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**University College Cork, Ireland**  
Coláiste na hOllscoile Corcaigh

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2 Ingestion of anthropogenic debris by migratory barnacle geese *Branta leucopsis* on a  
3 remote north-eastern Atlantic island  
4

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34 Abstract

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

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

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## 58 1. Introduction

59 The ingestion of synthetic debris, i.e. debris composed fully or partially of synthetic  
60 polymers, by birds can lead to increased rates of morbidity and mortality through physical damage,  
61 reduced digestive capacity and appetite, and blockage of the gastrointestinal tract (Pierce et al.  
62 2004; Lavers et al. 2014). Transmission of toxic chemicals from ingested synthetic debris,  
63 especially plastic particles, can also have deleterious effects such as mortality or reduced  
64 reproductive output (Tanaka et al. 2013; 2020). Although relatively large items (pieces > 5mm) of  
65 synthetic debris are most frequently implicated as environmental pollutants, the ingestion of small  
66 synthetic debris (pieces ≤ 5mm) by wildlife is of growing concern (Cole et al. 2011).

67 To date, although seabirds are frequently used as indicator species for anthropogenic  
68 pollution, ingestion of synthetic debris by shorebirds and other water birds remains poorly studied  
69 (Lourenço et al. 2017; Reynolds & Ryan 2018; Rossi et al. 2019). Accordingly, the extent to  
70 which synthetic debris is ingested by water birds inhabiting a combination of terrestrial, freshwater  
71 and marine habitats in coastal areas remains largely unknown (English et al. 2015). Further, the  
72 majority of studies that detail the ingestion of synthetic debris by birds have only focused data  
73 acquisition through necropsies and examination of regurgitation pellets, i.e., boluses comprised of  
74 items that cannot be digested such as shell fragments and stones (Provencher et al. 2017; 2018).  
75 Despite this, birds can also egest synthetic debris that are small enough to pass through the entire  
76 gastrointestinal tract (Reynolds & Ryan 2018; Provencher et al. 2018). As such, the lack of studies  
77 attempting to report ingestion of synthetic debris through assessment of bird faecal samples is a  
78 missed opportunity for environmental monitoring (Provencher et al. 2018), especially as many bird  
79 species will not be readily available for necropsy in sufficient numbers to provide a meaningful  
80 sample size. In addition, as the contents of regurgitated pellets will only correspond to the most  
81 recent meal or meals consumed throughout the previous day, detection of synthetic debris in  
82 regurgitated pellets does not reliably represent the quantities of debris that are retained for an  
83 extended period of time within the gastrointestinal tract (Johnstone et al. 1990; Acampora et al.  
84 2017a). Further, for many species, pellets are often egested relatively quickly, synthetic debris  
85 recovered from regurgitated pellets are not necessarily representative of the shape-types or sizes of  
86 debris that are entering more delicate sections of the gastrointestinal tract where absorption of  
87 contaminants or physical blockages could more readily occur. Synthetic debris recovered from  
88 egested faecal samples could be more representative of non-retained debris, i.e. items that pass  
89 through the gastrointestinal tract relatively quickly with digestive boluses, and larger debris items  
90 that have been broken down to smaller pieces that can pass through the gut and intestines  
91 (Provencher et al. 2018; Le Guen et al. 2020).

92 Barnacle geese, *Branta leucopsis*, inhabit a mixture of marine, freshwater and terrestrial  
93 habitats (Black et al. 2014). As obligate herbivores, *B. leucopsis* traditionally graze on the leaves,  
94 stems, roots and seeds of *Plantago/Bellis/Festuca* swards, however, they are increasingly observed  
95 to forage on semi-improved agricultural grasslands (Cabot & West 1973; Mason et al. 2017).  
96 Between October and April, migratory *B. leucopsis* are found wintering in north-western locations  
97 of Ireland and Scotland, where flocks typically forage in coastal pastures, salt marshes, river  
98 estuaries, tidal mud flats and offshore islands (Doyle et al. 2018; Mitchell & Hall 2018). Like most  
99 obligate herbivores, the digestive system of *B. leucopsis* is relatively inefficient with a low  
100 assimilation efficiency of ~30%, i.e. ~70% of ingested food items rapidly moves through the gut  
101 undigested (Owen 1971; Black et al. 2014). In general, *B. leucopsis* have gut retention times of  
102 between 1.9–3.1 hrs (Prop & Vulink 1992). Accordingly, we suspect that *B. leucopsis* could act as  
103 a sentinel species for the presence of synthetic debris in coastal environments through non-  
104 invasive analysis of egested faecal samples. Therefore, in the present study, we assess *B. leucopsis*  
105 faecal samples for the presence of synthetic debris across multiple sampling periods, from 2015–  
106 2019. Faecal samples were obtained from a *B. leucopsis* wintering on a remote, offshore north-  
107 eastern Atlantic island. As the geese tend to remain on the island for the duration of their wintering  
108 season and given their gut retention times, any recovered egested synthetic debris are most likely  
109 representative of on-island pollution.

