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Plastic ingestion in adult and fledgling Manx Shearwaters *Puffinus puffinus* on Skomer Island, Wales

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Abstract

Plastic debris is ubiquitous in the marine environment, and seabirds are among the most impacted marine vertebrates. Plastic ingestion was investigated in an internationally-important breeding population of Manx Shearwaters *Puffinus puffinus* on Skomer Island, Wales in 2018–19. Opportunistic necropsies were carried out on a total of 34 birds to collect contents of the gastrointestinal tract: 13 adults in April – July 2018 and 12 fledglings during September 2019, and a further nine opportunistic necropsies carried out from adults in July – September 2018. The presence, quantity, size and colour of plastic in the gastrointestinal tract was investigated. Plastic was found in 71% of stomach contents (68% of adults, 75% of fledglings). Adults were found to have larger plastic pieces in their gastrointestinal tract than fledglings, and lighter birds had larger pieces of plastic in their tracts than heavier birds. This study shows that Manx Shearwaters on Skomer Island are vulnerable to plastic ingestion, and that adults are likely to pass plastic to their chicks.

Introduction

Approximately 20 million tonnes of plastics enter aquatic ecosystems annually, with plastics continuing to accumulate as inputs exceed attempts at mitigation (Borrelle *et al.* 2020). The impacts of plastic pollution have been well-documented, affecting marine fauna via both entanglement and ingestion (Wilcox *et al.* 2015; O’Hanlon *et al.* 2017). Amongst marine vertebrates, seabirds are most affected by plastic (Lavers *et al.* 2014), being known to forage in convergence zones (Gould *et al.* 1997; Pichel *et al.* 2007) with high concentrations of ocean plastic (Wilcox *et al.* 2015). Therefore, plastic pollution in the marine environment may be contributing to the global decline of seabirds, a decline which is occurring more quickly than in any other bird group (Lavers *et al.* 2014).

Plastic ingestion in seabirds was first noted in the 1960s (Moser & Lee 1992), in the Laysan Albatross *Phoebastria immutabilis*. In the 1970s, Northern Fulmars *Fulmarus glacialis* in the North Sea were found to have 1–2 plastic pieces per stomach, but by the 1980s this had risen to an average of 10 pieces per stomach (van Franeker, 1985; van Franeker & Law 2015). Since then, it has been reported that more than 100 different seabird species have ingested plastic (Provencher *et al.* 2018; Kühn & van Franeker 2020), exceeding 90% of individuals in some studies (van Franeker *et al.* 2011; Codina-Garcina *et al.* 2013; Roman *et al.* 2016). By 2050 it is predicted that 99% of seabirds will ingest plastic regularly (Wilcox *et al.* 2015; Provencher *et al.* 2018). Entanglement in plastic litter is also a threat, with 36% of seabird species known to have been affected in this way. Furthermore, of the 265 bird species entangled globally, 83% were caught up in fishing gear (Ryan 2018). Additionally, many seabirds use plastic as nesting material, including Double-crested Cormorants *Phalacrocorax auritus* (Podolsky & Kress 1989) and Northern Gannets *Morus bassanus* (Montevecchi 1991; Votier *et al.* 2011; Rodríguez *et al.* 2013), increasing the risk of plastic entanglement and ingestion for both breeding adults and their chicks (Podolsky & Kress 1989).

Seabirds may ingest plastic either directly, often mistaking plastic for potential prey items (Azzarello & van Vleet 1987; Moser & Lee 1992; Shaw & Day 1994; Derraik 2002; van Franeker *et al.* 2011), or indirectly, via lower trophic levels, including filter-feeding organisms and other prey species (Graham & Thompson 2009; van Franeker *et al.* 2011; Ryan 2015; O’Hanlon *et al.* 2017). Plastic ingestion may increase mortality rates in seabirds through a number of mechanisms: shredding the stomach lining, causing blockages that lead to starvation, taking up stomach space that results in poor energy return (Petry *et al.* 2008), suppressing appetite, or decreasing fat deposition (Auman *et al.* 1997). Additionally, chemicals, such as alkylphenols, phthalates and organophosphates, are used in the

production of plastic (Lahens *et al.* 2018; Rhodes 2018) and, once ingested, can leach into an organism's tissues and potentially disrupt endocrine systems (Oehlmann *et al.* 2009), with adverse effects on development and reproduction (Oehlmann *et al.* 2009; Lusher *et al.* 2015). In water, plastic can also absorb persistent organic pollutants (POPs) such as polychlorinated biphenyls (PCBs), persistent bioaccumulative and toxic substances (PBTs) and trace metals, posing additional threats if they are ingested by organisms (Rochman *et al.* 2013; Lahens *et al.* 2018).

