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## **Plastic occurrence in six different species of Arctic seabirds**

Harmonizing methods and closing knowledge gaps on plastic occurrence and polymer identity

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Photo by Vegard Stürzinger

# Abstract

The once pristine Arctic is now facing negative alterations with a rapidly changing climate, increasing human activity, and plastic pollution. Seabirds are intrinsic to the marine ecosystems in the Arctic, but most of the seabird populations are declining, and many species are listed as threatened. In recent years, plastic ingestion by seabirds has been of increasing concern because of the potential negative impact on individual and population levels. There is an urgent need for more information on plastic pollution in seabirds to enable risk assessment and monitoring, as the rate of ingestion is expected to increase continuously. The aim of this study was to investigate the plastic occurrence and characterize the polymer identity of plastics found in six seabird species breeding in the Arctic. The study species were Atlantic puffins (*Fratercula arctica*), Common guillemots (*Uria aalge*), Razorbills (*Alca torda*), Great cormorants (*Phalacrocorax carbo*), European shags (*Phalacrocorax aristotelis*), and Glaucous gulls (*Larus hyperboreus*). We showed that the Atlantic puffins had a high frequency of occurrence of plastic and proved plastic ingestion for the first time in Glaucous gulls. The plastics in the Atlantic puffins were identified as polyethylene (PE) and polypropylene (PP), and the plastics in the Glaucous gull were identified as PP and polystyrene (PS). User plastics dominated over industrial plastics in both of the seabird species. No plastic was detected in the Common guillemots, Razorbills, Great cormorants, and European shags. Non-invasive methods to detect and measure plastic in seabirds should be further investigated, and research on the toxic effects from plastic exposure should be intensified. The study has proven to be valuable by providing new quantitative and qualitative data on plastic loading and polymer type reported in a standardized manner which can be used to establish a baseline for future research and monitoring of Arctic seabirds on a national and international level.

# Table of Contents

Abstract.....	i
Acknowledgements.....	iv
Abbreviations .....	v
Chapter 1 .....	1
Introduction.....	1
Marine plastic pollution .....	1
Plastic occurrence in Arctic Seabirds .....	4
Study species.....	7
Aim of study .....	14
Chapter 2.....	15
Material and method .....	15
Seabird sampling.....	15
Stomach dissection and plastic analysis .....	17
Fourier-Transform infrared (FT-IR) spectroscopy analysis .....	18
Quality assurance .....	21
Data analysis and statistics.....	21
Chapter 3.....	22
Results.....	22
Birds in the study .....	22
Plastic found in the stomachs.....	22
Chapter 4.....	26
Discussion .....	26

Plastic occurrence .....	26
Type of plastic.....	30
Sample size .....	34
Sample collection and dissection .....	35
Future research.....	36
Conclusion .....	38
References.....	39
Appendix A.....	45



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# Abbreviations

**ATR** Attenuated Total Reflectance.

**DMS** Dimethyl Sulfide.

**EcoQO** Ecological Quality Objective.

**FO** Frequency of Occurrence.

**FTIR** Fourier Transform Infrared.

**GIT** Gastrointestinal tract.

**HQI** Hit Quality Index.

**IUCN** International Union for Conservation of Nature.

**PE** Polyethylene.

**PET** Polyethylene Terephthalate.

**PP** Polypropylene.

**PS** Polystyrene.

# Chapter 1

## Introduction

### Marine plastic pollution

Plastic pollution in the marine environment is an increasing global concern and plastics are found everywhere in the ocean, from the Arctic to the Antarctic, occurring in all marine environmental compartments; it has become a ubiquitous problem (UNEP 2016; Tirelli et al. 2020; Baak et al. 2021).

In 2019 the global plastic production reached almost 370 million metric tons (Plastics-Europe 2020). Plastics are synthetic polymers, and the majority of plastic materials produced are still fossil-based. The plastic polymers that dominated in terms of production volume are polyethylene (PE, high and low density), polypropylene (PP), polyethylene terephthalate (PET), polyvinyl chloride (PVC), polystyrene (PS), and polyurethane (PUR) (Bergmann et al. 2015; UNEP 2016; Plastics-Europe 2020).

Jambeck et al. (2015) calculated that 4.8 to 12.7 million metric tons of plastic entered the ocean based on the plastic waste generated in 192 coastal countries in 2010. The quantity available to enter the ocean from land will increase by an order of magnitude by 2025 if no improvements on waste managements are made. One of the major routes for plastic to enter the marine environment on a global scale are rivers transporting plastic originating from mismanaged waste and inadequate wastewater treatment into the ocean (Barnes et al. 2009; Schmidt et al. 2017). Fisheries are a major contributor to plastic pollution in form of lost, abandoned, or discarded fishing gear (Lebreton et al. 2018), and according to an expert survey by Wilcox et al. (2016), fishing gears pose a substantial ecological threat

In the ocean, plastic is largely distributed by ocean currents, but small plastic particles are also susceptible to long-range atmospheric transport (Allen et al. 2019).

When a plastic item has entered the marine environment, several modes for degradation occur, i.e., photodegradation, mechanical abrasion, and biodegradation, which forms smaller and smaller plastic particles. When in the environment, plastic pieces are often categorized to

size in addition to polymer type. In this study the size categories follow the definitions by Barnes et al. (2009), as recommended by Provencher et al. (2017); microplastics (1– 5 mm), mesoplastics (>5–20 mm), macroplastics (>20– 100 mm), and megaplastics (>100 mm).

The negative impact of marine plastic pollution is complex and include several aspects, both economically (i.e., impeded fisheries and clean-up costs) and ecologically. In a new review article by Kühn and van Franeker (2020) at least 914 species were found to be affected by marine debris, of which plastics constituted almost all cases. For marine wildlife, ingestion and entanglement are the most documented impacts (Linnebjerg et al. 2021). In addition, plastic pollution enables a pathway for chemical contaminants to enter the food chain and can potentially cause negative health effects on humans as well (Thompson et al. 2009; Engler 2012).

### **Marine plastic pollution in the Arctic**

*In this paper the Arctic is defined as the Arctic boundary according to the Conservation of Arctic Flora and Fauna (CAFF) group in the Arctic Council (see figure 1).*

The Arctic environment is characterized by its high latitude, cold temperature, and prominent seasonality. The ecosystems in the Arctic are closely connected to sea ice and have short food webs with few trophic interactions from bottom-level species to top-level species (Tirelli et al. 2020).

The Arctic climate is changing faster than anywhere else in the world, and the region is losing its ice and snow, causing the loss of unique habitats and altering marine primary production (IPCC 2019). Despite its remote location, harmful persistent organic pollutants (POPs) are deposited in the Arctic via long-range atmospheric transport, ocean currents, and rivers (AMAP 2018). The Arctic is not spared from plastic contamination either. Reports indicate that the Arctic is not significantly less polluted than more densely populated areas further south. Plastic pollution derives from both local and distant sources, and observations indicates that a higher prevalence of plastic is connected to areas where fishery and ship traffic are high.

The entire size range of plastic particles are found in the Arctic, and microplastics dominate in abundance. Studies have found that the Arctic sea ice serves as a global sink for microplastic and that microplastics are released back into the seawater during the summer melt (Sebille et



al. 2012; Obbard et al. 2014; Reviewed in Halsband and Herzke 2019 and references therein; von Friesen et al. 2020).

Plastic pollution in the Arctic is increasing, and a sixth garbage patch is likely to emerge in the Barents Sea if we continue littering in the same manner as we do today (Seville, England, & Froyland, 2012). The negative impacts from increasing plastic pollution give rise to concern, as the arctic biota are vulnerable to several other environmental threats already, such as climate change and ocean acidification; offshore oil and gas activity; increased shipping traffic which overlap with key seabird areas; commercial fisheries; altered and destroyed habitats; and hazardous chemical pollution (Huntington 2009; Letcher et al. 2010; Humphries 2012; IPCC 2019; Linnebjerg et al. 2021).



Figure 1 Boundaries of the Arctic as defined by the working groups in the Arctic Council (CAFF in yellow). Source: Grid Arendal ( <https://bit.ly/3u76P57> )

## Plastic occurrence in Arctic Seabirds

The Arctic region is a very important area for seabirds, and the Barents Sea is one of the most productive ecosystems in the world, supporting around 4 million pairs of breeding seabirds (Gabrielsen 2009).

Seabirds are long-lived top predators that are intrinsic to the marine ecosystem (González-Bergonzoni et al. 2017). As such, seabirds can be valuable indicators of ecosystem health and marine pollution trends due to their variable position in the marine food web and their feeding ecology (Schreiber and Burger 2002; Mallory et al. 2010; van Franeker et al. 2011). Seabirds are important vectors of nutrients and contaminants from the marine environment to the terrestrial environments through guano (Gabrielsen 2009). Seabird colonies have recently been coined as sinks for marine plastics, as findings of microplastic in fecal precursors (i.e., guano) indicate that the seabirds are vectors for microplastics as well (Provencher et al. 2018; Bourdages et al. 2021; Grant et al. 2021).

### Foraging behavior in seabirds and bioavailability of plastics

The diet of seabirds often consists of pelagic fish, crustaceans or squid, and some species also forage on other seabirds or seabird eggs (Schreiber and Burger 2002; Gabrielsen 2009). The foraging range of seabirds is often distinguished between coastal and pelagic. Seabirds have several modes of foraging, where pursuit diving and surface-feeding are the most common methods. Pursuit diving is when birds dive from the surface and pursues prey underwater; as for surface-feeding, the birds are picking prey from the surface (Schreiber and Burger 2002). The foraging mode of birds are assumed to play a role in the amount of plastic ingested (Ryan 1987; Hallanger and Gabrielsen 2017).

Most seabird species feed in zones where plastic pollution occurs in high density, such as in surface layers along eddies and ocean fronts, making them particularly vulnerable to ingestion when plastic is mistaken for prey or by incidental swallowing (Amélineau et al. 2016; Provencher et al. 2019). Seabirds may also be susceptible to plastic ingestion through trophic transfer from prey (Hammer et al. 2016) or via parental transfer to chicks (Lavers et al. 2014; Acampora et al. 2017b). Surface-feeding birds, like the Procellariiformes (e.g., Northern fulmars (*Fulmarus glacialis*)) and pelicans, are especially susceptible to ingest plastic due to unselective feeding behavior and because plastics tend to float at the surface (Tourinho et al. 2010; Provencher et al. 2014; Poon et al. 2017). However, surface feeders are not the only

seabirds that demonstrate plastic ingestion, as diving seabirds foraging below the surface also display plastic ingestion in the Arctic (Provencher et al. 2010).

