelly organisms represented 24.33% of the sample, which is assumed to be at the very recent moult stage.

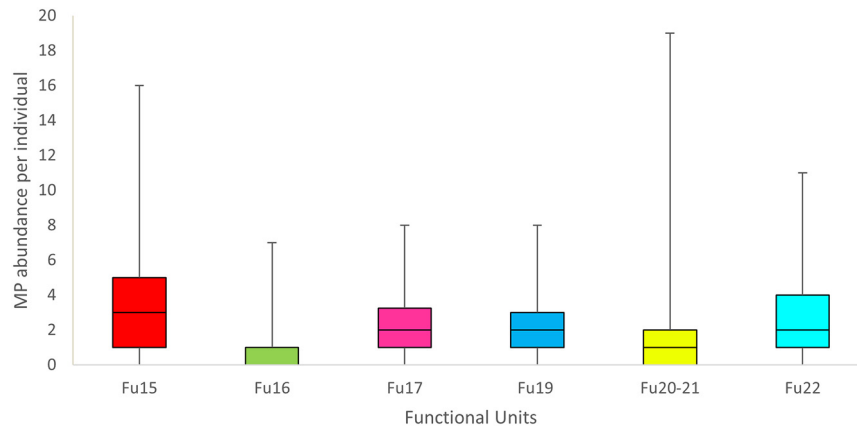


Fig. 2. Boxplot showing the range in the abundance of MPs extracted from the digestive tracts of *N. norvegicus* at each Functional Unit (FU) ($n = 100$; $N = 600$). Boxes represents the first and third quartile, middle bar the median and error bars maximum and minimum values.

3.1. Microplastics and *N. norvegicus*

A total of 1322 particles were extracted from the digestive tracts of 600 *N. norvegicus*, from 6 FU's in the North East Atlantic, with an average of 2.20 ± 2.47 MP items per individual. Of these samples, 430 out of 600 individuals (c. 72%) had ingested at least 1 MP particle.

Samples collected from the Western Irish Sea (FU15) exhibited the highest MP abundance, with an average of 3.66 ± 3.47 items per individual, while the lowest abundance was recorded in the Porcupine Bank (FU16) with 0.80 ± 1.21 items per individual (Fig. 2). The FU's furthest from shore had the smallest abundance of MPs (FU16 and FU20–21), while those in the proximity or within the Western Irish Sea had the highest abundance (FU15 and FU22), however the SE and SW Coasts of Ireland (FU19) which is the closest site to shore recorded a lower abundance of MPs. Procedural blanks and air control contamination recorded while processing was minimal (0.38 ± 0.49 and 0.27 ± 0.45 respectively); therefore, no corrections were made to the analysis.

The percentage of MP occurrence for each sampling station is presented in Table 1, where samples from the Aran Prawn Grounds (FU17) and the Western Irish Sea (FU15) recorded the highest percentage of individuals with MP's (84% and 82% respectively), while the lowest recorded was at the Porcupine Bank (FU16) (42%). The abundance of MPs ranged from 1 to 19 items per individual, with the highest abundance recorded from the Western Irish Sea FU15 ($n = 366$) and the lowest recorded at the Porcupine Bank FU16 ($n = 80$).

FU16 and FU 20–21 were significantly different from all FU's. FU16 had the lowest number of MPs recorded. FU15 was significantly different from all FU's apart from FU22 and FU17. FU15 had the highest MP abundance followed by FU22.

Two main categories of MPs were recorded, with the majority identified as fibres (98.2%) and the remainder fragments (1.8%). Fibres ranged in length from $45 \mu\text{m}$ to 13.34 mm with one outlier measuring 53.88 mm , giving an average length of 1.43 mm . The most common size recorded was $<1 \text{ mm}$ (51%). Results show that 97.4% of all extracted MPs were within

the defined size of MPs ($>1 \mu\text{m}$ and $<5 \text{ mm}$), while the rest consisted of particles $>5 \text{ mm}$ (2.6%) highlighting the presence of macroplastics among extracted particles.

A range of colours of MPs were extracted (Fig. 3), with blue (62.6%) being the most prevalent, followed by black (8.8%), red (8.3%), grey (7.4%), transparent (4.8%), and other (8.2%), which included colours such as green, pink, purple, orange, multicoloured, yellow and white.

A subsample of MPs ($n = 367$, 27.8%) was randomly selected for polymer identification, to include the factors sex, moult stage and length. The most common particles identified in the digestive tract of *N. norvegicus* were Polystyrene (PS), Nylon (polyamide) (PA), Polypropylene (PP) and Polyester. These polymers were recorded from all the FU's and combined they made up 36% of the MP particles analysed (see Fig. 4).

