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The northern fulmar as a sentinel species for monitoring the environmental health impacts of plastic exposure

Christina Biamis ^a, Susanne Kühn ^b, Ana Payo-Payo ^c, Jaimie Dick ^{a,d}, Pierre Bize ^e, Jan van Franeker ^b, and Gary Hardiman ^{a,d}

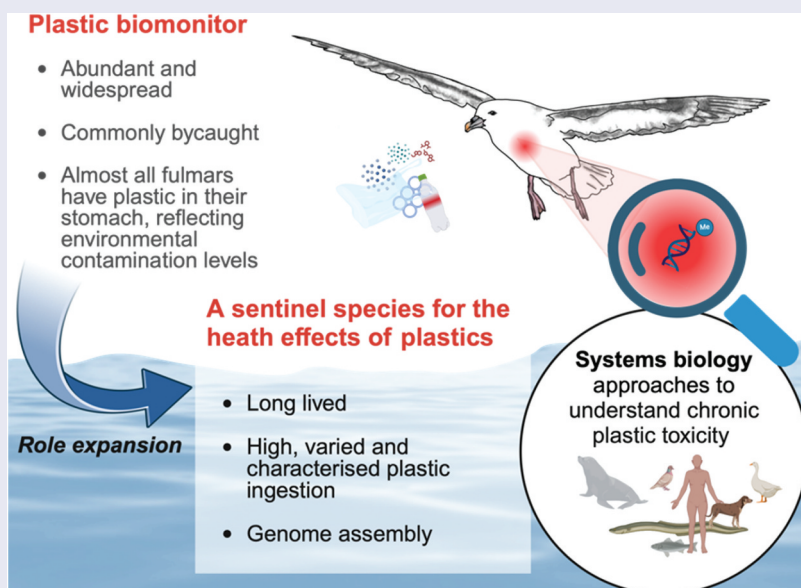
^aSchool of Biological Sciences, Queen's University Belfast, Belfast, UK; ^bWageningen Marine Research, Den Helder, The Netherlands; ^cDepartment of Biodiversity, Ecology and Evolution, Complutense University of Madrid, Madrid, Spain; ^dQueen's Marine Laboratory, Queen's University Belfast, Belfast, UK; ^eSwiss Ornithological Institute, Sempach, Switzerland

ABSTRACT

The northern fulmar (*Fulmarus glacialis*) is well suited to contribute to a “One Health” approach for understanding plastic-associated toxicity. Plastics contain additives that act as endocrine-disrupting chemicals (EDCs), which leach into the environment, often disrupting hormonal signaling in wildlife. As an ecotoxicological model, the fulmar, a seabird, provides a unique opportunity to investigate these effects under real-world exposure conditions. Fulmars are currently utilized in The EU's Marine Strategy Framework Directive as passive bioindicators of marine litter in the Northeast Atlantic, based upon the proportion of individuals with significant plastic burdens in their stomachs. The aim of this review is to advocate for elevating the fulmar's role from a passive indicator to active sentinel species to better understand long-term environmental health impacts of chronic plastic exposure. The fulmar's unique ecological niche, high trophic level, and wide-ranging foraging behavior make it particularly vulnerable to plastic ingestion, providing a relevant model for exposure assessment. The role of epigenetic mechanisms was examined in mediating gene–environment interactions, particularly how plastic exposure may influence gene expression and contribute to adverse health outcomes. Further, advances in genome sequencing and annotation enabled identification of molecular pathways affected by plastic-derived EDCs, contributing to the development of complex adverse outcomes that might be characterized through systems biology. Finally, practical benefits and methodological challenges of integrating molecular and genomic analyses into fulmar-based monitoring, focusing upon bycaught individuals and long-term study cohorts, was determined. This framework supports the fulmar's potential as a sentinel species for monitoring plastic-related health risks in marine ecosystems.