The ongoing Atlantification of the Barents Sea has generated a shift in prey availability and feeding behavior in some seabird species (CAFF 2017), which can affect the bioavailability of plastics. Another interesting aspect of plastic ingestion is olfaction, as several seabird species are assumed to rely on their sense of smell to detect food (Schreiber and Burger 2002). Marine plastics can produce the odorant dimethyl sulfide (DMS), which is also an info-chemical cue for prey patches, triggering foraging activity (Savoca et al. 2016).

When seabirds ingest plastic, the plastic particles are usually either regurgitated if possible (Ryan 1987) or retained in the gizzard and mechanically broken down until the birds can excrete the plastic, but very small pieces of microplastics may pass the digestive system without being retained in the gastrointestinal tract (GIT) (Bourdages et al. 2021). However, plastics can lead to deadly obstruction in the gut when the pieces are large (Pierce et al. 2004) or result in sub-lethal effects like damages in the gastrointestinal tract, reduced body condition and impaired growth and development in the affected organism (Lavers et al. 2019; Puskic et al. 2020)

In a study by Robards et al. (1995) they revealed that plastic ingestion rates had increased significantly both in the number of species containing plastic and the frequency of occurrence within species, as well as the mean number of plastic particles in individuals. Trevail et al. (2015) found that plastics ingestion in Northern fulmars had increased over the years and that the results contradicted the established trends of a decrease with latitude and distance from human impacts. The occurrence of plastic in seabirds and the ingestion rate of plastics are expected to continually increase proportionately with more plastic entering the ocean (Wilcox et al. 2015; Baak et al. 2021).

### **Knowledge gaps in plastic exposure in many Arctic seabird species**

The Norwegian population of seabirds alone constitutes 20-25 % of all seabirds breeding in Europe (Anker-Nilssen et al. 2015), but populations in the Atlantic Arctic are declining (CAFF 2017), and several species have been classified as threatened (Christensen-Dalsgaard et al. 2020). The negative population trends in Arctic seabirds portends a significant loss of biodiversity on a global scale (Gaston 2011).

The potential for negative effects from plastic pollution on seabirds on individual and population levels is of great concern, and seabirds are the most studied organism in the Arctic in terms of plastic ingestion (Tirelli et al. 2020). However, we lack knowledge about the occurrence and spatiotemporal trends of plastic ingestion in many of the arctic seabird species, and the available studies are outdated, as approximately half of the published data report plastic ingestion prior to year 2000 (O'Hanlon et al. 2017 b; Baak et al. 2021). In addition, studies often fail to report essential metrics because of a lack of standardized guidelines or protocols (Provencher et al. 2017; O'Hanlon et al. 2017 b; Provencher et al. 2019; Baak et al. 2021).

Currently, the unselective surface-feeding Northern fulmar is the only arctic seabird species (and the only species of any biota on a global scale) for which a published and standardized protocol has been implemented to monitor plastic ingestion. Fulmars are used as a monitoring species to assess plastic pollution in the North Sea for Ecological Quality Objectives (EcoQO) in OSPAR (Oslo/Paris convention for the protection of the Marine Environment of the North-East Atlantic) (van Franeker et al. 2011; Provencher et al. 2017; Baak et al. 2021). Wilcox et al. (2015) suggest that the negative impacts from plastic ingestion may be widespread among species, but plastic ingestion will vary between geographical areas and seabird species depending on their feeding ecology, age, and time of the year (Van Franeker and Meijboom 2002). Dehnhard et al. (2019) concluded that it is necessary to provide baseline information across Arctic seabird species to be able to identify which species and populations are most suitable for monitoring, and have suggested Northern fulmar, European shag (*Phalacrocorax aristotelis*), Great skua (*Catharacta skua*), Great black-backed gull (*Larus marinus*), Black-legged kittiwake (*Rissa tridactyla*), Common guillemot (*Uria aalge*), Brünnich's guillemot (*Uria lomvia*), and Atlantic puffin (*fratercula arctica*) as interesting species of Norwegian seabirds to monitor for plastic ingestion. The Northern fulmar, Brünnich's guillemot, and Black-legged kittiwake are suggested by the Conservation of Arctic Flora and Fauna (CAFF) group in the Arctic Council as indicator species of plastic pollution in the Arctic (Baak et al. 2021).

In recent years, research on plastic pollution in seabirds have been complemented by polymer identification and the related potential for chemical exposure, as different plastic types will contain different chemicals and have different behavioral properties (Tanaka et al. 2013; Amélineau et al. 2016; Avery-Gomm et al. 2016; Tanaka et al. 2019; Kühn et al. 2021). One method of polymer identification is Fourier-transform infrared spectroscopy (FTIR), and the









assessment of polymer composition of plastics ingested by seabirds can be used to evaluate potential toxic effects from a specific polymer (Lithner et al. 2011; Kühn et al. 2021).

## Study species

This study investigates plastic occurrence six arctic seabird species breeding in Norwegian territory (table 1).

*Table 1 The six seabird species investigated in this study and their distribution, ecological niche, and population status in mainland Norway (N) and on Svalbard (S) (IUCN red list: least concern (LC), near threatened (NT), vulnerable (VU), endangered (EN) or critically endangered (CR)).*

	Species	Scientific name	Distribution	Ecological niche	Population status (mainland Norway/Svalbard)
	Atlantic puffin	<i>Fratercula arctica</i>	Arctic & temperate	Pelagic, diving	VU (N) LC (S)
	Common guillemot	<i>Uria aalge</i>	Arctic & temperate	Pelagic, diving	CR (N) VU (S)
	Glaucous gull	<i>Larus hyperboreus</i>	Arctic & temperate	Pelagic/coastal, surface-feeding/opportunistic	NT (S)
	Razorbill	<i>Alca torda</i>	Arctic & temperate	Pelagic diving	EN (N) EN (S)
	Great cormorant	<i>Phalacrocorax carbo</i>	Arctic & temperate	Coastal (benthic), diving	LC (N)
	European shag	<i>Phalacrocorax aristotelis</i>	Arctic & temperate	Coastal (benthic), diving	LC (N)

## Atlantic puffin (*Fratercula arctica*)



Figure 2 Atlantic puffin on cliff. Photo: Geir W. Gabrielsen

The Atlantic puffin (also referred to as puffin) is a medium-sized pelagic seabird in the Alcidae family under the Charadriiformes order, and it is the most common seabird in Norway (Gjershaug and Lorentsen 2020). Puffins breed on both sides of the North Atlantic. In Europe the large majority of the population breeds along the Atlantic Ocean and the Norwegian Ocean, where the Norwegian population shares the same distribution as the Common guillemot (Lydersen 2006; Gjershaug and Lorentsen 2020). Atlantic puffins forage in coastal and offshore areas during the summer (Shoji et al., 2015 in Dehnhard et al. (2019), whereas outside the breeding season, the birds are fully pelagically distributed (Lydersen 2006).

In regards to size, puffins can reach 28-34 cm and 300-600 grams, and the sexes are similar (Lydersen 2006). They reach sexual maturity at 5-6 years, and their maximum life expectancy is 41 years (Gjershaug and Lorentsen 2020).

Puffins are pursuit divers and catch small schooling fish like sand eels (*Ammodytes* spp.) and young herring (*Clupea harengus*), and sometimes crustaceans, squid and polychaete worms (Nereidae). They typically feed in the surface layers down to 15 meters but are known capable of diving to 70 m (Lydersen 2006; Gjershaug and Lorentsen 2020; Fayet et al.).

To the best of our knowledge, there has been only one previous study documenting plastics ingestion in Atlantic puffin in the European Arctic (Berland, 1971 in O'Hanlon et al. 2017 a) and one in the Canadian Arctic in 2004 (Provencher et al. 2014).

### **Common guillemot (*Uria aalge*)**



Figure 3 Common guillemot colony. Photo: Svein-Håkon Lorentsen (NINA)

The Common guillemot is the largest of the existing auk species and belongs to the Alcidae family under the Charadriiformes order. The Common guillemot has a circumpolar low arctic-boreal distribution, and it is one of the most abundant seabirds in temperate and sub-arctic areas of the northern hemisphere (Anker-Nilssen et al. 2000; Lydersen 2006). The Norwegian breeding populations are found on the mainland from Rogaland to Finnmark and on Jan Mayen and Bear Island (Lydersen 2006). It is not a regular migratory bird, although it disperses in winter and outside breeding season, it appears both inshore and further out to sea (Lydersen 2006; Fauchald et al. 2019).

The Common guillemot is a diving seabird with a size of 38-43 cm and 900-1300 grams. The sexes are similar, and the birds breed for the first time at 4-5 years (Anker-Nilssen et al. 2000; Gjershaug and Lorentsen 2020).

These pursuit diving auks are food specialists (predominantly piscivorous) and catch small schooling fish (max 20 cm long) like capelin (*Mallotus villosus*), herring, and gadoids (*Gadidae* spp.), but may also feed on squid. They generally feed at depths of 20-50 m (Anker-Nilssen et al. 2000; Lydersen 2006; Gjershaug and Lorentsen 2020).

To the best of our knowledge, there are only two previous reports of plastic ingestion from Canadian waters (reviewed in Provencher et al. 2015 and references therein), and no previous studies reporting or investigating ingested plastic in Common guillemots in the European Arctic (O'Hanlon et al. 2017 a; O'Hanlon et al. 2017 b). However, it was an incidental discovery of plastic in a few birds examined after a mass mortality event in southern parts of Norway in November 2020 ( <https://bit.ly/33Jer2J> ) .

### **Razorbill (*Alca torda*)**



*Figure 4 Razorbill with sandeel in its beak. Photo: by Geir Systad (NINA)*

The Razorbill is an auk in the Alcidae family under the Charadriiformes order and is distributed in temperate, boreal, and low-arctic coastal regions on both sides of the North Atlantic (Anker-Nilssen et al. 2000). In Norway, the Razorbill breeds together with other auk species on bird cliffs along the coast of mainland Norway and on Jan Mayen and Bear Island (Lydersen 2006; Gjershaug and Lorentsen 2020).