N. norvegicus characteristics such as total length, sex, weight, and moult stage were examined for differences in MP abundance. In an overview of all FU's the smaller individuals ($<82 \text{ mm}$) were recorded to have a higher MP abundance in comparison to larger individuals ($>127 \text{ mm}$) (Fig. 5). While no statistical significance was recorded (Spearman's correlation; $p = 0.297$; $n = 430$, excluding zero values) an inverse relationship was observed. An individual analysis on each FU showed that FU15 and FU16 had statistically significant relationships between TL and MP abundance ($R_s = -0.236$, $p = 0.033$ and $R_s = 0.439$, $p = 0.004$ respectively). The body weight for *N. norvegicus* was examined in one FU (FU16) with a mean of $21.85 \pm 11.1 \text{ g}$, and maximum and minimum values equivalent to 69.99 g and 6.89 g , respectively. A Spearman's correlation analysis between body weight (g) and MP abundance indicated a positive correlation between the variables ($R_s = 0.346$, $p\text{-value} < 0.001$).

Individuals with a hard carapace condition contained a mean of 2.03 ± 1.97 items per individual, soft carapace individuals 2.10 ± 3.01 items per individual and Jelly carapaces 2.24 ± 2.64 items per individual. Correlation analysis indicated no significant association between number of MPs present and carapace condition (Spearman's rank; $p = 0.908$).

Females had a higher MP abundance ($n = 786$) than males (Fig. S1) with a mean of 2.47 ± 1.37 items/individual and males 1.88 ± 0.61

Table 1

Variation in MP occurrence and abundance at each Functional Unit (FU) and the proportion of individuals at each site ($n = 600$) that recorded MPs. A post-hoc Dunn's test indicated significant differences between FU's indicated here as letters (A, B, C, D).

Sampling station (prawn grounds)	Total MP's recorded	Maximum MP count recorded	% Containing MP's	Median MP's recorded
The Western Irish Sea (FU15)	366	16	82	3 ^A
Porcupine Bank (FU16)	80	7	42	0 ^B
Aran Prawn Grounds (FU17)	231	13	84	2 ^{A, C}
SE and SW Coasts of Ireland (FU19)	215	8	77	2 ^C
Labadie Jones and Cockburn (FU20–21)	156	19	64	1 ^D
The Smalls (FU22)	274	11	81	2 ^{A, C}

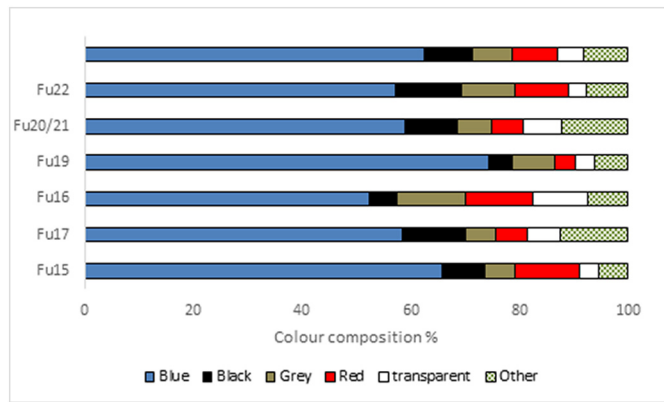


Fig. 3. Colour composition of MPs in *N. norvegicus* across Functional Units ($n = 1322$).

items/individual ($n = 536$). Spearman correlation between sex and MP abundance ($R_s = 0.105$) indicated a trend between the variables. An individual analysis on each FU showed that FU15 and FU22 had statistically significant relationships between sexes and MP abundance at the $\alpha = 0.05$ level ($p < 0.001$ and $p = 0.016$ respectively), indicating that sex has an impact on MP ingestion for these FU's.

Almost half (47.9%) of dissected *N. norvegicus* showed heavy external damage as classified by Ridgway et al. (2006), while 34.8% were lightly damaged and 17.3% had no damage. Following the colour diagnostic method, 9% of examined *N. norvegicus* were infected with *Hematodinium* spp. across the six FU's (Fig. S2), with FU20–21 having the highest rate and FU16 the lowest. Correlation analysis between *Hematodinium* spp. presence and MP abundance showed a positive correlation ($R_s = 0.063$) but was not statistically significant ($p = 0.125$).

The pleopod method of detecting *Hematodinium* spp. infection (Field and Appleton, 1995) showed a larger prevalence of the parasite. Using the index, infections were ranked into stages 0 to 4. Results showed that 54.8% were uninfected (stage 0), 39% were stage 1, 6% stage 2, 0.2% stage 3 and 0% at stage 4 (Fig. S3). As with the colour method, FU 20–21 had the highest level of infection and FU16 the lowest, with correlation analysis to MP abundance, again indicating no significant association ($p = 0.586$).