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

116

## 117 2. Methods

### 118 2.1 Sample collection

119 A total of 71 faecal samples were collected from adult or first-winter *B. leucopsis* wintering  
120 on Inishkea Islands, situated in the north-east Atlantic Ocean off the west coast of Ireland  
121 ( $54^{\circ}07'30.2''\text{N}$ ;  $10^{\circ}12'31.9''\text{W}$ ; Figure 1). Samples were collected from the south island (1.84 km<sup>2</sup>).  
122 Faecal samples were collected in March 2015 ( $n = 17$ ), November 2016 ( $n = 14$ ), April and  
123 November 2017 ( $n = 13$  and  $7$ , respectively), and March 2019 ( $n = 20$ ). Samples obtained in 2015,  
124 2016 and 2017 were taken directly from individual birds captured during routine population  
125 monitoring. In 2019, fresh faecal samples were collected from a monospecific roosting site. These  
126 samples were collected at distances of at least one metre apart, to ensure that they were produced

127 by different individual birds. Samples collected in 2019 were refrigerated ( $\leq 4$  months), while all  
128 others were kept frozen ( $-20\text{ }^{\circ}\text{C}$ ) until required. All birds were handled under licence from the  
129 National Parks and Wildlife Service of the Government of Ireland (Section 32, Wildlife Acts  
130 1976–2012), with relevant ringing permits from the British Trust of Ornithology.

131

## 132 2.2 Digestion, separation and microscopy

133 Samples were transferred into individual glass beakers and weighed on an analytical balance  
134 (0.01 g). To eliminate labile organic matter (including non-synthetic anthropogenic debris deriving  
135 from a natural source, e.g. 100% cotton, linen or wool fibres), samples were digested in solutions  
136 of Iron(II) sulfate heptahydrate ( $\text{FeH}_{14}\text{O}_{11}\text{S}$ ) and 30% hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) at  $60\text{ }^{\circ}\text{C}$  (0.75 g  
137 of  $\text{FeH}_{14}\text{O}_{11}\text{S}$  per 50 mL  $\text{H}_2\text{O}_2$ ), until total digestion had occurred (Masura et al. 2015). An  
138 approximate application ratio of 20 mL per 10 g of faecal sample was employed. Following  
139 digestion, a density based separation technique was used to isolate synthetic debris from denser  
140 undigested mineral components through flotation, using a saturated solution of NaCl, i.e.  $360\text{ g L}^{-1}$ .  
141 The resulting supernatant was carefully decanted and vacuum filtered onto filter pads (Whatman  
142 41, 47 mm,  $20\text{ }\mu\text{m}$  pore). All filter pads were placed in clean glass petri dishes and dried at room  
143 temperature. All samples were then examined under a stereomicroscope (Olympus SZX16). All  
144 synthetic debris particles were visually identified using the criteria outlined by Zhao et al. (2016).  
145 Recovered synthetic debris was classified into shape-type categories (e.g. fibre, fragment), size  
146 range and colour tone (Provencher et al. 2017; Bessa et al. 2019).

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

157

## 158 2.3 Statistical Analyses

159 All data were assessed for normality of residual distributions (Shapiro-Wilk test,  $P > 0.05$ )  
160 and homoscedasticity of variances (Fligner-Killeen,  $P > 0.05$ ). As all data were non-normal ( $P <$   
161  $0.05$ ), separate Kruskal–Wallis test with a Dunn’s post hoc were used to examine total count data

162 in relation to the dependent variable of winter season. Wintering season spans the months of  
163 October to April, inclusive, and represents the time period for which the *B. leucopsis* population  
164 dwells on Inishkea Islands. Where residuals did not meet homoscedasticity assumptions ( $P <$   
165  $0.05$ ), a White-adjusted ANOVA with Tukey LSM post hoc tests were employed instead.  
166 Similarly, the number of synthetic particles per gram of faecal sample was also assessed in relation  
167 to the dependent variable of winter season. The biomass of collected faecal samples was also  
168 considered in relation to winter season using Kruskal–Wallis tests. All statistical analyses were  
169 performed using R version 3.5.3 (R Core Team 2019).