The Razorbill is slightly smaller than the Common guillemot, with its 600-900 grams and approximately 38 cm (Lydersen 2006). The sexes are similar, and they reach sexual maturity at 4-5 years (Gjershaug and Lorentsen 2020).

The alcids are great divers, and the Razorbill catch pelagic prey, like small fish (e.g., sand eel, capelin, herring, and some gadoids), planktonic crustaceans, and polychaetas (Gjershaug and Lorentsen 2020), normally in the depth range of 25-30 meters. Throughout the year, the Razorbill forage both in coastal and offshore environments (Dehnhard et al. 2019) but are fully pelagic outside breeding season.



To the best of our knowledge, there have been no previous published studies reporting plastic ingestion in Razorbills in the arctic (Provencher et al. 2014; Provencher et al. 2015; O'Hanlon et al. 2017 a).

### **Great cormorant (*Phalacrocorax carbo carbo*)**



*Figure 5 Great cormorant on nest. Photo: Tycho Anker-Nilssen (NINA)*

The Great cormorant is a pursuit diving coastal seabird and one of the largest cormorant species in the Phalacrocoracidae family under the Pelecaniformes order (Anker-Nilssen et al. 2000). Great cormorants can be found on all continents except for South America; this study has investigated the subspecies *carbo* (also referred to as “Atlantic great cormorant”), which is distributed in the coastal areas of the North Atlantic where main breeding areas are Norway, British Isles and Iceland (Anker-Nilssen et al. 2000; Gjershaug and Lorentsen 2020). The Norwegian population of Atlantic Great cormorant breeds in the three northern counties of Norway (Gjershaug and Lorentsen 2020).

The birds weigh 1500-3600 grams, and the females are smaller than the males. They breed for the first time when they are between 2-4 years of age, and the oldest known specimen lived to be over 21 years (Gjershaug and Lorentsen 2020).

The Atlantic Great cormorant feeds almost exclusively on medium-sized fish, preferably gadoids, sand eel, and capelin, caught near the bottom in shallow waters (<10 m deep), whereas studies on food choice in Barents indicate that the eastern Finnmark population feed

on capelin in addition and that the birds may also feed on shoaling fish in open waters (Barrett et al., 1990 in Anker-Nilssen et al. 2000).

To the best of our knowledge, there are no published studies reporting ingested plastic in Great cormorants in the Arctic (O'Hanlon et al. 2017 a).

### **European shag (*Phalacrocorax aristotelis*)**



*Figure 6 European shag in flight. Photo: Tycho Anker-Nilssen*

The European shag is a medium-sized cormorant in the Phalacrocoracidae family under the Pelecaniformes order, and it is a coastal bound seabird. The subspecies investigated in this study breed along the coasts of the Atlantic Ocean. Regarding the Norwegian population, western part of Finnmark is the most densely populated area (Gjershaug and Lorentsen 2020).

They are smaller than the Great cormorant, and adults weigh 1800-2500 grams. The birds reach sexual maturity at 2-3 years, and maximum life expectancy is 30 years (Gjershaug and Lorentsen 2020).

They are rarely seen inside the fjords and usually feed in deeper waters than the Great cormorant, where they catch pelagic fish close to the bottom in the depth range of 20-40 meters (Anker-Nilssen et al. 2000; Gjershaug and Lorentsen 2020). In the Barents area, sand eel and young gadoids predominate the diet, and the European shag is considered a specialized piscivore (Anker-Nilssen et al. 2000).

To the best of our knowledge there are no previous reports of ingested plastic in European shags in the Arctic (O'Hanlon et al. 2017 a).

### **Glaucous gull (*Larus hyperboreus*)**



Figure 7 Glaucous gulls in Svalbard. Photo: Geir W. Gabrielsen

The Glaucous gull is a coastal-bound surface feeder that belongs to the Laridae family in the Charadriiformes order, and it has a circumpolar, high arctic distribution. There are four subspecies, where the Svalbard population, which is investigated in this study, belongs to the subspecies *L.h. gunnerus*. The Svalbard population spend the winter dispersed in the North Atlantic

It is one of the largest gulls breeding in the Arctic, with lengths varying between 65-78 cm and a normal weight of 1250-2700 grams (Lydersen 2006). The sexes have a similar appearance, but the males are somewhat larger than the females. The birds reach sexual maturity at the age of 4-5 years, and the maximum life expectancy for a Glaucous gull is 19 years (Lydersen 2006; Gjershaug and Lorentsen 2020). (Lydersen 2006).

The Glaucous gull is a generalist predator that exploits many different types of prey like fish, mollusks, echinoderms, crustacea, bird eggs, chicks, seeds, berries, offal, and food scraps from humans (Anker-Nilssen et al. 2000; Lydersen 2006; Gjershaug and Lorentsen 2020). The Glaucous gull is found in the uppermost part of the food chain, and in Svalbard, they occupy the same ecological niche as birds of prey further south (Anker-Nilssen et al. 2000;

Lydersen 2006). Consequently, their diet often consists of seabird chicks, seabird eggs, and seal blubber leading to high level of pollutants in the Glaucous gulls (Sagerup et al. 2009; Erikstad et al. 2013).

There are currently no published studies regarding polymer composition of plastic found in stomachs of Glaucous gulls to the best of our knowledge. There have been three previous studies examining stomach content in Glaucous gulls, two from Svalbard (Mehlum and Giertz 1984; Lydersen et al. 1989) and one from Frans Josef Land (Weslawski et al. 1994), with no findings of plastic. Therefore, this study may provide new quantitative and qualitative data on plastic loading and polymer type which can establish a baseline for future research and monitoring of Glaucous gulls.

### Aim of study

The aims of this study are (1) to provide information on plastic occurrence in Arctic seabirds by investigating six species of seabirds that have breeding populations in the Arctic, (2) to characterize the findings through visual inspection and FTIR, and (3) to harmonize methods and assess the feasibility of the set of recommendations on collection, processing method and reporting made by Provencher et al. (2019).

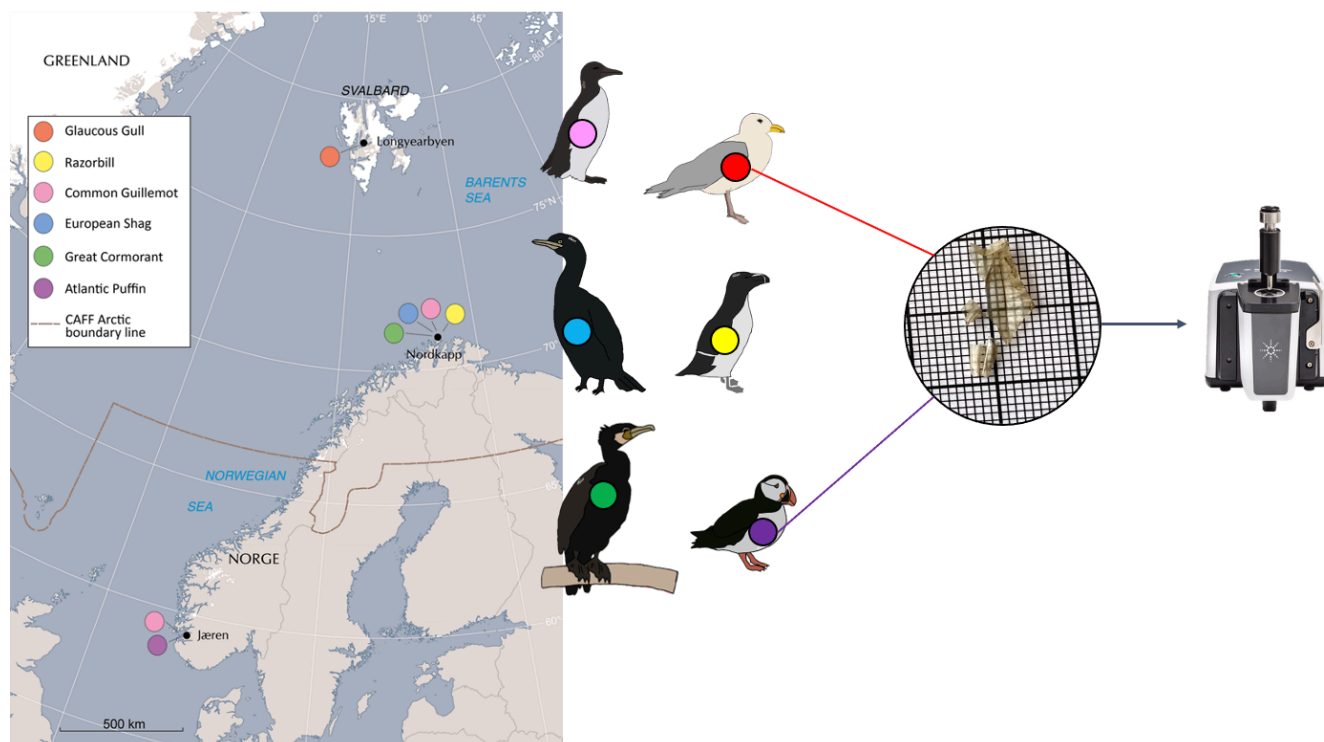


Figure 8 Graphic illustration of the study



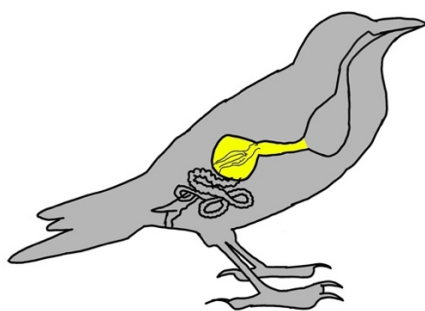
## Chapter 2

### Material and method

*This study follows the recommendations by Provencher et al. (2017) for reporting plastic ingestion in marine megafauna combined with the protocol for stomach procedure from the fulmar EcoQO-methodology (OSPAR 2015). This will facilitate the comparability for future studies and support the process of enforcing standard guidelines for reporting plastic ingestion.*

#### Seabird sampling

This study investigated six species of seabirds: Atlantic puffin, Common guillemot, Razorbill, Great cormorant, European shag, and Glaucous gull. The sampling method was necropsy of the gastrointestinal tract to examine the proventriculus and gizzard. The individuals collected in the field were carcasses from fisheries bycatch, beached birds, or birds from specific hunting provided by the Norwegian Institute for Nature Research (NINA; Trondheim; all the stomachs except for Glaucous gull) and the Norwegian Polar Institute (NPI; Glaucous Gull stomachs). Information about the age, sex, and body weight of the birds are listed in table 3. NPI and NINA dissected the birds and only the stomachs (proventriculus and gizzard) were processed and analyzed for plastic in this particular study (by Stine Benjaminsen).



