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Title	Ingestion of anthropogenic debris by migratory barnacle geese Branta leucopsis on a remote north-eastern Atlantic island
Authors	Coughlan, Neil E.;Doyle, Susan;Baker-Arney, Connie;Griffith, Rose M.;Lyne, Linda;Williams, Hannah;Kelly, Thomas C.;McMahon, Barry J.;Dick, Jaimie T. A.;Cunningham, Eoghan M.
Publication date	2020-08-24
Original Citation	Coughlan, N. E., Doyle, S., Baker-Arney, C., Griffith, R. M., Lyne, L., Williams, H., Kelly, T. C., McMahon, B. J., Dick, J. T. A. and Cunningham, E. M. (2020) 'Ingestion of anthropogenic debris by migratory barnacle geese Branta leucopsis on a remote north-eastern Atlantic island', Marine Pollution Bulletin, 160, 111588 (5 pp). doi: 10.1016/j.marpolbul.2020.111588
Type of publication	Article (peer-reviewed)
Link to publisher's version	http://www.sciencedirect.com/science/article/pii/S0025326X20307062 - 10.1016/j.marpolbul.2020.111588
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Download date	2024-03-28 21:11:25
Item downloaded from	https://hdl.handle.net/10468/10606

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Ingestion of anthropogenic debris by migratory barnacle geese *Branta leucopsis* on a remote north-eastern Atlantic island

Neil E. Coughlan[1,2,3,*], Susan Doyle[4], Connie Baker-Arney[1,2], Rose M. Griffith[1,2], Linda Lyne[3], Hannah Williams[1,2], Thomas C. Kelly[3], Barry J. McMahon[4], Jaimie T.A. Dick[1,2] & Eoghan M. Cunningham[1,2,5]

[1] Institute for Global Food Security, School of Biological Sciences, Queen's University Belfast, 19 Chlorine Gardens, Belfast, BT9 5DL, Northern Ireland, UK

[2] Queen's Marine Laboratory, Queen's University Belfast, 12-13 The Strand, Portaferry, BT22 1PF, Northern Ireland, UK

[3] School of Biological, Earth and Environmental Sciences, University College Cork, Distillery Fields, North Mall, Cork, Ireland

[4] School of Agriculture & Food Science, University College Dublin, Belfield, Dublin, Ireland

[5] School of Biological and Environmental Sciences, Liverpool John Moores University, 3 Byrom St, Liverpool L3 3AF, UK

*Corresponding author:
neil.coughlan.zoology@gmail.com

Abstract

Although seabirds are frequently used as sentinel species for anthropogenic pollution, the extent and impacts of synthetic debris ingestion remains poorly studied for many water bird species. Here, we assess ingestion of synthetic particles (≥0.5mm) by barnacle geese, *Branta leucopsis*, wintering on a remote island. Faecal samples were collected over a period of four wintering seasons. In total, 71 individual samples were assessed, with 79% of samples displaying at least one debris particle (maximum lengths 0.5–5mm) from anthropogenic sources. The recovered synthetic debris were identified as micro-fibres (*n*= 166) and micro-fragments (*n*= 165). The number of synthetic particles detected per sample was generally low at 4.7 ±0.9, 43 (mean ±SE, maximum): micro-fibres 2.3 ±0.3, 10; micro-fragments 2.3 ±0.8, 40. Particle numbers detected per gram of faecal sample differed amongst wintering seasons. Our results suggest that non-marine water birds can frequently ingest low quantities of synthetic particles in remote coastal habitats.

Keywords: Anthropocene; micro-fibre; micro-fragment; plastic pollution; waterfowl; wetland bird,

1. Introduction

The ingestion of synthetic debris, i.e. debris composed fully are partially of synthetic polymers, by birds can lead to increased rates of morbidity and mortality through physical damage, reduced digestive capacity and appetite, and blockage of the gastrointestinal tract (Pierce et al. 2004; Lavers et al. 2014). Transmission of toxic chemicals from ingested synthetic debris, especially plastic particles, can also have deleterious effects such as mortality or reduced reproductive output (Tanaka et al. 2013; 2020). Although relatively large items (pieces > 5mm) of synthetic debris are most frequently implicated as environmental pollutants, the ingestion of small synthetic debris (pieces ≤ 5mm) by wildlife is of growing concern (Cole et al. 2011).