3.2. Microplastics and sediments

A total of 104 MPs were recorded in 4 kg of dry weight (d.w.) sediment from the six FU's, with a mean abundance of 2.99 ± 1.80 microplastics/100 g dry sediment. The range in the abundance of MPs of sediments at each FU can be seen in Fig. 6.

Two main categories of MPs were recorded with the majority identified as fibres (97.1%) and the remainder fragments (2.9%). The highest average abundance of MPs was recorded at the Western Irish Sea (FU15), with the lowest abundance recorded at SE and SW Coasts of Ireland (FU19). There was a range of MP colours extracted from the sediment ($n = 104$) with blue (75%) being the most prevalent, followed by red (11.5%), white (4.8%) black (3.8%), green (1.9%), and other (pink, grey and multicoloured) taking up the remaining 3%. The length of the MP fibres ranged from 126 μm to 15.269 mm. Four fibres were greater than the upper limit of 5 mm but were included in the analysis as they had a width $< 20 \mu\text{m}$. The most common size of fibres recorded was $< 2 \text{ mm}$ (73%). No significant differences were observed in the level of MPs recorded in the sediments between the FU's (Kruskal-Wallis test; $p = 0.120$).

A subsample (47%) of MPs were analysed for polymer identification from sediment samples across all six of the FU's. Polystyrene (PS), Polypropylene (PP) and Nylon (polyamide) (PA) were the most prevalent polymers identified in the sediment across all six of the FU's, with PS and PP found at all six sites and PA found at three (see Fig. 7).

3.3. Sediment characterisation

Two sediment types were identified based on their particle composition textural group (Folk, 1954): Muddy Sand (FU 19, 20–21, 22) and Sandy Mud (FU15, 16, 17). The average MP abundance in Sandy Mud was greater than Muddy Sand but no significant relationship was found between the sediment type and the abundance of MPs (Kruskal-Wallis test; $p = 0.172$). No significant relationship was found between the level of Total Organic Content (TOC) and the abundance of MPs recorded (Kruskal-Wallis test; $p = 0.416$).

The relationship between MP abundance and depth was examined using a Kruskal-Wallis test, illustrating that there was no significance between MP depth and abundance ($p = 0.453$). Similarly, MP abundance at distance to shore of each FU station was explored and no statistical significance was recorded (Kruskal-Wallis test; $p = 0.479$). In addition, no statistically significant correlation was found between the abundance of MPs

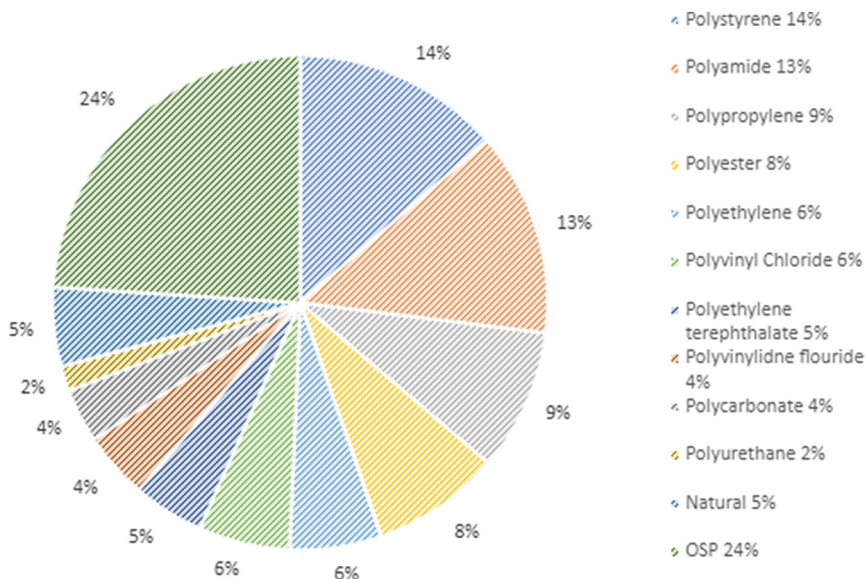


Fig. 4. Polymer composition (%) of MPs ingested by *N. norvegicus* across all 6 Functional Units (FU's). The main plastics are categorised by resin types according to Plastics Europe (2019) in conjunction with Other Synthetic Polymers (OSP) and natural fibres e.g., cotton and linen.