Although plastic is not biodegradable, wave action and ultraviolet light causes macroplastics to fragment, producing microplastics (Ter Halle *et al.* 2016; Weinstein *et al.* 2016; Ostle *et al.* 2019). This is problematic as microplastics are harder to remove from the environment and more easily ingested than larger plastic items (Auta *et al.* 2017).

Olfactory foraging, using dimethyl sulphide (DMS) as a cue to find productive areas of ocean and thus prey, can lead seabirds to confuse plastic for food items, because plastics in the marine environment also emit a DMS signature (Savoca *et al.* 2016). The surface, shallow-diving foraging habit of Procellariiformes compounds this by bringing birds into contact with buoyant plastics at the surface (e.g. Hammer *et al.* 2016). Furthermore, Procellariiformes have a constriction in the digestive system between the gizzard and proventriculus, along with a small gizzard, making it difficult for ingested plastic pieces to be regurgitated (Azzarello & van Vleet 1987); for example, ingested balloons present a particularly high risk of mortality to seabirds (Roman *et al.* 2019a). Indeed, Manx Shearwaters *Puffinus puffinus* (one of the most widely studied Procellariiformes) are known to ingest plastics: a study in Brazil found that 83% of items found in the stomach contents (N = 25) were plastic (Colabuono *et al.* 2009). Interestingly, Manx Shearwater fledglings may have an increased risk of plastic ingestion through intergenerational transfer, as adults regurgitate plastic during chick feeding, whereas chicks regurgitate less frequently, causing plastic to accumulate more within their bodies (Ryan 1988; Carey 2011). This can lead to increased pollutant concentrations in tissues, consequently reducing fledgling body condition (Day *et al.* 1985; Sileo *et al.* 1990; van Franeker & Meijboom 2002; Hutton *et al.* 2008; Colabuono *et al.* 2009; Carey 2011; Acampora *et al.* 2016; Lavers & Bond, 2016).

In this study, the presence, quantity, colour and size of plastic found in the gastrointestinal tract of Manx Shearwaters was investigated to discern: 1) differences between adult and fledgling birds, 2) associations between body weight and plastic ingestion, and 3) patterns of ingested plastic colour, as previous studies have found preferential selection due to plastics being mistaken for prey (e.g. Azzarello & van Vleet 1987; Verlis *et al.* 2013). This study is the first to investigate plastic ingestion in Manx Shearwaters in the United Kingdom on their breeding grounds and, more specifically, a

population in the north-eastern Atlantic; an area poorly studied in terms of plastic pollution (O'Hanlon *et al.* 2017).

Methods

Study site

The study was undertaken on Skomer Island, Wales (51°44' N, 5°19' W). Approximately 350,000 pairs of Manx Shearwater breed on Skomer and neighbouring Skokholm Island, an estimated 50% of the world population (Perrins *et al.* 2012; 2019).

Sample collection and analysis

Manx Shearwater carcasses were collected opportunistically around the island, following collisions with buildings or partial predation attempts by Great Black-backed Gulls *Larus marinus* and Ravens *Corvus corax*. Only fresh carcasses with apparently intact stomach contents and negligible decay or predation were collected. Adult birds were collected in July – September 2018 (N = 9) and April – July 2019 (N = 13), whilst fledglings were collected during September 2019 (N = 12). Carcasses were placed into individual plastic bags and frozen until necropsied.

Necropsies took place in facilities to minimise cross-contamination between carcasses – only one necropsy was conducted at a time, and the lab bench and dissection equipment cleaned between necropsies. Lab coats were worn to minimise the risk of contamination from clothing. Each carcass was weighed (dry weight), using a Force Gauge with Gram-Division (1,000 g), and, if possible, the suspected cause of death was determined by assessing body condition (van Franeker *et al.* 2011). Carcasses were dissected following van Franeker (2004). Once the gastrointestinal tract was free, both the proventriculus and gizzard were cut open and the contents collected. Samples were rinsed in cold water (as advised in Provencher *et al.* 2017) and filtered using a 0.3 mm stainless steel sieve onto unbleached filter paper, to retain plastic pieces of a minimum of 0.3 mm. The filtered samples were rinsed again with cold water directly onto filter paper, which we assume to have made little difference to the plastics sampling. All samples were also rinsed separately with diluted disinfectant to prevent cross-contamination.