To date, although seabirds are frequently used as indicator species for anthropogenic pollution, ingestion of synthetic debris by shorebirds and other water birds remains poorly studied (Lourenço et al. 2017; Reynolds & Ryan 2018; Rossi et al. 2019). Accordingly, the extent to which synthetic debris is ingested by water birds inhabiting a combination of terrestrial, freshwater and marine habitats in coastal areas remains largely unknown (English et al. 2015). Further, the majority of studies that detail the ingestion of synthetic debris by birds have only focused data acquisition through necropsies and examination of regurgitation pellets, i.e., boluses comprised of items that cannot be digested such as shell fragments and stones (Provencher et al. 2017; 2018). Despite this, birds can also egest synthetic debris that are small enough to pass through the entire gastrointestinal tract (Reynolds & Ryan 2018; Provencher et al. 2018). As such, the lack of studies attempting to report ingestion of synthetic debris through assessment of bird faecal samples is a missed opportunity for environmental monitoring (Provencher et al. 2018), especially as many bird species will not be readily available for necropsy in sufficient numbers to provide a meaningful sample size. In addition, as the contents of regurgitated pellets will only correspond to the most recent meal or meals consumed throughout the previous day, detection of synthetic debris in regurgitated pellets does not reliably represent the quantities of debris that are retained for an extended period of time within the gastrointestinal tract (Johnstone et al. 1990; Acampora et al. 2017a). Further, for many species, pellets are often egested relatively quickly, synthetic debris recovered from regurgitated pellets are not necessarily representative of the shape-types or sizes of debris that are entering more delicate sections of the gastrointestinal tract where absorption of contaminants or physical blockages could more readily occur. Synthetic debris recovered from egested faecal samples could be more representative of non-retained debris, i.e. items that pass through the gastrointestinal tract relatively quickly with digestive boluses, and larger debris items that have been broken down to smaller pieces that can pass through the gut and intestines (Provencher et al. 2018; Le Guen et al. 2020).

Barnacle geese, *Branta leucopsis*, inhabit a mixture of marine, freshwater and terrestrial habitats (Black et al. 2014). As obligate herbivores, *B. leucopsis* traditionally graze on the leaves, stems, roots and seeds of *Plantago/Bellis/Festuca* swards, however, they are increasingly observed to forage on semi-improved agricultural grasslands (Cabot & West 1973; Mason et al. 2017). Between October and April, migratory *B. leucopsis* are found wintering in north-western locations of Ireland and Scotland, where flocks typically forage in coastal pastures, salt marshes, river estuaries, tidal mud flats and offshore islands (Doyle et al. 2018; Mitchell & Hall 2018). Like most obligate herbivores, the digestive system of *B. leucopsis* is relatively inefficient with a low assimilation efficiency of ~30%, i.e. ~70% of ingested food items rapidly moves through the gut undigested (Owen 1971; Black et al. 2014). In general, *B. leucopsis* have gut retention times of between 1.9–3.1 hrs (Prop & Vulink 1992). Accordingly, we suspect that *B. leucopsis* could act as a sentinel species for the presence of synthetic debris in coastal environments through non-invasive analysis of egested faecal samples. Therefore, in the present study, we assess *B. leucopsis* faecal samples for the presence of synthetic debris across multiple sampling periods, from 2015–2019. Faecal samples were obtained from a *B. leucopsis* wintering on a remote, offshore north-eastern Atlantic island. As the geese tend to remain on the island for the duration of their wintering season and given their gut retention times, any recovered egested synthetic debris are most likely representative of on-island pollution.

This study focused on the detection of synthetic particles ≥ 0.5 mm in size, as the analysis of smaller synthetic particles is considered problematic given uncertainties around airborne contamination by ultra-small micro-fibres (Torre et al. 2016). Here, we add to the current paucity of studies that have attempted to quantify the extent of synthetic debris in bird faecal samples. In addition, to our knowledge, the ingestion of synthetic debris by *B. leucopsis* has been documented for the first time.