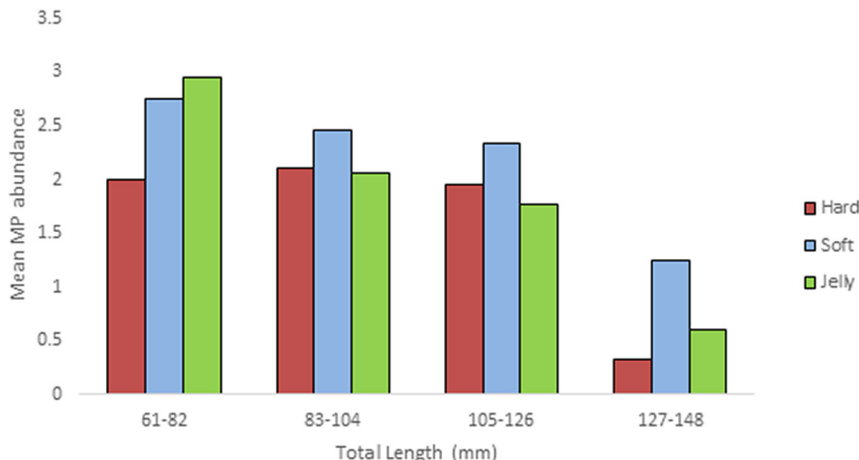


Fig. 5. Mean abundance of MPs in relation to total length and carapace condition.

present in *N. norvegicus* and the abundances found in the sediment (Spearman's rank; $p = 0.623$).

4. Discussion

4.1. Microplastic abundance and polymer types

This study recorded slightly higher MP abundance (72%) than those previously reported in the North East Atlantic (Hara et al., 2020), based on a wild population sample of 150 individuals, where 69% of samples contained MPs. Similarly, Welden and Cowie (2016a) recorded 67% prevalence of MPs in *N. norvegicus* populations from North and West of Scotland. A higher prevalence of MPs (83%) was reported by both Murray and Cowie (2011) in the Clyde Sea examining 120 individuals and by Cau et al. (2019) in the Mediterranean Sea examining 89 individuals. Another smaller study by Cau et al. (2020) recorded 100% MP abundance ($n = 27$) in the Mediterranean Sea and (Martinelli et al., 2021) similarly recorded 100% prevalence in 23 individuals from the Adriatic Sea.

A significant difference in MP presence in *N. norvegicus* between FU's was recorded in this study. The FU's within the Western Irish Sea (FU15) and the Smalls (FU22) showed similar high abundances and were both significantly different from all other FU's. Similarly in a recent study, the Western Irish Sea showed a higher frequency of MPs in comparison to other FU's (Hara et al., 2020). Proximity to shore was not seen to be a significant factor affecting MP abundance in this study with SW and SE coasts (FU19) having lower abundances of MPs although it was closest to shore. However, the proximity to MP sources has been recognised as a potential driver of MP

ingestion by marine organisms (Franceschini et al., 2021) and in this study proximity to highly industrialised coasts of Northern Ireland and Great Britain showed higher levels of MPs recorded in both sediments and *N. norvegicus* from FU15. Similarly, Welden and Cowie (2016a) demonstrated that nearshore habitats near anthropogenic pressures recorded a higher abundance of MPs in the gastrointestinal tract (GIT) of *N. norvegicus*. The Porcupine Bank (FU16) had the lowest abundance of MPs and was significantly different from all other FU's. This may be because FU16 is more isolated (further from shore and deeper) and is likely to be exposed to fewer anthropogenic impacts. Spatial distribution of MPs is strongly controlled by ocean currents (Hill et al., 1997; Ng and Obbard, 2006; Kane Ian et al., 2020) which would suggest that MP concentrations could also be diluted, potentially attributing to the lower MPs levels observed in the SW and SE coasts which are exposed to the Atlantic Ocean (FU19).

The type of MPs recovered from *N. norvegicus* and the surrounding sediment in this study were predominantly blue fibres. This finding is similar to other studies looking at benthic organisms (Welden and Cowie, 2016a; Wang et al., 2019; Hara et al., 2020; Fang et al., 2021) and in sediments in Irish waters (Martin et al., 2017; Pagter et al., 2020b). This highlights the likelihood of fibres as the most abundant type of MP readily available in the marine environment (Wright et al., 2013; Rebelein et al., 2021). The results are similar to other studies which found blue to be the most common MP colour recorded (Hara et al., 2020; Pagter et al., 2020a; D. Zhang et al., 2020). Entangled balls of fibres have been previously recorded in the GIT of *N. norvegicus* (Murray and Cowie, 2011; Welden and Cowie, 2016a), however, they were not a prominent feature in this study.