*Figure 9 Illustration of the avian digestive system (GIT) with proventriculus and gizzard highlighted in yellow.*

Seabirds have two stomachs; the first is the proventriculus which is a glandular and elastic stomach where food is stored and mixed with digestive stomach fluids. From the proventriculus, the food is pushed into the second stomach, the gizzard, which has thick muscular walls that grind hard food items into small enough pieces to be passed on further down the digestive system (see Figure 2) (Kühn and van Franeker 2012).

The birds provided by NINA were measured and dissected following the standardized protocol described in van Franeker (2004). The Razorbills (n=10), Great cormorants (n=8), and the shags (n=9) were collected at Nordkapp (71°10'N, 25°47'E), the northernmost point in Norway, in May/June 2012 as bycatch from a lumpsucker fishery. The Common guillemots (n=5) and the Atlantic puffins (n=17) were beached birds collected south of the Arctic boundary, as part of the EcoQO-project Beached Bird Survey (BBS) at Jæren (58°71'N, 5°71'E), Rogaland County, Norway in February/March in 2019 and 2016, respectively. The puffins were part of a mass mortality event in SW Norway during a winter storm, and the birds had presumably starved to death (Anker-Nilssen et al. 2018). The Glaucous gulls (provided by NPI) were collected in Longyearbyen (78°23'N, 15°59'E) in May 2018 as part of a project registered in the Research in Svalbard (RiS) database (RIS 10654). The birds were shot with a shotgun and decapitated, then blood was collected, and the birds dissected in the field. The stomachs were wrapped in aluminum foil and individually bagged. The below zero temperatures allowed the samples to quickly cool down before they were stored in a -20°C freezer at the end of the sampling day. In this study, 21 stomachs of Glaucous gulls were analyzed for plastic.

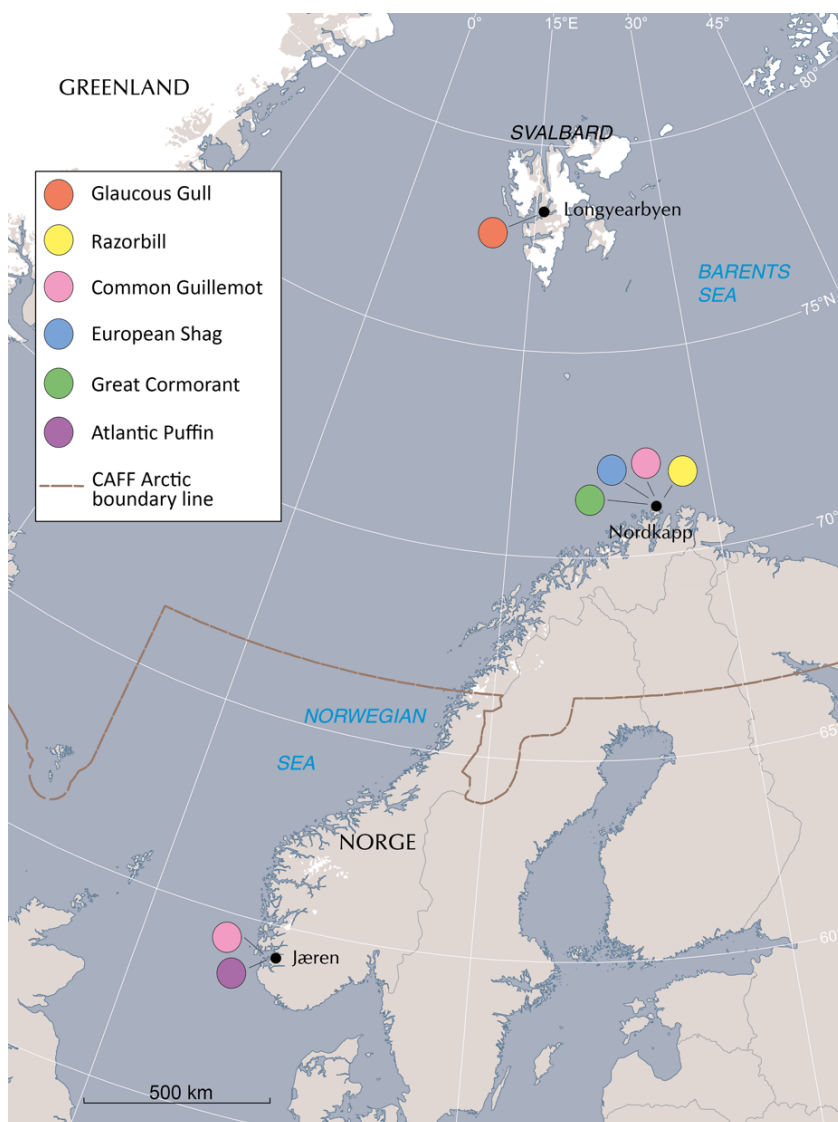


Figure 10 Map (courtesy of The Norwegian Polar Institute) of the sampling area for the seabird species investigated in this study.

## **Stomach dissection and plastic analysis**

All the stomachs were analyzed at NPI (Tromsø). The Glaucous gull stomachs were analyzed in November 2019, and the other birds were analyzed in December 2020. In 2020, we included a 0.5 mm sieve in addition to the 1 mm sieve and weighed the stomachs before the dissection.

Each stomach was thawed a couple of hours before the analysis and closed with a zip tie with the proventriculus and gizzard connected. First, the stomachs were opened up by cutting the zip tie and gently cutting along the stomach with scissors. The stomach content was then emptied over a sieve, and the stomach gently rinsed using cold fresh tap water to collect all the content. The content of the proventriculus and the gizzard were analyzed together. The stomach walls were rinsed thoroughly to ensure that no particles were overlooked. The Glaucous Gull stomachs were rinsed over a 1 mm sieve and then re-frozen, and the stomachs from the other species were rinsed over a 1 mm sieve with a 0.5 mm sieve underneath and then discarded. The plastic pieces were identified by visual examination of the sieve, and in addition, a Leica M60 stereomicroscope (magnification range 6.3x – 40x) was used when pieces were either very small or when the plastic nature of the piece was questioned. All the particles assumed to be plastic were collected, and the diet material from the birds provided by NINA was stored for future analysis. The plastic pieces were counted, dried, and weighed separately on an analytic scale (Sartorius and Acculab, Sartorius Group) to the nearest 0.0001 g, and the length was measured in mm with a digital caliper (including two decimals). The plastic pieces were categorized into the size group megaplastics (>100 mm), macroplastics (>20–100 mm), mesoplastics (>5–20 mm), or microplastics (1–5 mm). The type of the plastic was determined by visual inspection by trained experts experienced with beach cleanups and classified as industrial plastics (nurdles) or user plastics (sheet, thread, foam, fragment, and other). The plastic pieces were given color assignments and grouped into eight broad color groups following the guidelines by Provencher et al. (2017); off/white-clear; grey-silver; black; blue-purple; green; orange-brown; red-pink; yellow. Finally, the polymer identity was determined by FT-IR analysis (explained in the section below).

## Fourier-Transform infrared (FT-IR) spectroscopy analysis




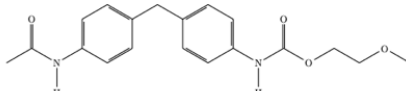
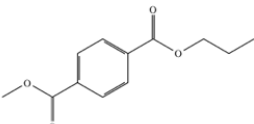
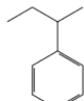
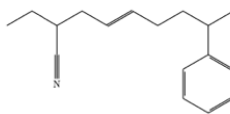
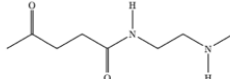
The FT-IR analysis was conducted in January 2021 at NPI in collaboration with the Norwegian Institute for Air Research (NILU) at the Fram center in Tromsø, Norway.

### Theoretical background

The Fourier transform infrared spectrometer is an instrument that rapidly provides the absorption bands for a given compound.

All molecules are built up of atoms, and the bonds connecting the atoms can absorb energy from an infrared beam when applied. Different molecules have unique patterns of absorption, which can be utilized in sample identification by collecting a spectrum with the absorption bands of a sample (Pavia et al. 2001).

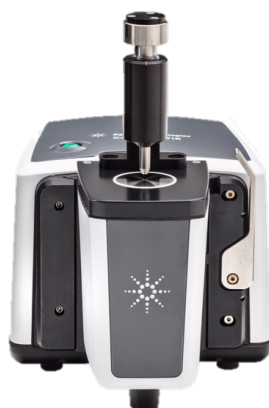
Table 2 Visual display of the monomers in main plastic polymers and their common area of use (Plastic Europe, 2018)

Name	Monomer	Common uses
Polypropylene		Food packaging, bags, automobile parts
Polyethylene		Food packaging, bottles, toys, containers
Polyvinyl chloride		Building materials, hoses, cable insulation
Polyurethane		Building insulation, pillows and mattresses
Polyethylene terephthalate		Bottles
Polystyrene		Packaging, cups
Acrylonitrile butadiene styrene		Medical devices, LEGO, keyboard caps
Polyamide		Nylons

The standard FT-IR instrument utilizes an interferometer generated by using a beam splitter to separate the source energy into two perpendicular beams. Computer-interfaced FT-IR and the

Diamond attenuated total reflectance (ATR) sampling accessory uses a single beam method and is most suitable for higher absorbing samples, like polymers (Technologies 2011).

To obtain the spectrum of the sample with Computer-interfaced FT-IR, one first collects an interferogram of the “background” and implement the mathematical technique Fourier transform to make a plot that can be read like a typical infrared spectrum. Then the sample is placed into the beam, and the detector of the interferogram signal registers the types of wavelengths absorbed by the sample. The spectrum with the absorption band of the sample is created as a result of the Fourier transform on the sample’s interferogram (Pavia et al. 2015). The computer software then removes the background spectrum from the sample spectrum and plot the infrared spectrum of the sample on the computer screen. The spectrum obtained can then be compared to a reference library of infrared spectra and identified if there is a match in the reference library.