KEYWORDS

Plastic-ingestion; northern fulmar; endocrine disrupting chemicals; sentinel; epigenetics; one health



CONTACT Christina Biamis  cbiamis01@qub.ac.uk; Gary Hardiman  g.hardiman@qub.ac.uk  School of Biological Sciences, Queen's University Belfast, 19 Chlorine Gardens, Belfast BT9 5DL, UK

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Expanding the Northern Fulmar's Role: From Plastic Biomonitor to Sentinel Species The northern fulmar is uniquely positioned to bridge environmental plastic monitoring and health impact assessment within a One Health framework. As a plastic biomonitor, the fulmar offers broad geographic coverage, frequent bycatch sampling opportunities, and near-universal plastic stomach content making it a direct mirror of marine plastic contamination and an established indicator. Role expansion to sentinel species is supported by several biological and logistical advantages: its longevity enables chronic exposure assessment; its high, varied and well-characterized plastic ingestion spans polymer types and sizes; an available genome assembly enables molecular investigation; and archived tissue banks allow retrospective analysis. Systems biology approaches including genomics, transcriptomics, metabolomics, proteomics and epigenomics applied to fulmar samples can reveal the molecular mechanisms of chronic plastic toxicity across biological scales. Within a One Health framework, findings integrate across wildlife, human and ecosystem health, generating insights that can directly inform plastic regulation and policy.

Introduction

Plastics are high molecular weight polymers of various sizes, from macroplastic (>25 mm) to mesoplastic (25–5 mm), microplastic (5 mm–100 nm) and nanoplastic (<100 nm) (Bibi et al. 2023). Larger plastics might initiate health issues for wildlife when ingested, including local inflammation, fibrosis and gastrointestinal obstructions (Charlton-Howard et al. 2023). Micro and nanoplastics (MNPs) similarly affect health, but due to their smaller size, they are more easily transported to distal tissues (da Silva Antunes et al. 2025; Feng et al. 2023; Fontes et al. 2024). All plastics contain a myriad of additives and monomers, many of which are endocrine disrupting chemicals (EDCs) including bisphenol A (BPA) and phthalates. However, in the environment, plastics also adsorb other EDCs such as polychlorinated biphenyls (PCBs) and polyfluoroalkyl substances (PFAS). These plastic-associated EDCs initiate broad-spectrum and systemic adverse health effects in both humans and wildlife through interference with fundamental physiological processes (Gore et al. 2015; Lee et al. 2024; Patlewicz et al. 2024). Their broad-spectrum toxicity is facilitated by epigenetic mechanisms (Jacobs et al. 2017). The end results of plastic-induced dysregulation of these processes include developmental, reproductive and metabolic dysfunction (Marlatt et al. 2022; Patlewicz et al. 2026). Given the pervasive environmental health issues posed by plastic pollution, a Global Plastics Treaty is being negotiated (Brander et al. 2024).

Traditional approaches to study plastic toxicity include *in vitro* and *in vivo* models, but these may not capture the real-world complexity of chronic exposures and maladaptation. Nonetheless, these

findings have contributed substantially to regulations limiting production of EDCs (Bolker 2017). However, regulatory barriers are often overcome merely by industry pivoting to newer chemical alternatives; many of which exhibit similar toxicities. The adverse health effects of plastic's chemical mixtures are further complicated by heterogeneity in size and morphology of plastic polymers (Xu et al. 2022). These diverse factors challenge the replication of environmental conditions in lab-based exposure studies. In addition, since epigenetic alterations often represent the initial molecular events underlying health impacts from environmental exposures and given that epigenetics mediate interaction between environmental factors and the genome, studies examining epigenetic responses to plastic exposure are particularly susceptible to environmental variability. The limitations of traditional toxicological approaches make ecotoxicological studies in wild species particularly valuable in mirroring real-world plastic exposure patterns and understanding the holistic adaptations to this chronic exposure.

Integrating diverse data sources is critical for elucidating the complex and dynamic biological responses to plastic exposure. Systems biology provides a holistic framework for environmental health research by applying computational models to analyze multifaceted datasets (Millar-Wilson et al. 2022). Comparative genomics complements this approach by revealing evolutionary relationships, gene functions, and genetic variants linked to disease susceptibility and adaptive responses. These methodologies are instrumental in conservation biology, as these methods facilitate identification of patterns of genetic diversity and adaptation. As

marine environments are major sinks for plastics and associated pollutants, apex marine species, often used as bioindicators, frequently ingest substantial quantities of plastic debris (Coyle et al. 2020; Coyle et al. 2023). Therefore, systems biology investigations targeting these species provide valuable insights into the ecological and health consequences of plastic contamination. By integrating molecular, physiological, and environmental data, these studies (1) enhance our ability to understand organismal responses, (2) identify biomarkers of exposure, and (3) model the broader impacts of plastic pollution within marine ecosystems.