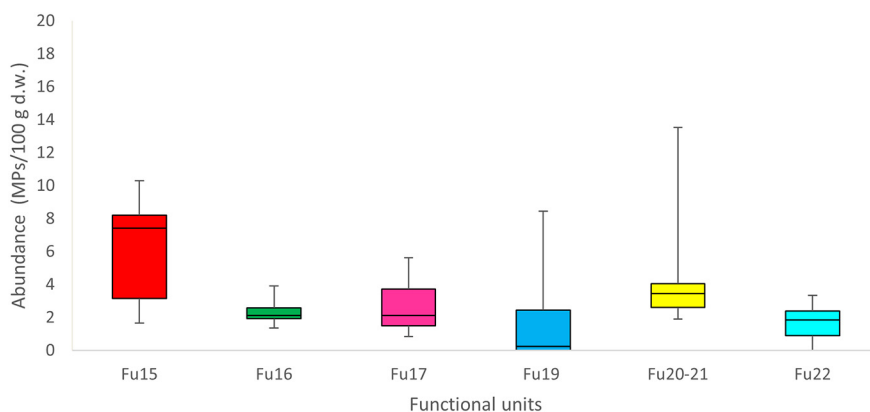


Fig. 6. Boxplot showing the range in the abundance of MPs of sediments at each Functional Unit (FU 15, 16, 17, 19, 20–21 and 22), ($n = 34$). Boxes represent the first and third quartiles, middle bar the median and the error bars, the maximum and minimum values.

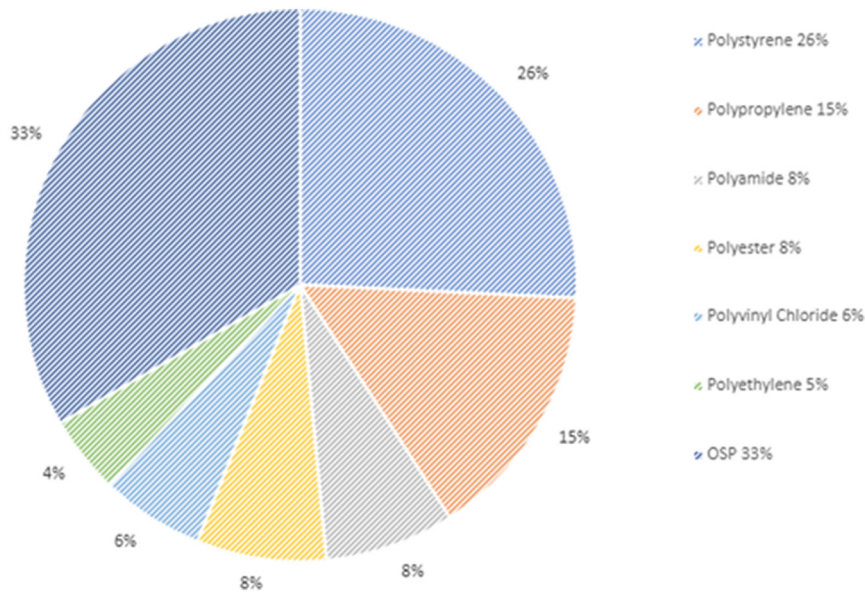


Fig. 7. Polymer composition (%) of MPs found in the sediment across all six Functional Units. The main plastics are categorised by resin types according to [Plastics Europe \(2019\)](#) in conjunction with Other Synthetic Polymers (OSP).

The importance of using a secondary form of MP identification such as a FT-IR Microscope was highlighted by [Pagter et al. \(2020a\)](#) as approximately 20% of the MPs retrieved were identified as natural. The current study identified 5% of the MP subsamples from *N. norvegicus* as natural fibres. The most common polymer of plastic found to be present in *N. norvegicus* was Polystyrene (PS), which is used for food packaging, electrical and electronic equipment, building insulation, inner liner for fridges etc. ([Plastics Europe, 2019](#)); this correlates with findings from [Hara et al. \(2020\)](#). The next two most common polymers were Polypropylene (PP) which is used for food packaging, sweet wrappers, microwave containers, pipes, bank notes, etc. ([Plastics Europe, 2019](#)) and Nylon which is used for fishing nets and ropes ([OSPAR COMMISSION, 2020](#)). It is notable that all three polymers are used in the fishing and aquaculture industry ([EUNOMIA, 2018](#)). Both PP and PA have previously been identified as the most frequently observed polymers in the GIT of *N. norvegicus* in the North and West of Scotland ([Welden and Cowie, 2016a](#)). Plastics with a higher density than water are expected to have increased settling rates in comparison to lower density plastics ([Schwarz et al., 2019](#)). In this study both higher density MPs (PS and PA) and lower density MPs (PP), were retrieved from benthic sediment. This may be due to microbial growth on pieces of plastic (biofilm) which can alter their density causing them to sink ([Semcesen and Wells, 2021](#)) and/or fragmentation of fishing gear already present in the environment which is known to shed MPs ([Saturno et al., 2020](#); [Napper et al., 2022](#)).