*Figure 11 Cary 630 FTIR Spectrometer with ATR sampling module, screenshot from: [www.agilent.com](http://www.agilent.com)*

The use of ATR accessory is the recommended method for analyzing solid sample compounds. The ATR method is based on the physical properties of light and the difference in the index of refraction of two materials (the sample and the crystal). The beam of infrared radiation travels back and forth off the crystal surface and penetrates the sample with a small and specific depth before it is reflected into the detector. This eliminates the necessity of both sample preparation and the need for a clear sample. The key to obtaining good measurements with the ATR is to ensure good contact between the sample and the crystal (Technologies 2011).

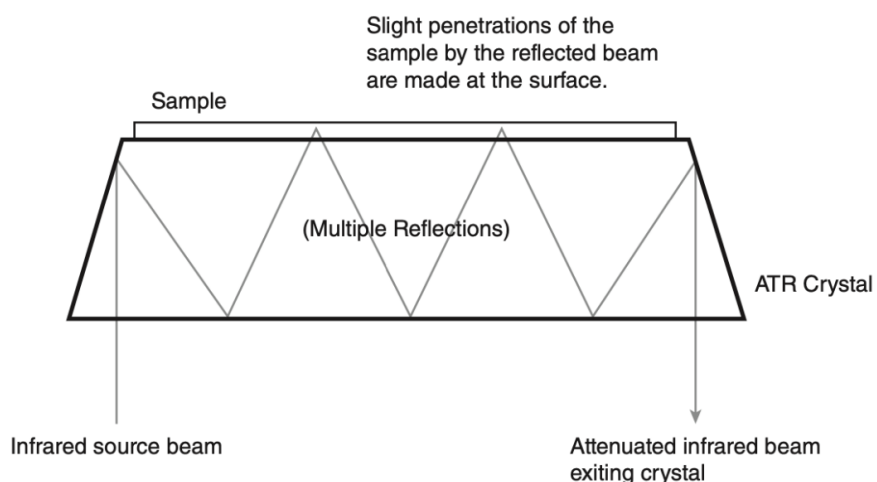


Figure 12 Description of the ATR sampling accessory (Pavia et al., 2015, p. 26)

### ATR FT-IR Instrument details and practical procedure

A Cary 630 FTIR spectrometer with Diamond Attenuated total reflectance (ATR) accessory (Agilent, CA, USA) was used to collect spectrums from  $4000\text{cm}^{-1}$  to  $650\text{cm}^{-1}$ . The resolution was set at  $8\text{cm}^{-1}$ .

The ATR diamond crystal was cleaned, and a background scan was performed between each sample. The pieces were placed on the crystal covering the entire surface area of the crystal and clamped down to ensure good contact between the crystal and the sample. The absorption bands of the sample were recorded and compared to the reference spectrums in the demo library of the computer by a similarity search algorithm. The match between the sample spectrum and the reference spectrum is given a score in the Hit Quality Index (HQI) between zero and one, where one is a perfect match.

Some pieces of sheet needed removal of biofilm by scraping with a sharp object, and other pieces, most often fragments and pellets, were cut with a knife to provide a thin and clean sample for a successful measurement if scraping was insufficient. Analyzing the end of threads gave a better reading than using the middle parts. All the 18 plastic pieces found in the seabird stomachs were analyzed with FT-IR spectroscopy, and the polymers were identified as a given plastic polymer if the HQI was  $\geq 0.7$ .

## Quality assurance

The stomachs were dissected using equipment in glass or metal to avoid contamination. All pieces of plastic were dried and weighed separately on a Sartorius balance (0.0001g accuracy). Undergoing the FT-IR, every sample was analyzed with the desired background established, and several analyzes were carried out on the same sample, if necessary, to obtain the accepted match of an HQI  $\geq 0.7$ . The different density types of polyethylene (linear low density, low density, and high density) could not be distinguished to avoid mislabeling due to the indistinct signature peaks between the three polyethylene types.

## Data analysis and statistics

All biometric and other numeric data were stored in Microsoft Excel, and the statistics were carried out in R version 1.4.1103 (R Development Core Team 2021) using R Studio. In R Studio, all zeros were included when statistics were run. The package `pastecs` (Grosjean and Ibanez 2018) with `stat.desc` were used to obtain the desired metrics. The frequency of occurrence with a 95% confidence interval was calculated for each species using the “Jeffreys” interval, which was calculated using the Epitool online calculator (see: <https://epitools.ausvet.com.au/ciproportion> ). Due to small sample size, no further tests were conducted. A tabular form was decided to be the most suitable and appropriate way to present the results.







## Chapter 3

### Results

#### Birds in the study

There were six different species of arctic seabirds included in this study, and the general information about the species is presented in table 1. A summary of the birds investigated in our study is presented below in table 3.

Table 3 Summary of the seabirds used in this study, showing sampling area, sample size(n), sex(male/female), age category, and mean weight in grams  $\pm$  standard deviation (SD).

	Species	Sample size (n)	Sex (male/female)	Age category	Weight (g) Mean $\pm$ SD
Southwest Norway	 Atlantic Puffin	17	71% m 29% f	13 adults, 3 immatures, 1 juvenile	251 $\pm$ 114.9 *
	 Common guillemot	5	40% m 60% f	1 adult, 1 immature, 3 juveniles	865 $\pm$ 205,9*
Northern Norway	 Razorbill	9	33% m 67% f	8 adults, 1 immature	930.4 $\pm$ 50.3
	 Great Cormorant	8	50% m 50% f	All adults	3265 $\pm$ 436.3
	 European Shag	8	75% m 25% f	5 adults, 3 immatures	2032.4 $\pm$ 183.6
Svalbard	 Glaucous gull	21	38% m 62% f	All adults	1688.7 $\pm$ 352

\* The birds were wet and/or sandy

#### Plastic found in the stomachs

Among the 17 stomachs of puffins, ten were found to contain plastic corresponding to a medium-high frequency of occurrence (FO) of 58.8% with a 95% confidence interval (Jeffreys interval) (CI) of 35.6-79.3%. Among the 21 Glaucous gull stomachs analyzed, three stomachs (gizzard) contained plastic with a low FO of 14.3% and a CI of 4.2-33.4%. The



stomachs of Razorbill (n=10), Common guillemot (n=5), Great cormorant (n=8), and European shag (n=9) showed no occurrence of plastic that could be detected with the method used in this study.

Table 4 gives the descriptive statistics for the mass and number, and table 5 gives the descriptive statistics for the length of plastic pieces found in the stomachs of Atlantic puffin and Glaucous gull. The metrics are given for the total plastic burden, for the industrial plastic (nurdles) and for the user plastics (sheet, thread, foam, fragment and other). Each individual piece of plastic is described in Appendix A.

Table 4 Mass (mg) and number of pieces of plastics found in stomachs of Atlantic puffins (n=17 individuals), Glaucous gull (n=21 individuals), (Razorbill, Common guillemot, Great cormorant and European shag had no occurrence of plastic). Summary statistics are given as mean  $\pm$  standard deviation (SD), standard error of the mean (SEM), median, and range (min-max). lack of values due to inadequate sample size are shown as not available (n.a)





		Mass (mg)				Number of pieces			
		Mean $\pm$ SD	SEM	Median	Range	Mean $\pm$ SD	SEM	Median	Range
 Atlantic puffin	Total plastics	9.95 $\pm$ 12.54	3.04	2.20	0-34.90	0.88 $\pm$ 0.99	0.24	1	0-3
	Industrial nurdles	2.73 $\pm$ 7.73	1.87	0	9.90	0.18 $\pm$ 0.53	0.13	0	0-2
	User plastic:								
	Sheet	2.02 $\pm$ 4.97	1.20	0	18.50	0.35 $\pm$ 0.70	0.17	0	0-2
	Thread	0.22 $\pm$ 0.65	0.15	0	1	0.12 $\pm$ 0.33	0.08	0	0-1
	Foam	0				0			
	Hard fragment	4.97 $\pm$ 11.10	2.69	0	31.90	0.23 $\pm$ 0.44	0.10	0	0-1
	Other	0				0			
 Glaucous gull	Total plastics	7.55 $\pm$ 31.30	6.83	0	0-143.5	0.14 $\pm$ 0.35	0.08	0	0-1
	Industrial nurdles	0				0			
	User plastic:								
	Sheet	0				0			
	Thread	0				0			
	Foam	0.07 $\pm$ 0.30	0.07	n.a	n.a	0.048 $\pm$ 0.22	0.048	n.a	n.a
	Hard fragment	6.83 $\pm$ 31.31	6.83	n.a	n.a	0.048 $\pm$ 0.22	0.048	n.a	n.a
	Other	0.65 $\pm$ 2.97	0.65	n.a	n.a	0.048 $\pm$ 0.22	0.048	n.a	n.a
Razorbill	Total plastics	0				0			
Common guillemot	Total plastics	0				0			
Great cormorant	Total plastics	0				0			
European shag	Total plastics	0				0			

Table 5 Length (mm), mean with standard deviation (SD), standard error of mean (SEM), median and range (min-max) of the plastic pieces found in the Atlantic puffins and Glaucous gulls.

		Length of plastic pieces (mm)			
		Mean±SD	SEM	Median	Range
 Atlantic puffin	Total plastics	9.75±7.93	2.05	6.36	3.38-29.05
	Industrial nurdle	3.94±0.50	0.29	4.12	3.38-4.33
	User plastics:				
	Sheet	9.51±5.69	2.32	8.22	4.31-20.43
	Thread	26.02±4.28	3.02	26.02	23-29.05
	Foam	-	-	-	-
	Hard fragment	6.33±2.67	1.38	5.37	4.43-10.17
Other	-	-	-	-	
 Glaucous gull	Total plastics	10.21±6.60	3.81	11.18	3.17-16.28
	Industrial nurdle	-	-	-	-
	User plastics:				
	Sheet	-	-	-	-
	Thread	-	-	-	-
	Foam	3.17	-	3.17	-
	Hard fragment	16.28	-	16.28	-
Other	11.18	-	11.18	-	

### Plastic type

The Atlantic puffin stomachs contained 15 pieces of plastic, of which 12 pieces were user plastic, and three pieces were industrial plastic nurdles. The user plastic comprised six pieces of sheet, two pieces of thread, and four pieces of fragment. The Glaucous gull stomachs contained in total three pieces of user plastic and zero pieces of industrial plastic. The user plastic comprised one piece of foam, one hard fragment, and one piece of “other” (a swift tack for price tags).