The contents of the gastrointestinal tract from each individual were analysed under a GXM XTL3 101LED2 binocular stereo microscope with a GXCAM Hichrome MET-M camera and screen. Any plastic was identified either following van Franeker *et al.* (2011), or via a float test where any substance that floated on water was classed as plastic (Nuelle *et al.* 2014; Norris 2019) and counted. Measurements (nm) of the length and width of each plastic piece were noted, to provide an average plastic size per

bird stomach, and the colour of each piece of plastic was categorised as either blue, clear, orange/yellow, black/brown, green or pink.

Statistical analysis

A binary logistic regression was undertaken to assess the probability of plastic presence using age (adult or fledgling) as a categorical predictor and body weight as a continuous predictor, and an interaction between the two terms. A Poisson regression was undertaken to investigate plastic count and average size, using age (adult or fledgling) as a categorical predictor and body weight as a continuous predictor, and an interaction between the two terms. A Chi-square test of association was undertaken to assess the number of plastic pieces of different colour category in relation to age (adult or fledgling). All analyses were undertaken using Minitab v.19.2020.

Results

Plastic presence

Overall, 24 out of the 34 (71%) Manx Shearwaters sampled contained plastic in their stomachs. In total, 15 out of 22 (68%) adult Manx Shearwater stomachs had plastic present, whereas nine out of 12 (75%) fledgling stomachs contained plastic. Plastic was only found in the ventriculus of the birds.

There was no significant effect of age ($Z = -0.02$, $N = 34$, $P = 0.980$) or weight ($Z = 0.91$, $N = 34$, $P = 0.365$) on the probability of plastic presence in Manx Shearwaters, and no significant interaction between the terms ($Z = 0.10$, $N = 34$, $P = 0.923$). It must be noted that sample sizes are small which may limit the power of these analyses.

Plastic count

The average number of plastic pieces found in Manx Shearwaters was 2.96 ± 2.77 SD. Where possible cause of death was determined (11/22, 50% adults), none of the deaths was directly linked to plastic ingestion. However, those killed by predation (3/11, 23%) had ingested the highest number of plastic pieces, accounting for 56% of the total number of plastic pieces found (Table 1).

Table 1. Suspected cause of death for adult Manx Shearwaters *Puffinus puffinus*.

Suspected cause of death	Number of individuals	Average number of plastic pieces per individual (\pm SD)
Predation	3	5.3 (\pm 6.66)
Building collision	5	1.2 (\pm 2.17)
Starvation	3	0.7 (\pm 0.58)
Unknown	11	3.6 (\pm 3.91)

Although the proportion of adult Manx Shearwaters with plastics in their ventriculus was lower than the proportion of fledglings, there was more variability in the number of pieces of plastic ingested by the adults, with several individuals ingesting relatively high numbers of plastic (Table 2).

There was no significant effect of age ($Z = -0.91$, $N = 24$, $P = 0.365$) or weight ($Z = 0.02$, $N = 24$, $P = 0.984$) on the number of items of plastic found in the ventriculus of Manx Shearwaters, and no significant interaction between the terms ($Z = 0.92$, $N = 24$, $P = 0.355$).

Table 2. The number of plastic pieces ingested by adult and fledgling Manx Shearwaters *Puffinus puffinus*.

Age of bird	Number of birds dissected	Number with plastic	Number of plastic pieces ingested	Average number of plastic pieces ingested per individual (\pm SD)
Adult	22	15	46	3.06 (\pm 3.07)
Fledgling	12	9	25	2.70 (\pm 1.93)

Plastic size and colour

All plastics found were microplastics (< 5 mm) with the largest plastic fragment measuring 33,331.90 nm² (mean size = 3,517 nm² \pm 4,967 SD). There was a positive effect of both age ($Z = 10.07$, $N = 24$, $P < 0.001$; Figure 1) and weight ($Z = 57.32$, $N = 24$, $P < 0.001$; Figure 1) on the size of plastic piece found in the ventriculus of Manx Shearwaters, whereby adults ingested significantly larger plastic pieces than fledglings, and heavier birds ingested significantly smaller plastic pieces. There was also a significant interaction between the terms ($Z = -26.45$, $N = 24$, $P < 0.001$; Figure 1), where there was a positive relationship between body weight and plastic size in adult birds, and a slight negative relationship between body weight and plastic size in fledgling birds.