170

### 171 3. Results

172 In total, 331 synthetic particles were detected; 166 were micro-fibres and 165 were micro-  
173 fragments. However, while 79% of samples were found to contain synthetic particles, the number  
174 detected per sample was generally low at  $4.7 \pm 0.9$ , ranging from 0–43 particles (mean  $\pm$  SE):  
175 micro-fibres  $2.3 \pm 0.3$  (0–10); micro-fragments  $2.3 \pm 0.8$  (0–40) (Figure 2). When standardised in  
176 relation to the quantity of faecal mass per sample, synthetic particles were detected at a mean  
177 ( $\pm$ SE) frequency of  $0.67 \pm 0.12$  particles  $g^{-1}$ , ranging from 0–4.88 particles  $g^{-1}$ . Micro-fibres and  
178 micro-fragments were observed at a frequency of  $0.52 \pm 0.12$  particles  $g^{-1}$  (0–4.88) and  $0.15 \pm 0.05$   
179 (0–2.10), respectively. Potential laboratory contamination was low, with only three micro-fibres  
180 being detected by control filter pads, i.e. 0.04 per processed faecal sample, therefore no  
181 adjustments to the results were made. All synthetic particles recovered from faecal samples had a  
182 maximum length within the size range of 0.5–5 mm, and therefore, could be categorised as either  
183 micro-fibres or micro-fragments (Torre et al. 2016; Zhao et al. 2016). Micro-fibres had dark (92%;  
184 e.g. navy-blue, black, dark red) or mid colour tones (8%; e.g. blue, red), while micro-fragments  
185 consisted of mid (13%; e.g. blue, blue-green) or light (87%; e.g. clear, white-blue, yellow) colour  
186 tones.

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

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

199

#### 200 4. Discussion

201 Here, to our knowledge, ingestion of synthetic debris by *B. leucopsis* has been recorded for  
202 the first time. Further, it is clear that this phenomenon has been ongoing over a number of years at  
203 an isolated and remote location. Although this is the first record of synthetic debris ingestion by  
204 Anatidae in Ireland, previous records have documented ingestion of debris by Anatidae dwelling  
205 within anthropogenically disturbed sites across continental Europe (Gil-Delgado et al. 2017),  
206 North America (English et al. 2015; Holland et al. 2016), and South Africa (Reynolds & Ryan  
207 2018). In addition, a variety of coastal dwelling seabird species have previously been recorded to  
208 ingest synthetic debris in coastal locations of Ireland, in-line with international trends for debris  
209 ingestion by seabird species (Acampora et al. 2016; 2017a,b). Nevertheless, this is the first study  
210 to consider multiyear assessment for a single population of a non-obligate marine bird species  
211 residing within a remote area, with 4–8 years of sampling being needed to reveal possible trends  
212 (e.g. van Franeker & Meijboom 2002). For example, although the total number of synthetic  
213 particles detected decreased between 2015/16–2018/19 wintering seasons, the total number  
214 particles recovered per gram of faecal sample has generally remained the same across the  
215 wintering seasons, suggesting that synthetic particles are ubiquitous on the Inishkea Islands.  
216 Although 2018/19 samples do suggest an increase in the number of particles detected per gram,  
217 this appears to be an artefact of the 2019 samples, which were taken from a unique sample location  
218 (i.e. a roosting site), as well as this relationship being largely driven by three outlier samples.  
219 However, it is worth noting that a truly accurate assessment of any change in debris ingestion  
220 frequencies (e.g.  $\pm 5\%$  detection rate with a sampling power of 80%) would likely require annual  
221 sampling of  $>14,000$  birds (Lavers & Bond 2016), which is simply not feasible for the vast  
222 majority of study systems.

223 In the present study, a relatively high frequency of synthetic debris ingestion by *B. leucopsis*  
224 was recorded, with 79% of samples having at least one synthetic particle. However, for Anatidae,  
225 frequency of ingestion appears to vary amongst species and sampling locations, e.g. 4.3–53.8%  
226 (English et al. 2015), 0–50% (Holland et al. 2016), 43.8–60% (Gil-Delgado et al. 2017), and 0–  
227 17% (Reynolds & Ryan 2018). However, despite outliers, the mean number of debris items  
228 recovered per sample tended to be similar, if slightly higher, to amounts reported by other studies  
229 (e.g. English et al. 2015; Gil-Delgado et al. 2017). To date, studies assessing waterfowl faecal  
230 samples for the presence of synthetic debris, including the present study, have not considered  
231 possible in-field contamination. This is particularly problematic for micro-fibres, as wind or soil-



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

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

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

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

281

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290

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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.