### Polymer identity, color, and size

The three types of plastic polymers that were identified in the Atlantic puffins and Glaucous gulls combined were polyethylene (PE), polypropylene (PP), and polystyrene (PS). There was one piece that remained unidentified and therefore labeled as “other”. The plastic pieces were visually categorized as off-white/white-clear, black and green. The sizes of the plastic pieces

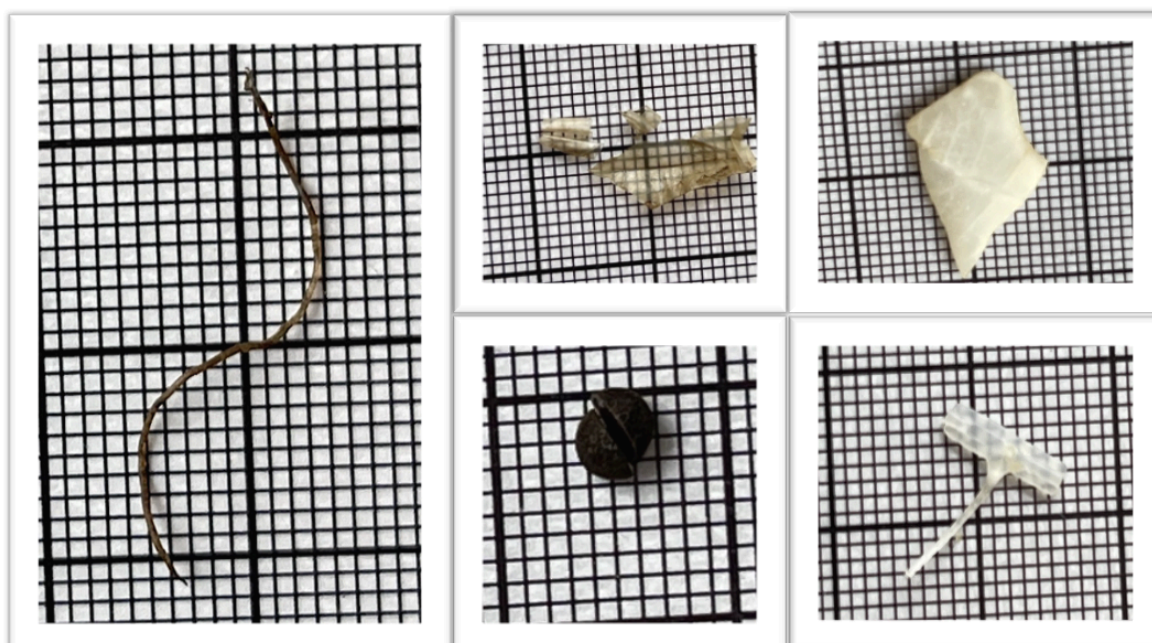
were in the classes macroplastic, mesoplastic, and microplastic, and the length of the plastic pieces ranged from 3.17 mm to 29.05 mm.

### Atlantic Puffin

There were six pieces of PP in the puffin stomachs. All were in the color off-white/white-clear and there were two pieces of macroplastic, two pieces of mesoplastic, and two pieces of microplastic. There were eight pieces of PE which included one piece of green mesoplastic, three black pieces (two pieces of microplastic and one piece of mesoplastic), and four off-white/white-clear pieces (one piece of macroplastic, two pieces of mesoplastic, and one piece of microplastic). In addition, there was one black piece of microplastic in the polymer category “other”.

### Glaucous gull

There were two mesoplastic pieces of off-white/white-clear PP and one microplastic piece of off-white/white-clear PS in the Glaucous gull stomachs.



*Figure 13 A selection of plastics found in the seabird stomachs photographed on millimeter paper. First picture from left: PE thread (puffin). From the top left corner: PP sheet (puffin and hard fragment (Glaucous gull). From bottom left corner: industrial nurdle (puffin) and swift tack (Glaucous gull). All pictures were taken after the FTIR-analysis and the plastics have been cut and/or scraped (see table 5 for correct lengths).*

## Chapter 4

### Discussion

#### Plastic occurrence

The main aim of this study was to provide information on plastic occurrence in six arctic seabird species. Until now, there has been little to no knowledge about the presence of plastic in these species. This study found a medium-high frequency of occurrence of plastic in the stomachs of Atlantic puffins and a low frequency of occurrence in Glaucous gulls. The stomachs of Razorbills, Great cormorants, European shags, and Common guillemots showed no occurrence of plastic.

**Plastic in Atlantic puffins** was below the EcoQO-target for Northern fulmars (less than 10% of fulmars should have 0.1 g plastic or more in their stomach) set by OSPAR (2008). This is the second study where plastic has been detected in the stomach of Atlantic puffins in Norway: one previous study was conducted outside Hordaland back in 1970 (Berland 1971). There have been several reports of plastic ingestion in puffins in the North-East (NE) Atlantic both recently and in the past, indicating that puffins are one of the auk species which are at risk for plastic ingestion (Parslow and Jefferies 1972; Blake 1984; Harris and Wanless 1994; Acampora et al. 2016). The puffins feed in the seawater's upper layers down to 15 meters, possibly making them more prone to ingest plastic debris than other deep diving seabird species. Plastic incorporation in puffin nests has also been observed in puffin colonies at Svalbard (Sebastien Descamps, personal communication, April 2021), strengthening the arguments that puffins are deliberately obtaining plastic from the marine environment.

**Plastic in Glaucous gulls** was above the EcoQO-target for Northern fulmar (sample sizes will be further discussed in the method section). This is, to the best of our knowledge, the first study reporting plastic occurrence in Glaucous gulls in the Arctic. There has been one previous report of a single particle assumed to be plastic in one Glaucous gull in Alaska (Day 1980), but the particle was lost before it could be thoroughly examined.

There are relatively few studies on plastic pollution in gulls (Laridae) (Wilcox et al. 2015; Seif et al. 2018) to compare with the results in the present study. Previous studies on Arctic

gull species suggested that gulls are at low risk for plastic ingestion in the Arctic, with the highest average FO ever observed in an arctic gull being 10% with an average sample size >40 (Herring gull, *Larus argentatus*) (Baak et al. 2021). In our study, the FO in the Glaucous gulls was slightly higher, but the sample size lower (<40) (further discussed in the sample-size section of the discussion). A study by Seif et al. (2018), investigating 41 birds from three different gull species (Herring gulls (*Larus smithsonianus*), Great Black-backed gulls (*Larus marinus*), and Iceland gulls (*Larus glaucooides*)) collected at a landfill in Newfoundland, showed a range of FO of 61-100%. Despite collected in the high Arctic, the Glaucous gulls were culled in near proximity to human settlements, including an open landfill and were therefore expected to have a high FO.

There is now only one open landfill in Longyearbyen (in a closing process), but there have been several open landfills in the past (Kjersti O. Ingerø, technical manager in Longyearbyen local council, personal communication, April 2021). Data on plastic ingestion prior to the closing of the previous landfills would have been desirable but are lacking. The availability of natural food items for the Glaucous gulls is high on Svalbard (Geir W. Gabrielsen, personal communication, April 2021) and may also have contributed to the low occurrence of plastic debris in their stomachs.

Another important feature of gulls is their high ability to regurgitate indigestible food items, making them capable of ridding themselves of plastic and preventing plastics from accumulating in the GIT (Ryan 1987). Indeed, the methodology used in the present study highlighted a wide and flexible passage (pyloric sphincter) between the proventriculus and gizzard of Glaucous gull's stomach, which may further facilitate plastic removal by regurgitation.

The plastic debris that is ingested may be different than what is accumulated. Therefore, it would be interesting to investigate regurgitated boluses from Glaucous gulls from the same area to compare plastic loading in stomachs versus boluses, as this shows that they ingest more plastic than what can be observed in their stomachs.

### **Interspecies comparison between Atlantic puffins and Glaucous gulls**

Both gulls (Laridae) and auks (Alcidae) are known to have lower rates of plastic ingestion than Procellariiformes, such as the Northern fulmar (Baak et al. 2021). Our findings are in accordance with this as the FO was much lower than what has been previously reported for

the Procellariiformes. Nevertheless, O'Hanlon et al. (2017 a) found that the auks, which is a group of diving seabird species had the second highest FO after the Procellariiformes. In another study from Brazil by Tavares et al. (2017) indicated that diving species feeding at intermediate and deep depths had a higher FO than surface feeders in the same area.

The puffins and Glaucous gulls were not collected in the same area, but our findings are in accordance with those of O'Hanlon et al., 2017, as the puffins showed the highest FO among the birds containing plastic in this study.

### **Common guillemots, Razorbills, Great cormorants and European shags**

To date, there have been no previous studies reporting plastic ingestion in the Arctic in Common guillemots, Razorbills, Great cormorant, or European shag (O'Hanlon et al. 2017 a). The lack of plastic in their stomach may be due to living at high latitudes and with long distance to densely populated areas. However, four out of five Common guillemots were sampled in southern Norway, south of the Arctic boundary. Another possible explanation to why the birds did not have plastic in their stomachs could be the feeding ecology, low sample size, analyzing method, or a combination of the latter.

The Common guillemots are predominantly piscivorous and feed in deeper waters than the puffins (20-50 m vs. 0-15 m) and may be less prone to mistake plastic, or at least microplastics, for prey. Day (1980) found in his study on 37 species from Alaska that the seabirds relying heavily on fish had a lower incidence of plastics than seabirds feeding on crustaceans and cephalopods. However, based on the incidental discovery of plastic in stranded birds in the southern parts of Norway in 2020 as well as two reports from the NE Atlantic (Weir et al., 1997 in Acampora et al. 2016; O'Hanlon et al. 2017 b) and two reports from Canadian waters (Provencher et al. 2015), we expected to find plastics in their stomach as well. The birds in our study were emaciated (except from the one bird from bycatch in Finnmark), like the birds from the stranding in Norway in 2020, and the fact that we did not detect plastic in the stomachs of our birds could be due to plastic ingestion rates being lower than the retention time of plastic in Common guillemots or as a result of very low sample size (<5).