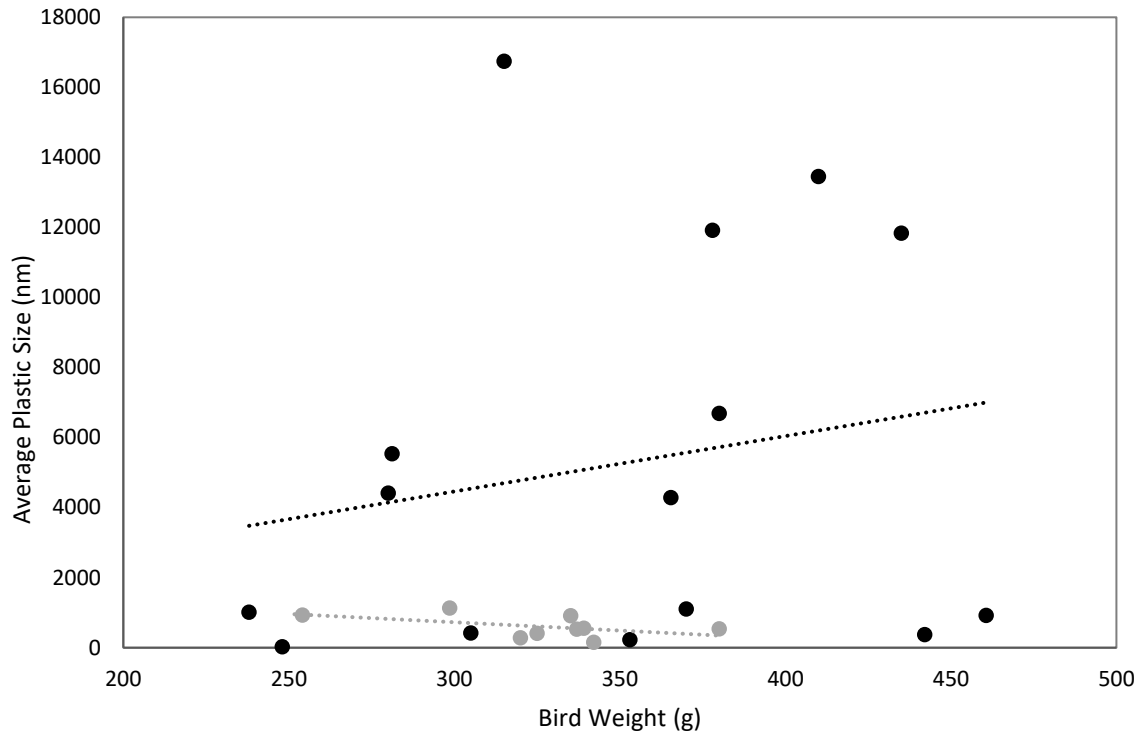


Figure 1. The average size of pieces of plastic ingested by Manx Shearwaters *Puffinus puffinus*, in relation to weight and age. Adults are represented by black dots and trendline, fledglings are represented by grey dots and trendline.

Significantly higher numbers of black/brown and pink plastic were found in the ventriculus of fledgling Manx Shearwaters, whereas significantly higher numbers of clear plastics were found in adults ($X^2 = 47.57$, d.f. = 4, $P < 0.001$; Figure 2).