It has been acknowledged that contaminant levels in organisms may be closely related to the levels found in the surrounding environment ([Qu et al., 2018](#)). To date, there are a lack of integrated studies investigating MPs in marine organisms and their surrounding environment; thus the relationship between them still remains unclear ([Qu et al., 2018](#)). In a study carried out in the coastal waters of China, a positive relationship between MP levels in two species of mussels and in the surrounding waters was established for not only the abundance of MPs but also for MP characteristics ([Qu et al., 2018](#)). In this study, the most common polymers found in the sediment were Polystyrene (PS), Nylon (PA) and Polypropylene (PP) which mirror the MPs found in the GIT of *N. norvegicus*, highlighting the potential, if not probable link between environmental prevalence and MP abundance in organisms. A study looking at MPs in bivalves had similar findings to this study, where MPs present in the organisms had the same characteristics as those found in the surrounding seawater although no significant relationship was observed ([Cho et al., 2021](#)).

4.2. Microplastic abundance and characteristics of *N. norvegicus*

Both [Murray and Cowie \(2011\)](#) and [Welden and Cowie \(2016a\)](#) suggested that MPs are excreted through ecdysis (moulting process) and assume this to be a key route of excreting MPs aggregations. Furthermore, [Welden and Cowie \(2016a\)](#) recorded lower levels of MPs in the stomachs of individuals that had recently moulted and identified fibres in the discarded gut lining of moulted individuals. The results of this study however, are in line with ([Hara et al., 2020](#)), where no significant association was found between MP abundance and moult stages. It has also been acknowledged that the abundance of plastic cannot be directly linked to a single factor due to many confounding variables ([Lusher et al., 2017](#); [Vendel et al., 2017](#)).

Crustaceans have a complex digestive tract in comparison to other invertebrates ([Welden and Cowie, 2016a](#)) with the presence of chitinous plates in the foregut ([Murray and Cowie, 2011](#)). The shape of the plates narrows at the entrance to the hindgut, which may prevent MPs from being egested. Research has suggested that as the organism grows, the gaps within the gastric mill also increase ([Welden et al., 2015](#)) therefore, allowing for the possibility of larger individuals to egest more MPs in comparison to smaller individuals ([Welden and Cowie, 2016a](#)). The current study shows *N. norvegicus* of less than 82 mm TL are seen to had a higher average MP abundance than larger individuals >127 mm TL, which are in alignment with findings by [Murray and Cowie \(2011\)](#) and [Welden and Cowie \(2016a\)](#) where *N. norvegicus* containing higher abundances of plastics in their stomachs had smaller carapace lengths (CL). However, each FU revealed variation between MP abundance and TL of organisms with contradictory findings further highlighting the ubiquity and the heterogeneity of MPs observed in the marine environment. The smallest organisms were recorded in the Western Irish Sea (FU15; TL of 87.4 ± 13.5 mm) in comparison to the larger *N. norvegicus* found in the Porcupine Bank (FU16; TL of 104.1 ± 16.3 mm). This aligns with current stock assessments where high burrow densities in FU15 are associated with relatively smaller organisms relative to the larger organisms and low burrow densities observed in FU16 ([Johnson et al., 2013](#); [Lundy et al., 2019](#); [Aristegui et al., 2020](#)). Furthermore, smaller organisms were recorded to have higher MP abundance in comparison to larger organism's, contradictory to findings by [Hara et al. \(2020\)](#) which suggested that the highest abundance of MPs were in larger organisms. [Welden and Cowie \(2016a\)](#) stated that the abundance of MP may be reflective of the discrepancy in size and moulting

frequency between males and females. Female *N. norvegicus* moult at a slower rate in comparison to males, and therefore, may have a smaller gastric mill making the egestion of MP particles more difficult and possibly retained for a longer period (Welden and Cowie, 2016a). Females have been thought to retain plastic for twice as long as males due to the reduced moult rate and resulting smaller size (Welden and Cowie, 2016a). Similarly, and in agreement, in this study females had the highest abundance of MPs; however, a significant difference was only illustrated within two FU's (FU15 and FU22). Despite this the authors remain cautious, as the results obtained in this study (variation in MP abundance as a function of sex and TL, and as a result of gastric mill size, and or moulting) all showed low overall MP abundance. Such low values increase potential for false positive/negative results, or indication of no relationship.