The only previous study reporting plastic in Razorbills with proximity to the Arctic was performed in Wales. In their study, they found a FO of 1% (Weir et al., 1997 in O'Hanlon et al. 2017 b). Razorbills have similar feeding ecology compared to Puffins and are in the same

family (Alcidae), which contributed to an assumption of similar plastic loading. However, it was not the case in this study, and we attribute this to the fact that the Razorbills feed at a greater depth than the puffins (25-30m vs. 0-15m). Another explanation for the reported discrepancies could be the large difference in the latitude and proximity to human settlement at the sampling sites, as the Razorbills were sampled in the northernmost part of Norway and the Puffins in the south-western parts of Norway.

When it comes to Great cormorants, one study from Finland (Salmi et al., 2015 in O'Hanlon et al. 2017 b) and two studies from NE Atlantic (Carss 2009; Acampora et al. 2017a) reported plastic as either of low FO or as “occasional”/“present” and included plastic found in pellets. For European shag, the only study reporting plastic in the NE Atlantic investigated pellets only and found a low FO (Acampora et al. 2016). Studies further south (North-West Spain) found a high FO of microplastic in pellets (Álvarez et al. 2018). The Cormorants and Shags prey on larger fish and may be less exposed to direct plastic ingestion but more likely to be exposed via fish. As Great Cormorant and European shags are able to regurgitate stomach content, they may also be able to rid themselves of plastic. This suggests that the investigation of smaller-sized plastic in pellets may provide better information on plastic loading in these seabird species. Cormorants exhibit plastic incorporation in nests (Baak et al. 2021), and research focusing on this may be a more relevant method to assess exposure to plastic pollution of cormorant species in the Arctic.

### **Seasonal and regional trends**

The occurrence of plastic and the ingestion rates by seabird species may differ within a year depending on where the birds are located. For example, some studies showed Brünnich's guillemots had a higher occurrence of plastics earlier in the breeding season (Provencher et al. 2010), indicating that the plastics in their stomachs was ingested during migration to the breeding site, while others have reported higher levels during the breeding season compared to the non-breeding season. This suggests that regional differences in plastic pollution affect the seasonal plastic loading in a given species (Baak et al. 2021). In addition, all seabird species investigated in our study were sampled in the same period, and we therefore lack baseline data to make a comparison.

Finally, the retention time of plastics in the GIT is unknown for many seabirds (Ryan 2015). Northern fulmars collected in the high Arctic showed a 90% decrease in the average number



of plastics particles in the stomach over the summer and for Brünnich's guillemots, among the 13% of the birds that arrived at the breeding location in the high Arctic with plastic in their stomach, none contained plastic two months later (Provencher et al. 2010; van Franeker and Law 2015). Based on the given information, it is suggested that the migration strategy in a given species may affect plastic loading in the stomach. Therefore, the findings from the present study may not be representative for the amount of plastic at the sampling location.

## **Type of plastic**

The second aim of this study was to characterize the plastic found in the birds, distinguishing user plastic from industrial plastic, and to determine the size, color, and polymer composition of each plastic piece.

### **The occurrence of user plastic and industrial plastic**

The present study shows that the plastic in the puffin stomachs was dominated by user plastic over industrial plastics (nurdles/pellets) and that the Glaucous gulls contained user plastics only.

The small sample size in this study may be a bias in the results, but it is nevertheless interesting that surface-feeders have been found to ingest mostly user plastic whereas diving seabirds ingested more industrial pellets/nurdles and that large species ingested larger user plastic pieces as opposed to small industrial pellets (Robards et al. 1995(including Day, 1980)).

When it comes to the occurrence of industrial plastics in Glaucous gulls, there are no previous studies to compare with. However, a study by Seif et al. (2018) examined 284 pieces of debris in other gulls and found only one single industrial pellet. There are few comparative studies for the puffins as well, but our findings are similar (at least for the puffins) to the plastic composition found ingested by fulmars after the 1980s, where industrial plastic is declining and user plastics dominate (Kühn and van Franeker 2012; Ask et al. 2020; Kühn et al. 2021).

There were in total, three industrial nurdles in the puffin stomachs and 12 pieces of user plastic which included six pieces of sheet, two pieces of thread, and four pieces of fragment. Old data on plastic composition in puffins in the NE Atlantic reported rubber thread cutting as the main plastic pollutant in puffins in the North sea and that the elastic threads, when tested

in a tank, could resemble swimming pipefish (Parslow and Jefferies 1972). There were no rubber threads found in the puffins in the present study, but two pieces of thread and one long and narrow piece of sheet, which could also maybe resemble fish or polychaetae, and be mistaken for prey. Industrial nurdles could resemble fish eggs or maybe eyes, but no food items were found in the emaciated puffins to help determine the current diet of the birds. The industrial nurdles may also have been accidentally swallowed because of their small size.

The Glaucous gulls contained one piece of styrofoam, one hard fragment, and one swift tack for price tags, illustrating that the gulls can ingest various types of plastics. Industrial nurdles may also have been present in the gulls at some point but excreted prior to the sampling time. Seif et al. (2018) found in decreasing order, foam, sheet, and fragments in Glaucous gulls, but our data (only three pieces of plastic) do not allow relevant comparison.

### **Origin of the plastic found in the stomachs**

The weathering of plastic in the marine environment followed by digestive processes in the stomach make the determination of origin close to impossible. The pieces of thread in the puffins are very likely to be fishing line, but the only piece whose origin could be identified was the swift tack found in the gull. Interestingly, two fulmars collected a few months prior on a research cruise sailing on a transect from Svalbard to Greenland also contained swift tacks, and the authors suspected that these items might have been discarded or lost during shipping (Ask 2019).

### **Size and color**

The size, when measured in length, was approximately the same for the puffin and Glaucous gull. The puffins had equal amounts of mesoplastic and microplastic, and the Glaucous gull had two pieces of mesoplastic and one small piece of microplastic (3.17 mm). The size range in debris ingested by gulls in another study showed they ingested items ranging from 1.0-81.3 mm, but mostly mesoplastics (Seif et al. 2018).

In terms of weight, the heaviest piece in the Glaucous gull was four times higher in weight than the heaviest piece in the puffins, and as the large pieces in the puffins in terms of length were thin and narrow pieces of sheet or thread, it appears that the Glaucous gulls have the ability to ingest larger pieces of plastic than puffins. This makes sense as the Glaucous gull is a much larger bird (1.0-1.5 kg) than puffin (0.3-0.6 kg) and can prey on larger food items.

Provencher et al. (2019) recommended not studying plastic smaller than 1 mm as such particles will be excreted by the birds at a fast rate. In our study, a 0.5 mm sieve was included as well, but all particles detected were larger than 1 mm. It was deemed insufficient to search for particles <1 mm using the given method, and the results would most likely have been more prone to human errors. If the main focus in a study is on microplastics, other more appropriate methods should be adopted.

The color composition that dominated the samples in our study was respectively clear, white, and black. The plastic debris collected in the gulls in Seif et al. (2018) was also mainly in the white/clear category (45%). That could reflect the most used colors in plastic production, but this has not been investigated in our study. The color pigments added to plastic can be insoluble organic or inorganic particles (<https://polymer-additives.specialchem.com/selection-guide/pigments-for-plastics>, accessed April 30<sup>th</sup>, 2021), and the toxic potential of different color pigments used in the plastic industry is another interesting subject to investigate in the future.

The clear and white pieces in our study had all undergone some degree of biofouling and appeared more “light” to “medium” dark brownish. The brownish color may cause the plastic particles to resemble natural prey items, and Day (1980) proposed that the light brown particles were mistaken for small crustaceans or planktonic larvae. Biofouling is also an important factor for plastic ingestion in terms of DMS (discussed further in the section below).

### **Polymer profile**

The plastic pieces in the stomachs of Atlantic Puffins were identified as polyethylene (PE) and polypropylene (PP). The user plastics were equally distributed between PE and PP. Two of the industrial nurdles were PE, and one was PP. The plastic in the stomachs of Glaucous gulls was PP, and one piece of polystyrene (PS), and all pieces were user plastics.

This reflects the production of plastic in Europe as PE and PP are the most produced plastic polymers (Plastics-Europe 2020).

To our best knowledge, no other studies have reported polymer identity and composition in neither of the species, but one recent study on several species of Procellariiformes found PE followed by PP to be the most dominant plastic polymer identified in seabirds, both in the

number of pieces and in mass (Kühn et al. 2021). Tanaka et al. (2019) also found the plastics to be mainly made of PE followed by PP when investigating stomachs of Northern fulmar fledglings and in regurgitates from Laysan (*Diomedea immutabilis*) and Black-footed albatrosses (*Phoebastria nigripes*). The findings are similar to the present study, as PE dominated in both mass and number in puffins. When we included the mass for PP found in the Glaucous gulls, the PP was the most dominating polymer by mass.

PE and PP are both of low relative density and are the dominating floating polymers (de Haan et al. 2019) which are reflected in the plastic loading in the surface-feeding seabirds mentioned in the studies above. The puffin, on the contrary, is a diving seabird and thus expected to ingest plastic polymers with higher density that would be negatively buoyant in the seawater. Ingested plastics in another diving auk species, the Little auks (*Alle alle*) (Avery-Gomm et al. 2016), were also mainly represented by PE followed by PP. It is probable that the formation of aggregates (de Haan et al. 2019) and biofouling cause plastics to sink and become available at the subsurface.

The formation of biofilm on marine plastics can cause emittance of the foraging cue, DMS (Savoca et al. 2016) which will further increase the chances for the plastic particles to be ingested. The same authors found virgin pieces of PE and PP in seawater to be rapidly colonized by such DMS-producing organisms.

An important observation from (Kühn et al. 2021) was that the available plastic category seemed to be the driving factor of the polymer types found in the birds and not the preference for the polymer type itself. Knowledge on polymer identity is still valuable when assessing the behavior and fate of marine plastics (Cincinelli et al. 2017; Jung et al. 2018), as well as plastic-associated contaminants that could be absorbed by animals (Provencher et al. 2019).