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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.

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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 =  $P < 0.001$ .

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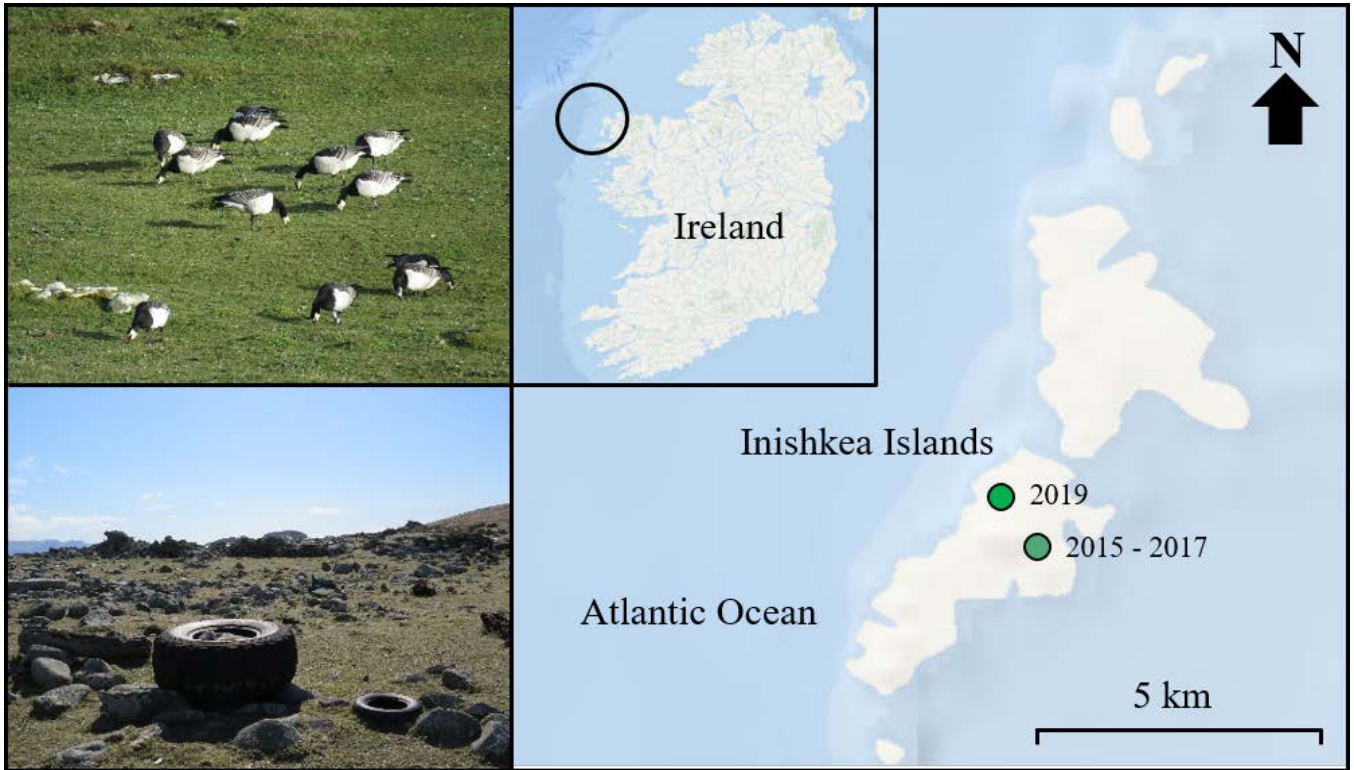
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427 Figure 1:

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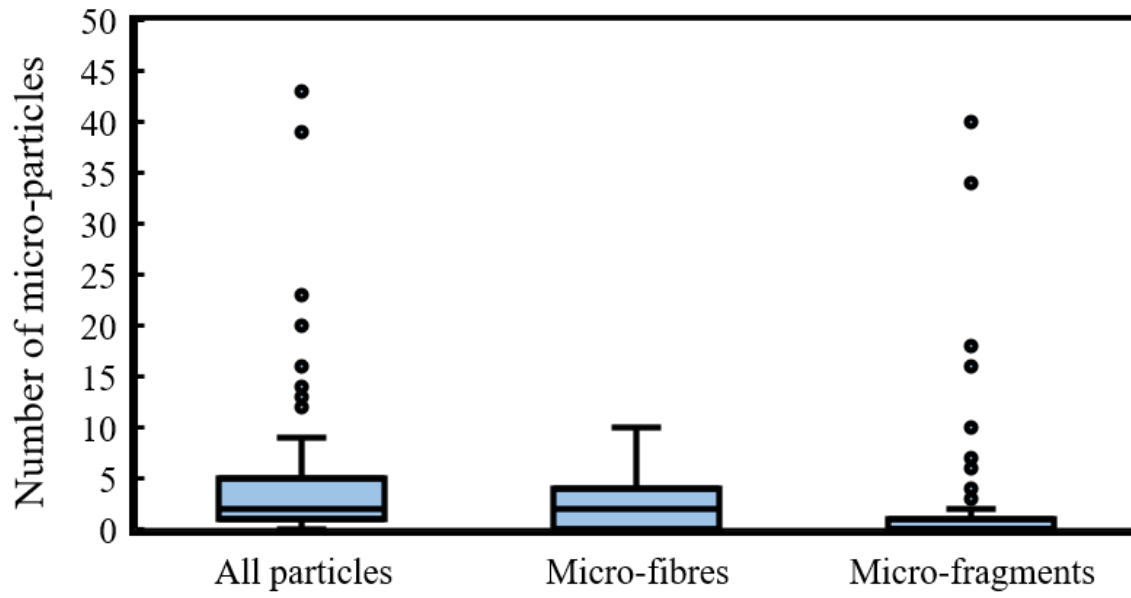
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449 Figure 2:

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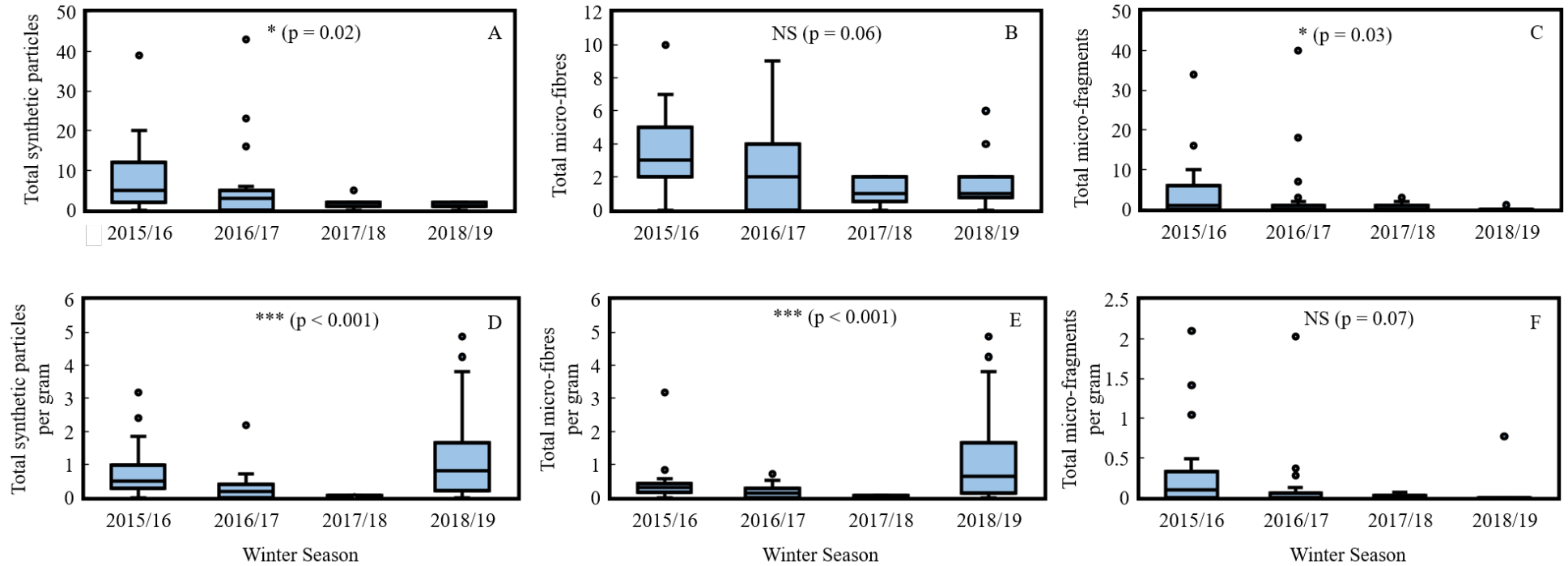
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