The dinoflagellate parasite *Hematodinium* spp. infects commercially valuable crustaceans such as *N. norvegicus* (Li et al., 2021; Stentiford and Shields, 2005), with the parasite having been previously recorded in Irish waters (Briggs and McAliskey, 2002). However, no relationship between the infected organisms and MP abundance was established in this study. The authors are aware of the limitations of the detection methods used, with the colour method having been shown to detect 50% less infections in comparison to the pleopod method (Stentiford et al., 2001). The pleopod method however, can only detect heavily infected individuals (Small et al., 2006) and is open to subjectivity (Stentiford et al., 2001). Therefore, the potential of infection rates is likely to be underestimated in this study.

Feeding behaviour and the prevalence of MPs in the surrounding environment are two of the main factors that can influence MP abundance in organisms (Murphy et al., 2017; Walkinshaw et al., 2020). A direct relationship between MP abundance in *N. norvegicus* and the surrounding environment has not been established to date (Murray and Cowie, 2011; Martinelli et al., 2021). However, diets of *N. norvegicus* have been found to mirror local food availability (Parslow-Williams et al., 2002), with a recent study suggesting a possible relationship between proximity to macroplastic hotspots and MPs in benthic organisms using a generalised additive model (GAM) (Franceschini et al., 2021). The results of the current study indicate that MPs do not accumulate, as larger organisms, who are older, had lower MP abundances. A recent study where separate stomach and intestines examinations of *N. norvegicus* were conducted revealed higher abundances of MPs in the intestine, suggesting their ability to move through the GIT, eventually being excreted (Cau et al., 2020). This is in alignment with the results of our current study.

The prevalence of similar types, colours, abundances, and proportional compositions of MPs in the GITs of *N. norvegicus* and in their habitat sediments empirically indicates MP deposition to the seafloor, through either direct sinking from the water column to the sea floor or through intermediary consumers, acts as a pathway to ingestion by *N. norvegicus*. The vertical transportation of MPs to the deep-sea is complex and poorly understood (Courtene-Jones et al., 2017; Barrett et al., 2020) with the sinking rates of plastic particles influenced by many factors including particle size, shape and polymer density (Zhang, 2017; Kooi et al., 2018). While this study may not explicitly substantiate this, it is a hard to conclude otherwise, and is in line with potential pathways suggested by Coyle et al. (2020). It may be hypothesised that the explicit amounts of MPs present in the GIT of *N. norvegicus* relate to complex interactions of individual anatomy, larger North Atlantic and localised oceanographic conditions, and environmental availability including proximity to point and diffuse sources and biotic and abiotic pathways to sediment and the food chain.

4.3. Microplastics, food security and consumer confidence

Seafood is an important part of healthy diets across Europe (FAO, 2020) with the total seafood production in 2018 amounting to 179 million tonnes and this is expected to further rise to 204 million tonnes by 2030 (FAO, 2020). Therefore, assessing potential human consumption of MPs from seafood is imperative, as a potential exposure pathway (Wright and Kelly, 2017; Smith et al., 2018; De-la-Torre, 2020). Furthermore, food security may be negatively impacted by the presence of MPs in seafood, however,

data and evidence pointing to the existence of a relationship between MPs and food security is still lacking (De-la-Torre, 2020; Walkinshaw et al., 2020).

Recent studies have identified MPs in human stools (Harvey and Watts, 2018; Zhang et al., 2021) indicative that humans can pass MPs. Reviews have identified that exposure can occur through skin contact, inhalation or ingestion (De-la-Torre, 2020). Although there have been more studies on ingestion, it has been hypothesised that humans are more exposed to microplastics via inhalation rather than ingestion (Vianello et al., 2019; Q. Zhang et al., 2020). The true effects of exposure are, however, still unknown (Barboza et al., 2018; Vethaak and Legler, 2021). A lower MP risk of human ingestion is evident in organisms where the GIT is discarded prior to human consumption (Murray and Cowie, 2011; Wright and Kelly, 2017). Furthermore, it must be noted that humans are not only exposed to MPs through seafood consumption but also from other sources such as drinking water (Oßmann et al., 2018), tea bags (Hernandez et al., 2019), beer (Liebezeit and Liebezeit, 2014), honey (Liebezeit and Liebezeit, 2013) airborne particles (Bergmann et al., 2019) and dust (Gallagher et al., 2015). MPs have recently been identified in the edible tissue of *N. norvegicus*, highlighting the possibility of translocation of small plastic particles, however, prudence must be taken when interpreting such results (Martinelli et al., 2021). Mechanisms of translocation of small microplastics remain unclear (Wang et al., 2016) and further studies need to be conducted to assess microplastics in commercially relevant species as well as providing consumer confidence. In comparison to other regions in the Mediterranean, MP abundances in *N. norvegicus* in the North East Atlantic are relatively low (Cau et al., 2019; Martinelli et al., 2021), hence, exposure by human consumption is also considered low (Hara et al., 2020). Of note is the prevalence in this study of MP fibres over fragments, while Martinelli et al. (2021) reported a predominance or fragments to fibres at a ratio of 3:1. Further, it may be speculated that the much higher abundances reported by Martinelli et al. (2021) are a result of samples being from the relatively enclosed Adriatic Sea in the Mediterranean basin in comparison samples from the North East Atlantic, Celtic and Irish seas analysed here.