## Sample size

The sample sizes for all species included in this study were small (<22), which resulted in the lack of statistical power. Even though the FO in the puffins was medium/high, the total number of particles in the two species, especially in the gulls, limited the opportunity for statistical analysis and made the summary statistics recommended reported by Provencher et al. (2019) was less feasible for the present study.

For most seabirds used in plastic studies, a minimum sample size has not been established in order to monitor the changes in prevalence and mass of ingested plastic by necropsy of seabirds. Nevertheless, (Provencher et al. 2015) conducted a power analysis using existing data from plastic ingestion in several species in Canada and for Common guillemots from Newfoundland and concluded that 106 Common guillemots would be needed annually to detect a 25% change in ingestion and up to 613 birds to detect a change of 20%. The sample size of >40 birds annually over a 4-8 year period, which is required for the Northern fulmar in the North sea to detect a 25% temporal change in ingested plastic (Van Franeker and Meijboom 2002), is often referred to when discussing sample sizes for birds, but the necessary annual sample size has proved to vary a lot between species and location (Provencher et al. 2017).

While a higher sample size would have been beneficial for statistical analysis and monitoring, this could result in the unwanted death of Arctic seabird species, some of the species being red-listed, especially when stranded birds are not available.

Although this study was small-scale and outside an established monitoring program, all plastic ingestion studies that use standardized methods will contribute with valuable data and enable the opportunity for comparison in future studies.

## Sample collection and dissection

The protocol for collection recommended by Provencher et al. (2019) could not be assessed in this study as it was not done by the author.

This study solely investigated carcasses, and the carcasses came from beached birds, fishery bycatch, and culling for scientific research. As Provencher et al. (2019) stated, the disadvantage of using carcasses is that the sampling is generally opportunistic, and some species may not be available for sampling. Sampling of dead birds (i.e., beached birds) may be biased as the source of mortality can influence the findings of plastic and the interpretation of data (Auman et al. 1998 in Rodríguez et al. 2018; Provencher et al. 2019). No significant difference in plastic burdens have been found when beached Northern fulmars were compared to birds that died in good condition (e.g., fishery bycatch) (Van Franeker and Meijboom 2002), but findings in a study of Short-tailed shearwater fledglings (*Ardenna tenuirostris*) were contradicting as the plastic burden in naturally beached fledglings were higher than in the those collected as road-kill (Rodríguez et al. 2018)

Similar studies have not been conducted for the seabird species used in the present study. However, it is noteworthy that the Glaucous gulls were culled while in good condition with stomachs containing mostly natural food items and had a low plastic FO, whereas the puffins were beached birds in poor condition with stomachs empty of natural food item. Although this could affect the results, this remains to be determined.

However, the protocol for dissection was straight forward and applicable for all the species investigated in this study. As recommended, only the stomachs were used as previous studies have shown (Van Franeker and Meijboom 2002) that the investigation of the small intestines was not relevant in studies on plastic occurrence in the stomach of seabirds as plastics are retained in the proventriculus and gizzard (Day et al. 1985; Provencher et al. 2019). The protocol recommends that, whenever possible, the plastic loading should be reported for each compartment of the GIT. This successfully applied to the Glaucous gulls but turned out to be more difficult for the puffins as their stomachs were a lot smaller and their contents more mixed between the compartments. For future studies in which the difference in plastic loading between the proventriculus and gizzard is of interest, the compartments should be sealed when the birds are sampled. Although this will never discard the risk for cross-contamination of compartments.

## Future research

It is important to distinguish between research aiming to monitor trends, and research focusing on the differences in plastic ingestion across regions, between and within seabird species. Both fields are highly essential to understand the distribution of marine plastics and how it affects seabirds (Provencher et al. 2015)

Regular monitoring should be implemented over different spatial and temporal scales, and for long-term monitoring, species with a low prevalence of plastic should be of interest as well. (Avery-Gomm et al. 2013; Provencher et al. 2014; Poon et al. 2017; Dehnhard et al. 2019; Kühn and van Franeker 2020; Baak et al. 2021).

Feeding ecology in terms of diet preference and foraging behavior of a species is of great importance when assessing plastic pollution in seabirds. Our findings and those of previously mentioned studies (e.g., Day et al. (1985) and Amélineau et al. (2016)) suggest that we gain limited information on plastic distribution in seabirds and exposure to seabirds focusing mainly on surface feeders.

As the future for many Arctic seabirds may include stress related to food access, and emaciated seabirds have been found with plastics in their stomach, it would be interesting to investigate the potential for correlations.

Areas of high productivity resulting from oceanographic phenomena such as upwellings could potentially lead to more ingested plastic by seabirds feeding in such areas. It would therefore be interesting to investigate birds caught in or near upwelling areas.

As human population and human activity increase in the Arctic region, the risk of plastic ingestion in Arctic seabirds may increase. In 2021 CAFF suggested that Northern fulmars, Thick-billed murre, and Black-legged kittiwakes should be indicator species of plastic pollution in the Arctic (Baak et al. 2021). We know little about the FO of plastic in seabirds across Norway and Svalbard (O'Hanlon et al. 2017 a), but European shags, Common guillemots, and Atlantic puffins are species that have been identified by Dehnhard et al. (2019) as interesting Norwegian seabirds species to monitor plastic ingestion.

The findings in this study support the use of Atlantic puffins as a relevant indicator species. The Glaucous gull is a sentinel species in the Arctic and may be a relevant species to study in



the light of long-range plastic pollution, increasing human activity, and the ongoing Atlantification of the Arctic.

Non-invasive methods should be further investigated to prevent birds from being culled for research. Pierce et al. (2004) suggested using ultrasound as a promising tool to detect plastic in the stomach of seabirds, but to our knowledge, there has been little to no development in that particular field, except for current investigation by researchers from the Norwegian Polar Institute (Geir W. Gabrielsen, personal communication, April 2021). Another non-invasive method could be biomarkers and in a study by Neumann et al. (2021), they found a brominated flame retardant (BDE209) in the liver tissue of birds that had ingested plastics. Their findings suggested that the BDE209 was derived from the ingested plastic and that a high concentration of BDE209 could be used as a general indicator of plastic exposure prior to sampling. More research on biomarkers and non-invasive methods to measure them can prove valuable.

The potential for hazardous chemicals being transported from marine plastic to seabirds is another area of plastic research that needs more attention as the future of many Arctic seabird species is threatened. The toxic effect from ingested plastic may have a great negative impact at the individual-, population- and ecosystem level (Tanaka et al. 2013; Tanaka et al. 2019; Tanaka et al. 2020).

## Conclusion

The present study has provided a snapshot of the current status of plastic occurrence in seabirds breeding in the Arctic. The main findings showed a medium-high FO in Atlantic puffins and a low FO in Glaucous gulls. The plastic polymers were identified as PP, PE, and PS, and user plastic dominated over industrial plastic in both seabird species. Our data will serve to establish a baseline for future research and monitoring of Atlantic puffins and Glaucous gulls. The null findings of plastics in the stomachs of Common guillemots, Razorbills, Great cormorants, and European shag contribute with valuable information and a point of comparison for future research on plastic ingestion in both Arctic seabirds in general and in Norwegian breeding populations

The findings in the present study suggest that we gain limited information by focusing mainly on surface feeding seabird species, and the Atlantic puffin is therefore suggested as a relevant indicator species for diving seabird species. The Arctic is affected by long-range plastic pollution, Atlantification, and increasing human activity, and in regard to that, the Glaucous gull may be a relevant sentinel seabird species for future studies. Non-invasive methods should be further investigated, and the research on potential toxic effects from ingested plastic should be intensified.

“To establish a better understanding of the growing issue of plastic marine debris in the marine environment, we require a region-wide, coordinated effort to collect information on both plastic ingestion and nest incorporation, collected and reported in a standardized manner” (O'Hanlon et al. 2017 a). In this context, opportunistic studies have proven to be of value, and as such, our study has met its goals by providing new quantitative and qualitative data on plastic loading and polymer type reported in a standardized manner.

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## Appendix A

Table 6 Polymer type, color code, length (mm), size category and plastic type (user/industrial) for each piece of plastic found in the birds.

Plastic ID	Species	Bird-ID	Polymer	Color code	Size (mm)	Weight (mg)	Size class	Origin
#1	Glaucous Gull	LH-36	PP	WH-CL	11.18	13.6	meso	user other
#2	Glaucous Gull	LH-32	PP	WH-CL	16.28	143.5	meso	user fragment
#3	Glaucous Gull	LH-16	PS	WH-CL	3.17	1.4	micro	user foam
#4	Atlantic Puffin	ROG-2016-011	PE	GREE	6.17	28.2	meso	user fragment
#5	Atlantic Puffin	ROG-2016-012	PE	BLA	4.43	3.0	micro	user fragment
#6	Atlantic Puffin	ROG-2016-017	PP	WH-CL	9.51	2.6	meso	user sheet
#7	Atlantic Puffin	ROG-2016-017	PP	WH-CL	4.31	2.0	micro	user sheet
#8	Atlantic Puffin	ROG-2016-017	Other	BLA	4.57	19.4	micro	user fragment
#9	Atlantic Puffin	ROG 2016-018	PE	WH-CL	7.11	7.7	meso	user sheet
#10	Atlantic Puffin	ROG 2016-018	PP	WH-CL	9.34	0.7	meso	user sheet
#11	Atlantic Puffin	ROG-2016-023	PP	WH-CL	23	1.4	macro	user thread
#12	Atlantic Puffin	ROG-2016-023	PE	BLA	3.38	11.6	micro	industrial nurdle
#13	Atlantic Puffin	ROG-2016-023	PP	WH-CL	4.12	13.3	micro	industrial nurdle
#14	Atlantic Puffin	ROG 2016-026	PE	WH-CL	6.36	2.2	meso	user sheet
#15	Atlantic Puffin	ROG-2016-043	PE	BLA	10.17	34.9	meso	user fragment
#16	Atlantic Puffin	ROG 2016-048	PP	WH-CL	20.43	19.2	macro	user sheet
#17	Atlantic Puffin	ROG-2016-049	PE	WH-CL	4.33	21.5	micro	industrial nurdle
#18	Atlantic Puffin	ROG-2016-052	PE	WH-CL	29.05	2.4	macro	user thread