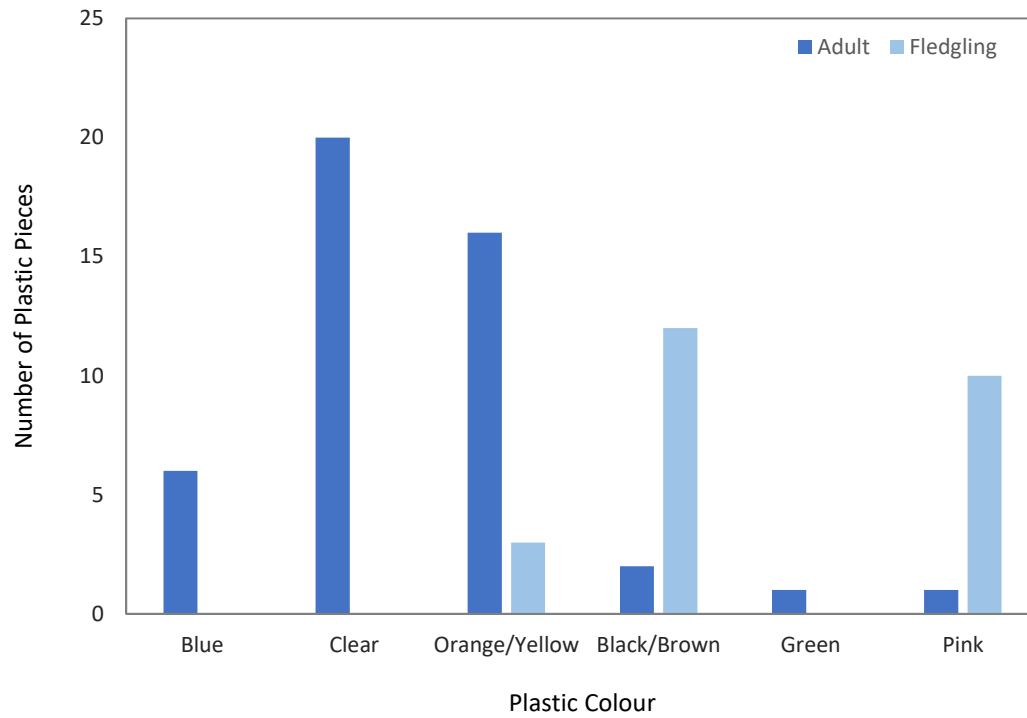


Figure 2. Different colours and quantities of plastic found in the stomach contents of Manx Shearwaters *Puffinus puffinus*.

Discussion

The majority of birds sampled (71% overall; 68% of adults and 75% of fledglings) had at least one plastic piece in their gastrointestinal tract. The evidence of plastic ingestion in this study is consistent with other studies which have highlighted that Procellariiformes are surface-feeders and, as such, are highly susceptible to plastic ingestion (van Franeker *et al.* 2011; Provencher *et al.* 2014; Acampora *et al.* 2016). It should be noted that the Manx shearwaters have not necessarily ingested the plastics from the Skomer area. For example, Acampora *et al.* (2016) found evidence of plastic ingestion in Manx Shearwaters on the nearby coast of Ireland, although only one piece of plastic was found from three carcasses. The foraging range of Manx Shearwaters is such that birds in our study could also have been feeding in the waters around Ireland during the breeding season (Wischnewski *et al.* 2019). However, as Manx Shearwaters winter in South America each year (Guilford *et al.* 2009), they could have ingested the plastics at any point of their journey.

Although a higher proportion of fledglings had ingested plastic in their digestive tracts compared to adults, the adults contained a greater number of pieces, with a higher variability in the number of pieces ingested between individuals. This could be explained by the fact that adults spend time at sea foraging and so are more likely to come across plastic than fledglings (Spear *et al.* 1995). Also, the most important pathway of plastic ingestion for fledglings is through adult regurgitation during feeding (Ryan 1988; Carey 2011). During this process, is it unlikely that all plastic pieces ingested by the adult would be regurgitated (Spear *et al.* 1995). It has also been noted that eliminating plastic through regurgitation may be difficult for Procellariiformes, due to the constriction between the proventriculus and the gizzard (Azzarello & van Vleet 1987; Spear *et al.* 1995). Therefore, unless the plastic had not yet reached the gizzard at the point of regurgitation it is likely to be retained by the adult (Spear *et al.* 1995).