2. Methods

2.1 Sample collection

A total of 71 faecal samples were collected from adult or first-winter *B. leucopsis* wintering on Inishkea Islands, situated in the north-east Atlantic Ocean off the west coast of Ireland (54°07'30.2"N; 10°12'31.9"W; Figure 1). Samples were collected from the south island (1.84 km^2). Faecal samples were collected in March 2015 (n = 17), November 2016 (n = 14), April and November 2017 (n = 13 and 7, respectively), and March 2019 (n = 20). Samples obtained in 2015, 2016 and 2017 were taken directly from individual birds captured during routine population monitoring. In 2019, fresh faecal samples were collected from a monospecific roosting site. These samples were collected at distances of at least one metre apart, to ensure that they were produced

by different individual birds. Samples collected in 2019 were refrigerated (≤ 4 months), while all others were kept frozen (-20 °C) until required. All birds were handled under licence from the National Parks and Wildlife Service of the Government of Ireland (Section 32, Wildlife Acts 1976–2012), with relevant ringing permits from the British Trust of Ornithology.

2.2 Digestion, separation and microscopy

Samples were transferred into individual glass beakers and weighed on an analytical balance (0.01 g). To eliminate labile organic matter (including non-synthetic anthropogenic debris deriving from a natural source, e.g. 100% cotton, linen or wool fibres), samples were digested in solutions of Iron(II) sulfate heptahydrate ($FeH_{14}O_{11}S$) and 30% hydrogen peroxide (H_2O_2) at 60 °C (0.75 g of $FeH_{14}O_{11}S$ per 50 mL H_2O_2), until total digestion had occurred (Masura et al. 2015). An approximate application ratio of 20 mL per 10 g of faecal sample was employed. Following digestion, a density based separation technique was used to isolate synthetic debris from denser undigested mineral components through flotation, using a saturated solution of NaCl, i.e. 360 g L^{-1}. The resulting supernatant was carefully decanted and vacuum filtered onto filter pads (Whatman 41, 47 mm, 20 μm pore). All filter pads were placed in clean glass petri dishes and dried at room temperature. All samples were then examined under a stereomicroscope (Olympus SZX16). All synthetic debris particles were visually identified using the criteria outlined by Zhao et al. (2016). Recovered synthetic debris was classified into shape-type categories (e.g. fibre, fragment), size range and colour tone (Provencher et al. 2017; Bessa et al. 2019).

Glassware rather than plastic apparatus was used throughout, and was acid washed prior to use. Glassware was covered with fresh aluminium foil to minimise potential contamination during the entire extraction procedure. Further, where possible, apparatus were periodically checked by stereomicroscope for the presence of synthetic particles prior to sample processing. In addition, immediately before processing the first sample of each batch, all glassware were double rinsed with distilled water and a procedural control sample was processed using this distilled water. This damp filter pad was then placed in a petri-dish and used as a laboratory contamination control, whereby it was placed directly alongside the benchtop area in use and remained exposed to the laboratory air during sample processing (n = 5). Additionally, 100% cotton lab coats and nitrile gloves were used during the sample analysis to reduce potential human contamination.

2.3 Statistical Analyses

All data were assessed for normality of residual distributions (Shapiro-Wilk test, $P > 0.05$) and homoscedasticity of variances (Fligner-Killeen, $P > 0.05$). As all data were non-normal ($P < 0.05$), separate Kruskal–Wallis test with a Dunn's post hoc were used to examine total count data

in relation to the dependent variable of winter season. Wintering season spans the months of October to April, inclusive, and represents the time period for which the *B. leucopsis* population dwells on Inishkea Islands. Where residuals did not meet homoscedasticity assumptions ($P < 0.05$), a White-adjusted ANOVA with Tukey LSM post hoc tests were employed instead. Similarly, the number of synthetic particles per gram of faecal sample was also assessed in relation to the dependent variable of winter season. The biomass of collected faecal samples was also considered in relation to winter season using Kruskal–Wallis tests. All statistical analyses were performed using R version 3.5.3 (R Core Team 2019).