However, multiple barriers exist in expanding the role of bioindicators to ecotoxicological models of plastic exposure, particularly in a systems biology context. Most wild species still lack completed genome builds and the associated annotation that are essential for successful analysis of epigenomic, transcriptomic and proteomic data. Further ethical considerations, conservation concerns, and logistical challenges pose significant barriers to the use of wild species in environmental research involving animal sacrifice.

The northern fulmar seabird (*Fulmarus glacialis*) has long been recognized as a plastic bioindicator in the North-East (NE) Atlantic, passively reflecting the extent of plastic pollution through its ingestion of plastic waste (van Franeker et al. 2021). However, the health impacts of such exposure remain understudied in this long-lived predator. The aim of this review is to propose the use of fulmars as an active sentinel species for elucidating environmental health impacts of exposure to plastics. The fulmar possesses many characteristics that make it uniquely suited to this purpose, which include (1) a tendency to ingest plastic, (2) a bioindicator status, (3) longevity, (4) behavioral and anatomical characteristics, (5) complete genome build, and (6) availability of diverse fulmar sample types (van Franeker et al. 2005). This role expansion of the fulmar from a passive indicator of plastic pollution to that of an active sentinel species is in agreement with the need to understand not only the presence, but also the environmental health impacts of

exposure to pollutants, filling this critical gap in risk assessment and management.

The objectives of this review were to (1) examine the implications of epigenetic mechanisms in understanding the integrated health impacts of plastic-associated EDCs; (2) delineate the unique ecological and biological characteristics of fulmars that qualify these birds as a sentinel species for investigating the adverse health effects attributed to exposure to plastics and their additives; and (3) outline the practical considerations required to integrate ecotoxicological data from fulmars into broader environmental health assessments.

Considerations for a systems biology and ecotoxicological approach to plastic toxicology

This section examines the integral role of epigenetics in how organisms biologically respond to chemicals released from plastics. The contributions of ecotoxicology and systems biology to understanding the environmental health impacts of these exposures are also described.

Role of epigenetics in biological responses to chemicals in plastics

Over the past two decades, the critical role of epigenetic mechanisms in responding to EDCs has become increasingly apparent (Jacobs et al. 2017). Endocrine-disrupting chemicals are defined by their ability to disrupt the endocrine system by mimicking, blocking, or altering the production and transport of hormones and their receptors (Gore et al. 2015; Kojima et al. 2010). Epigenetics describes the study of changes in gene expression without alterations to the DNA sequence itself. This field, emerging in the 1940s, was redefined in 2011 as encompassing “molecular factors and processes around DNA that regulate genome activity independent of DNA sequence and are mitotically stable,” meaning these compounds persist through cell divisions (Skinner 2011). Such epigenetic alterations modify gene expression profiles, potentially leading to cellular dysfunction and adverse health outcomes (Montjean et al. 2022). An intricate bidirectional relationship exists between the endocrine system and epigenetic

mechanisms in mediating these effects. Estrogen receptors play an important role in responding to exposure to plastic's chemicals (van Hassel et al. 2025). This relationship is complex because different epigenetic states play essential roles in cell differentiation and development, as well as in regulating hormonal responses and adaptations to environmental changes. However, exposure to exogenous EDCs might lead to aberrant epigenetic changes, resulting in deviations from normal developmental and wellness end-points (Safi-Stibler and Gabory 2020).

Epigenetic changes might occur through processes involved in alterations in DNA methylation (DNAm), non-coding RNA regulation, and histone modification, often leading to changes in gene expression (Skinner 2011). Further, these alterations may be long-lasting. High-throughput sequencing (HTS) approaches are emerging as instrumental in unraveling DNAm changes in non-model species. Investigating these perturbations might provide crucial insights into the regulatory mechanisms disrupted by exposure to plastics and their EDCs. DNAm-Seq characterizes the presence or absence of a methyl group ($-CH_3$) in DNA. DNAm aberrations induced by plastic-associated EDCs might result in dysregulation of transcription and biological processes. For example, exposure to di(2-ethylhexyl) phthalate (DEHP), a common plasticizer, was linked to significant alterations in DNAm patterns and increased expression of methyl-CpG-binding proteins and DNA methyltransferases (DNMTs) involved in DNAm, resulting in transcriptional repression (Martinez-Arguelles and Papadopoulos 2015). Similarly, Huff et al. (2018) reported that DEHP exposure was associated with differential expression of genes involved in key metabolic processes. Several investigators observed that adult reproductive success and survival rates was frequently found to decrease due to negative impacts experienced during early development, when organisms are more developmentally sensitive (Bateson et al. 2014; Douhard et al. 2016; Monaghan 2008). Alarming, some epigenetic modifications that persist throughout meiosis are heritable through the germline. In the context of plastic exposure, this finding suggests that EDC-related epigenetic alterations may be transmitted in a transgenerational manner to subsequent generations not directly