4.4. Monitoring

Given that the marine environment is everchanging, concentrations of MPs are known to fluctuate over time (Hill et al., 1997; Frias et al., 2020), and although the current risk to humans is low, it is still of paramount importance to monitor plastic contamination to ensure MPs levels remain low. *N. norvegicus* have been previously proposed as an indicator species for plastic pollution (Cau et al., 2019; Welden, 2015; Franceschini et al., 2021). Even though this study does not allow for a distinct relationship to be identified it does demonstrate that *N. norvegicus* may be used as a bioindicator for marine MP pollution, as it meets the selection criteria and reflects the concentration of contaminants in its surrounding environment (Markert et al., 2003; Fossi et al., 2018). For example, *N. norvegicus* are benthic opportunistic feeders of high commercial importance and are widely distributed around the North East Atlantic and the Mediterranean (Ungfors et al., 2013; Cau et al., 2019; Hara et al., 2020). Furthermore, in the Mediterranean, *N. norvegicus* has already been suggested as a bioindicator for MP presence on the seafloor for small – scale (FAO Geographical subareas, GSAs) (Fossi et al., 2018).

It is important to note that a single species approach may not give a complete overview of MP prevalence in the environment, as it only represents a snapshot of what that organism has recently consumed, while organisms from different FU's, guilds (groups of species exploiting a comparable series of resources) and trophic levels could present different levels of MPs available in the environment (Pagter et al., 2020a). This suggests that the limitations of a single species indicator need to be acknowledged, and preferable further investigated, if implemented in a monitoring programme or that a more ecosystem-wide approach should be applied, and clearly so where a suitable single species bioindicator is not available (Pagter et al., 2021).

5. Conclusion

This study provides baseline data on the occurrence of MPs in *N. norvegicus* and its surrounding environment in the North East Atlantic Ocean. It is apparent from the results that *N. norvegicus* in the Porcupine Bank FU16 had substantially lower MP abundance in comparison to the other sites and the Western Irish Sea FU15 had the highest abundance. Importantly, results imply that MPs do not bioaccumulate in *N. norvegicus* and the size of organism may have an influence on MP abundance with the larger *N. norvegicus* having lower MPs, however, there is no individual biological parameter that correlates with the MP abundance recorded.

While research suggests an ecosystem-based approach is the most reliable, *N. norvegicus* in combination with the sediment does provide potential as a monitoring tool but limited to the presence or absence of microplastics in an area and could potential be the basis of a new traffic light system to reflect the levels of bioavailable microplastics. Further research is required to develop this system to define categories of MP abundance.

When assessing the bioavailability of MPs in the environment it may be hypothesised that an ecosystem-based approach, reporting MP loadings in a number of pathways should be applied in order to collect environmentally relevant data to fully inform the MSFD (Descriptor 10 - Marine Litter). This study demonstrated MP presence in *N. norvegicus* and associated substrata, and complexities in ascertaining a robust contaminant level relationship. *N. norvegicus* may form a readily available bioindicator for marine MPs, however further investigation into the MP abundance in *N. norvegicus* and its surrounding environment is recommended to better establish MP retention, mechanisms, patterns, and hotspots.

CRedit authorship contribution statement

Haleigh Joyce: Conceptualization, Methodology, Investigation, Formal analysis, Writing - original draft. **Róisín Nash:** Conceptualization, Writing - review & editing, Supervision. **João Frias:** Writing - review & editing, Supervision. **Fiona Kavanagh:** Writing - review & editing, Supervision. **Jonathan White:** Conceptualization, Methodology, Formal analysis, Writing - review & editing. **Rachel Lynch:** Investigation. **Elena Pagter:** Investigation.

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 A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2022.154036>.

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