3. Results

In total, 331 synthetic particles were detected; 166 were micro-fibres and 165 were micro-fragments. However, while 79% of samples were found to contain synthetic particles, the number detected per sample was generally low at 4.7 ± 0.9, ranging from 0–43 particles (mean ± SE): micro-fibres 2.3 ± 0.3 (0–10); micro-fragments 2.3 ± 0.8 (0–40) (Figure 2). When standardised in relation to the quantity of faecal mass per sample, synthetic particles were detected at a mean (±SE) frequency of 0.67 ± 0.12 particles g^{-1}, ranging from 0–4.88 particles g^{-1}. Micro-fibres and micro-fragments were observed at a frequency of 0.52 ± 0.12 particles g^{-1} (0–4.88) and 0.15 ± 0.05 (0–2.10), respectively. Potential laboratory contamination was low, with only three micro-fibres being detected by control filter pads, i.e. 0.04 per processed faecal sample, therefore no adjustments to the results were made. All synthetic particles recovered from faecal samples had a maximum length within the size range of 0.5–5 mm, and therefore, could be categorised as either micro-fibres or micro-fragments (Torre et al. 2016; Zhao et al. 2016). Micro-fibres had dark (92%; e.g. navy-blue, black, dark red) or mid colour tones (8%; e.g. blue, red), while micro-fragments consisted of mid (13%; e.g. blue, blue-green) or light (87%; e.g. clear, white-blue, yellow) colour tones.

The total number of synthetic particles recovered in wintering seasons significantly decreased over the sampling period (ANOVA $F = 3.49$, df = 3, 67, $P < 0.05$), as did the number of detected micro-fragments (ANOVA $F = 3.15$, df = 3, 67, $P < 0.05$: Figure 3). However, the number of detected micro-fibres did not differ (KW; $P > 0.05$). The total number of synthetic particles observed per gram significantly differed amongst wintering seasons, which was driven by a substantial increase of particles in 2019 samples (ANOVA; $F = 10.62$, df = 3, 67, $P < 0.001$: Figure 3). The number of micro-fibres recovered per gram of faecal sample also significantly differed amongst wintering seasons due to greater detection of micro-fibres in 2019 samples (ANOVA; $F = 9.28$, df = 3, 67, $P < 0.001$), whilst micro-fragments detected per gram of faecal sample did not significantly differ amongst wintering seasons (ANOVA; $P > 0.05$: Figure 3).

There was no apparent difference in the biomass of collected faecal samples amongst winter seasons (KW; $P > 0.05$).

4. Discussion

Here, to our knowledge, ingestion of synthetic debris by *B. leucopsis* has been recorded for the first time. Further, it is clear that this phenomenon has been ongoing over a number of years at an isolated and remote location. Although this is the first record of synthetic debris ingestion by Anatidae in Ireland, previous records have documented ingestion of debris by Anatidae dwelling within anthropogenically disturbed sites across continental Europe (Gil-Delgado et al. 2017), North America (English et al. 2015; Holland et al. 2016), and South Africa (Reynolds & Ryan 2018). In addition, a variety of coastal dwelling seabird species have previously been recorded to ingest synthetic debris in coastal locations of Ireland, in-line with international trends for debris ingestion by seabird species (Acampora et al. 2016; 2017a,b). Nevertheless, this is the first study to consider multiyear assessment for a single population of a non-obligate marine bird species residing within a remote area, with 4–8 years of sampling being needed to reveal possible trends (e.g. van Franeker & Meijboom 2002). For example, although the total number of synthetic particles detected decreased between 2015/16–2018/19 wintering seasons, the total number particles recovered per gram of faecal sample has generally remained the same across the wintering seasons, suggesting that synthetic particles are ubiquitous on the Inishkea Islands. Although 2018/19 samples do suggest an increase in the number of particles detected per gram, this appears to be an artefact of the 2019 samples, which were taken from a unique sample location (i.e. a roosting site), as well as this relationship being largely driven by three outlier samples. However, it is worth noting that a truly accurate assessment of any change in debris ingestion frequencies (e.g. ± 5% detection rate with a sampling power of 80%) would likely require annual sampling of >14,000 birds (Lavers & Bond 2016), which is simply not feasible for the vast majority of study systems.