exposed to the contaminant (Chamard-Jovenin et al. 2017). These chronic effects are particularly concerning in the context of plastics considering their biodegradation-resistant characteristics.

Role of ecotoxicology and systems biology in capturing complexity of biological responses to exposure

The highly context-specific nature of epigenetic states emphasizes their role as a critical intermediary linking gene expression changes with environmental exposures to EDCs (Sepers et al. 2019). Despite the advances provided by many models for examining EDC effects, *in vitro* cellular models cannot replicate the systemic effects induced by EDCs. *In vivo* preclinical controlled studies typically use rodent models (Benjamin et al. 2017; Jennings et al. 2012). It is of interest that Dolinoy, Huang, and Jirtle (2007) showed that in the well-known Agouti mouse, BPA exposure induced heritable hypomethylation-related health changes. However, the lab environment in which rodents are housed is known to induce epigenetic changes and unique adverse health effects (Cait et al. 2022). This may render the study of these environmentally sensitive states and their alterations challenging in lab environments.

Amidst the prevailing and regrettable circumstance of wildlife exposure to improperly managed plastic waste, ecological models to study plastic toxicity emerged as a byproduct (Biamis et al. 2021; Huang et al. 2020). Natural experimental groups exist where groups of individuals are compared that differ in their exposure to plastics and EDCs temporally or by geographical differences. These open *in vivo* systems offer promising alternatives to lab animal testing (Vidal-Dorsch et al. 2013). Open systems are characterized by numerous interacting variables, which create significant complexity when trying to establish direct causality, especially with many confounding factors present. However, causality may be gauged by the scale and reproducibility of the effect, as well as temporal sequences and gradient effects (Matthiessen et al. 2018). Wild feeding studies are also useful in this regard but might be associated with ethical barriers (Tanaka et al. 2020).

Research in the wild is necessary to understand the consequences, together with the mechanisms, in an ecologically and evolutionary relevant setting where environmental factors fluctuate and resources are often limited, and thus subject to strong natural selection. Humphries et al. (2021) in a study in the Nile crocodile, *Crocodylus niloticus*, found that periods of environmental stress were associated with increased toxicological risk from environmental organochlorine bioaccumulation. Indirect, often subtle, adverse effects of anthropogenic compounds, such as behavioral modifications may be observed in nature that might exert pervasive effects on individual reproduction and survival. PFAS and PCBs for example were associated with egg turning behavior in kittiwakes, *Rissa tridactyla* (Blévin et al. 2020). Related to this, exposure to PCBs has been linked to egg-shell thinning in aquatic birds (Faber and Hickey 1973). Population level effects of EDCs were reported in humpback dolphins (*Sousa chinensis*), with alkylphenols as the most impactful contaminant of emerging concern in this capacity (Luo et al. 2024). This indicates the value of ecotoxicological studies in understanding the effects of exposure to plastics and their EDCs on health.

Role of sentinel species in plastic and EDC ecotoxicology

Bioindicator or biomonitoring species play a passive role in assessing contamination levels compared to sentinels which play an active role in assessing the impacts of contamination, however the terms are often used interchangeably (Galimany et al. 2023). Bioindicators track changes in the environment, assess effectiveness of management practices, and serve as early warning systems for imminent ecological transformations (Siddig et al. 2016). A review of terrestrial and aquatic avian bioindicators of plastics and other contaminants over the past 30 years found that plastic-associated compounds such as EDCs are threatening birds (Mansfield et al. 2024). However, the health effects of these exposures in bioindicators have been largely neglected.