A high proportion of plastic fragments found in the stomach contents of Manx Shearwaters can be considered as microplastic (< 5 mm) and are reported to be the most-ingested size of plastic in seabirds (Bond *et al.* 2014). This supports the fact that the average size of plastic particles in the oceans are decreasing (Barnes *et al.* 2009). Manx Shearwaters' prey may ingest microplastics, so indirect plastic consumption is possible (Provencher *et al.* 2015; Ryan 2015). Alternatively, fragments may have broken down in the stomach due to mechanical and chemical damage (Andrady 2000; Barnes *et al.* 2009). Also, individuals that ingest macroplastics might die at sea, due to obstruction of the gastrointestinal tract, so ingestion of macroplastics may be underestimated (Pierce *et al.* 2004). There

was a significant difference between the size of plastic pieces found in the stomach contents of adults and fledglings, with adults found to have ingested larger pieces. This could be explained by adults regurgitating smaller pieces when feeding chicks, whilst the larger pieces may remain stuck in the adult's digestive tract (Petry *et al.* 2008). A positive relationship was found between the size of plastic pieces and adult bird weight, with larger plastic pieces associated with heavier birds. In comparison, a negative correlation was found between plastic size and fledgling body weight, where larger plastic pieces were associated with lighter birds. This could be because larger plastic pieces are more likely to cause internal injuries, and may increase PCB concentrations, so are more likely to reduce the overall condition of the bird, leading to weight loss (Auman *et al.* 1997). In addition, bird weight may be lower as plastic can retain in the gut, causing blockages and reducing the deposition of fat and absorbing nutrients (Ryan 1990; Spear *et al.* 1995; Auman *et al.* 1997). Furthermore, plastic induces the feeling of satiation and infrequent feedings, causing a high chance of physiological stress, thus lowering their overall body weight (Auman *et al.* 1997).

Clear and yellow plastics were most commonly ingested by adult Manx Shearwaters, which could suggest that they are mistaking them for prey (Azzarello & van Vleet 1987). Indeed, a study on Wedge-tailed Shearwaters *Ardenna pacifica* found that 62.5% of plastics ingested were of light tone (clear/white, yellow and green), a similar colour to their prey (Verlis *et al.* 2013). Alternatively, it has been reported that light-coloured plastics dominate the marine environment, so it could be that birds are simply more likely to come across these colours whilst foraging (Moser & Lee 1992; Shaw & Day 1994). Indirect ingestion of certain plastic colours through the prey of Manx Shearwaters, or in water ingested when catching prey, could also be a factor (Sileo *et al.* 1990). In comparison, fledglings were found to have black and pink plastics in their gastrointestinal tract, with no clear plastic found at all. As the fledglings in this study were not yet at an age to forage for themselves, they will have accumulated plastic that was fed to them by their parents before fledging (Carey 2011; Rodríguez *et al.* 2012), i.e. they were unlikely to have ingested plastics when foraging on their own. There is also the possibility of human error in identifying colours, as well as the fact that the plastic samples from the fledglings were washed with a disinfectant cleaning solution which could have affected the colour of the plastic (Provencher *et al.* 2017). A final possibility for the colours of plastic being ingested, could be due to the ultraviolet (UV) light that the plastic absorbs (Verlis *et al.* 2013). As many birds are sensitive to light in the ultraviolet range, UV information is used in foraging and signalling and so, may be a factor in Manx Shearwaters selecting the plastic found in this study (Verlis *et al.* 2013).

Whilst no deaths could be confirmed to be caused by plastic ingestion, there was a high incidence of plastic ingestion for birds that died from predation (although the sample size of three birds was very small), which might indicate that ingested plastic lowered the condition of those birds, making them more vulnerable to predation (Provencher *et al.* 2019). Causes of death were mostly unknown, so mortality due to plastic-related causes, such as toxicity, or internal wounds, may remain undetected (Codina-García *et al.* 2013).

This study shows that plastic ingestion is a frequent occurrence for Manx Shearwaters that breed on Skomer Island. Although the ingestion rates reported here are not as high as areas of the Pacific Ocean, where the risk is much greater (van Franeker and Law 2015; Wilcox *et al.* 2015; Roman *et al.* 2019b), the potential impacts on Manx Shearwaters warrant further study.

Author statement

C.L.A., S.A. & E.A. carried out fieldwork, necropsies and data collection. C.L.A., L.K.G. and S.A. wrote the manuscript, and C.L.A. and L.K.G. conducted statistical analyses. M.A., L.K.G. & M.J.W. advised on project design, analysis and writing, E.A. commented on the manuscript, and M.J.W. provided support and training in the field.

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