In the present study, a relatively high frequency of synthetic debris ingestion by *B. leucopsis* was recorded, with 79% of samples having at least one synthetic particle. However, for Anatidae, frequency of ingestion appears to vary amongst species and sampling locations, e.g. 4.3–53.8% (English et al. 2015), 0–50% (Holland et al. 2016), 43.8–60% (Gil-Delgado et al. 2017), and 0–17% (Reynolds & Ryan 2018). However, despite outliers, the mean number of debris items recovered per sample tended to be similar, if slightly higher, to amounts reported by other studies (e.g. English et al. 2015; Gil-Delgado et al. 2017). To date, studies assessing waterfowl faecal samples for the presence of synthetic debris, including the present study, have not considered possible in-field contamination. This is particularly problematic for micro-fibres, as wind or soil-

surface borne micro-fibres could theoretically attach to faecal samples prior to collection. Therefore, we argue that future studies should consider back-ground levels of wind and soil-surface borne synthetic debris, as contamination prior to collection could inflate water bird debris consumption data. Nevertheless, in some instances, the ingestion of synthetic debris by water birds has been linked to the availability of historical or current sources of synthetic debris at study sites (e.g. Gil-Delgado et al. 2017; Reynolds & Ryan 2018). Although the Inishkea Islands have become a key wintering site for *B. leucopsis* since the last islanders were evacuated by the Irish Government in 1932 (Cabot 1963), and despite being rarely visited by people, an extensive amount of synthetic debris has been washed in from the sea along low lying shoreline (e.g. domestic waste and fishing gear) (S.D. pers. obs.). In addition, this synthetic debris can be pushed further inland during storm conditions, and is sometimes found in *B. leucopsis* foraging areas. Accordingly, we argue that ingestion of synthetic debris by *B. leucopsis* is most likely linked to the prevalence of pelagic synthetic debris that is deposited by tidal forces.

Interestingly, synthetic debris >5 mm in length was not detected by the present study. However, micro-fibres and micro-fragments can result from the fragmentation of larger particles due to biotic and abiotic effects (e.g. Mateos-Cárdenas et al. 2020). Including within digestive tracts of birds (Provencher et al. 2018), especially granivorous waterfowl with strong gizzards (Mayhew & Houston 1993), that are potentially capable of mechanically disintegrating larger synthetic debris items over time (Reynolds & Ryan 2018). However, species-specific gut retention times of synthetic debris remain poorly understood (but see Charalambidou et al. 2005). In addition, the abundance of debris in bird gastrointestinal tracts is largely determined by their retention period (Holland et al. 2016; Ryan 2016). Further, longer retention times may aid inter-habitat dispersal of synthetic debris by water birds (Coughlan et al. 2017). In particular, as a long-distance migrant, Ireland's wintering *B. leucopsis* population originates exclusively from remote areas of north-east Greenland with staging grounds in Iceland, unlike Scottish populations that arrive from both Greenland and Svalbard (Wernham et al. 2002). As such, despite generally short gut retention times for food items (i.e. 1.9–3.1 hrs; Prop & Vulink 1992), *B. leucopsis* could be a potential vector of synthetic debris and other contaminants amongst remote areas of Greenland, Iceland and Ireland, if ingested synthetic debris remains within the gastrointestinal tract for an extended period of time.

Whilst the present study demonstrates frequent and sustained ingestion of synthetic debris by a migratory water bird species inhabiting marine coastal habitats, more in-depth assessments are required to ascertain the overall impacts of synthetic debris ingestion on water bird health. In particular, greater quantification of the amounts and types of synthetic debris available in coastal environments is required, as well as the rates of ingestion and retention by water bird species

(English et al. 2015; Holland et al. 2016; Reynolds & Ryan 2018). Although visual identification is considered relatively reliable (Zhao et al. 2016; Reynolds & Ryan 2018; Stanton et al. 2019), there remains a risk that some misidentification of natural fibres as synthetic fibres could have occurred in the present study. This is especially relevant given that a growing number of studies have documented a higher prevalence of natural textile based micro-fibres than synthetic micro-fibres in freshwater and marine ecosystems (see Stanton et al. 2019; Suaria et al. 2020). Accordingly, future research should consider the use of analytical chemistry techniques (e.g. Raman and FTIR) to ascertain polymer identification, thereby reducing the potential for misidentification of debris (Zhao et al. 2016; Stanton et al. 2019). Yet, natural micro-fibres such as textiles also represent a harmful environmental contaminant (e.g. Stone et al. 2020). In addition, the extent of absorption and subsequent impacts of chemical contaminants requires greater consideration. Nevertheless, our findings suggest that synthetic debris could be a problematic pollutant for non-marine coastal dwelling water birds, with possible bird-mediated dissemination of synthetic debris to remote arctic regions by from *B. leucopsis* wintering grounds.

5. Acknowledgements

The authors acknowledge support from the G. & M. Williams Research Fund, Queen's Marin Laboratory, and the Irish Research Council. We gratefully thank Ryan Johnston and Craig Holmes for assisting this study, and Patrick Manley and Brian Burke for assistance collecting samples. We also gratefully acknowledge support for Dr Katherine Booth-Jones and the British Trust for Ornithology. In particular, we thank Emma Healey and Allen Whitaker for technical support, and Ricky Whelan, Marcel Jansen and Alicia Mateos-Cárdenas for encouraging discussions.

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406

407 Figure 1: Yearly faecal sampling sites for Barnacle geese, *Branta leucopsis*, wintering on Inishkea
408 Islands. The islands are situated in the north-east Atlantic Ocean off the west coast of Ireland
409 (54°07'30.2"N; 10°12'31.9"W). Grazing *B. leucopsis* are shown, as is a collection of very large
410 debris items washed ashore by tidal forces (i.e. a large wooden beam, a tractor machinery tyre, and
411 a regular car tyre). Photo credits: Susan Doyle.

412

413 Figure 2: Median counts with interquartile ranges (IQR), and maximum and minimum IQR values,
414 are shown for the total number of recovered synthetic particles, micro-fibres, and micro-fragments.
415 Outlier values are shown.

416

417 Figure 3: Median counts for the total number of recovered synthetic particles, micro-fibres, and
418 micro-fragments in relation to winter season are shown (A, B, C, respectively), as are median
419 counts per gram of faecal sample (D, E, F). Interquartile ranges (IQR), maximum and minimum
420 IQR values, and outlier values are denoted. NS = non-significant, * = $P < 0.05$, ** = $P < 0.01$, ***
421 = $P < 0.001$.

422

423

424

Figure 1:

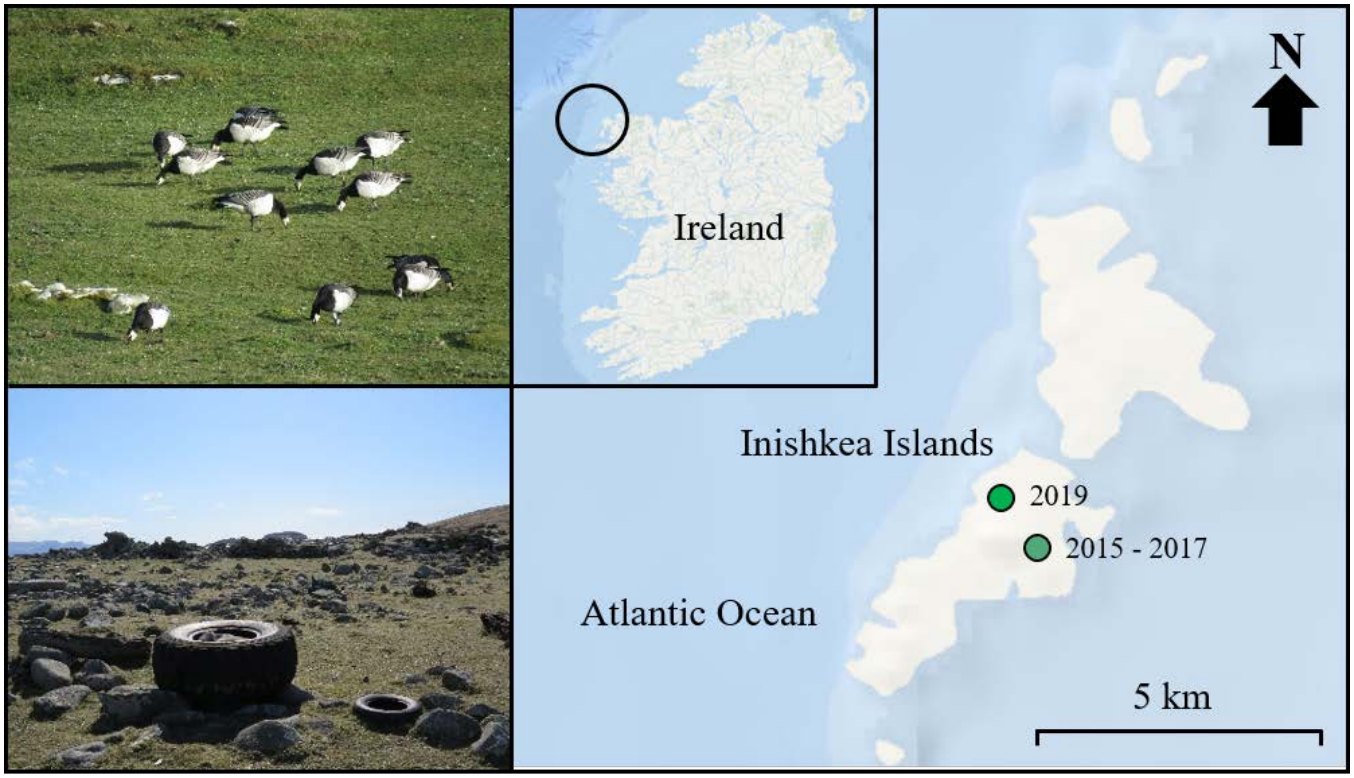

N
Ireland
Inishkea Islands
2019
2015 - 2017
Atlantic Ocean
5 km

Figure 2:

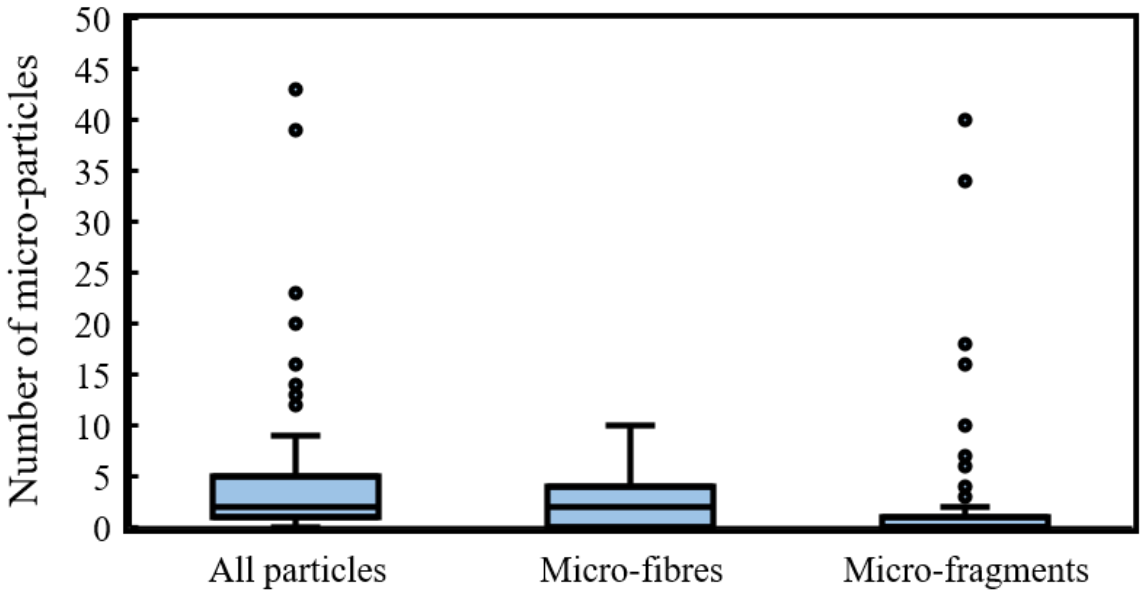
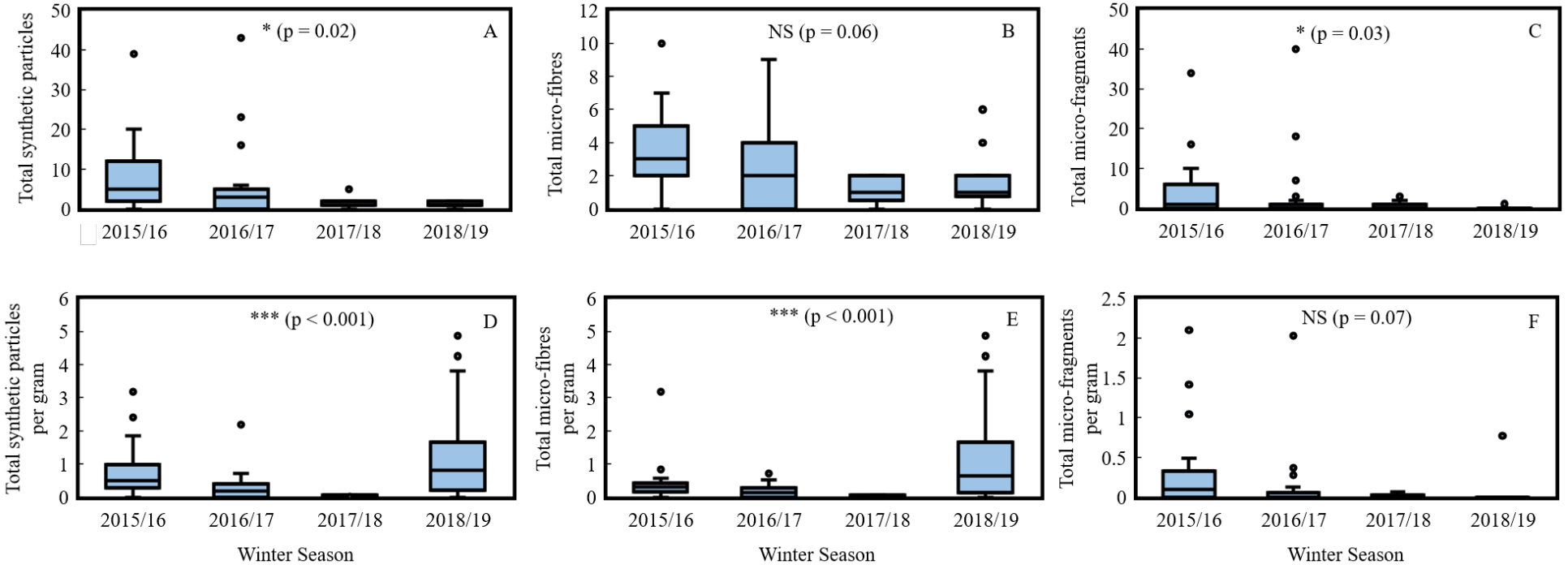

Number of micro-particles
50
45
40
35
30
25
20
15
10
5
0
All particles
Micro-fibres
Micro-fragments

Figure 3:


Total synthetic particles
* (p = 0.02)
A
Total micro-fibres
NS (p = 0.06)
B
Total micro-fragments
* (p = 0.03)
C
Total synthetic particles per gram
*** (p < 0.001)
D
Total micro-fibres per gram
*** (p < 0.001)
E
Total micro-fragments per gram
NS (p = 0.07)
F
2015/16
2016/17
2017/18
2018/19
Winter Season