Sentinels in contrast provide advanced warnings of risks and hazards, due to their environmental niches, habitats, and behaviors. These species

provide a measurable response to the hazard, which may subsequently be employed to infer risk to humans and other species (US 1991). The first historic image of a sentinel species is the canary utilized in coal mines. Owing to their small size and rapid breathing, canaries are highly sensitive to carbon monoxide. Thus, if the canary fell from its perch, advance warning of carbon monoxide hazard was provided to the miners (Spencer 1962). Other examples include pet dogs as sentinels for environmental chemicals that affect fertility (Sumner et al. 2020), osprey as sentinels for environmental contaminants in lakes, reservoirs rivers and estuaries (Grove et al. 2009) and fish sentinels for environmental pollution in Brazilian river reservoirs and coastal California (Baker et al. 2009; Rubio-Vargas et al. 2024). The American alligator, *Alligator mississippiensis*, is a reliable sentinel species due to its role as an apex predator in sensitive wetland ecosystems, where this animal bioaccumulates contaminants over its long lifespan (Guillette et al. 2016). Their well-documented physiology and susceptibility to pollutants in these sentinels enable investigators to detect deviations from baseline health and assess broader ecological impacts (Guillette et al. 2016; Kohno et al. 2023). Investigators have effectively used this model to investigate the epigenetic effects of anthropogenic chemicals in reptiles in aquatic environments (Guillette et al. 2016; Kohno et al. 2023). Given that large-ranging marine birds are frequently used as bioindicators for environmental plastic pollution, data suggests the possibility of leveraging the avian model, like the crocodilian approach, to explore biological responses to plastic ingestion (Clark et al. 2023; Rodríguez et al. 2024; Roman et al. 2020; Savoca et al. 2024; Thibault et al. 2024).

Wild birds as sentinel populations to assess environmental health impacts of plastic exposure

All vertebrates have complex endocrine systems that regulate stress responses and energy levels through hormone secretion, notably through the hypothalamus-pituitary-thyroid (HPT) and the hypothalamus-pituitary-adrenal (HPA) axes, which maintain homeostasis (Galligan et al. 2023). However, birds are unique in their high metabolic rates due to the metabolic energy and

oxygen requirements for flight, as well as their relatively complex social systems (Carere et al. 2010). These requirements come at a cost and may enable the effects of EDCs to be more visible in terms of health markers. Seabirds differ from many other birds in their life history as they are K-selected, meaning these birds produce a low number of slow growing offspring over a long lifespan (Hasimuna et al. 2023). For K-selected species, life history theory suggests that the fitness of individuals at one stage of their life is conditioned by occurrences in previous stages and this makes them more sensitive to the cumulative effect of chronic toxicant exposures (Dobson and Jouventin 2007; National Research Council (US) 1991).

Although the number of wild seabird studies quantifying plastic-associated EDCs in tissues are numerous, with a particular focus on legacy contaminants like polybrominated diphenyl ethers (PBDEs) (Burger and Gochfeld 2001; Tanaka et al. 2015; Yamashita et al. 2011), studies of plastic-derived EDCs in these populations are rare (Tanaka et al. 2020). Thus far, most investigators accessed plastics in the digestive system from dead birds (of bycatch and beached origins) (Hanifen et al. 2025). Correlations with plastic ingestion in these populations are limited by a cross-sectional approach or a “snapshot” problem whereby plastic stomach contents at the time of death do not necessarily reflect longer term EDC exposure patterns (Kühn et al. 2022). Recently, there has been a move toward using minimally invasive methods to measure exposure to plastics and EDCs in live birds, including determination of phthalate levels in the preen oil gland (Gray et al. 2024; Hardesty et al. 2015; Provencher et al. 2019). These observations may permit dynamic measurements over time.

Investigations of the epigenetic adaptations in birds in response to plastic-derived EDCs are limited. Ecological epigenetic studies usually focus on plants, humans, and lab rodents, with few studies of epigenetic marks in wild birds (Sepers et al. 2019). Nonetheless, it is known that maternal exposures in birds might alter DNA methylation patterns of the offspring (Ahmed et al. 2014; Liu et al. 2018), and transgenerational inheritance was noted in chickens (He et al. 2023). Evidence that the

consequences of early life exposures on DNA methylation persist throughout a lifetime was demonstrated in birds (Sepers et al. 2019).

The biological pathways by which EDCs exert their toxicity are largely shared across species, in part due to the conservation of disease-related genes and biological pathways, enhancing applicability and relevance of ecotoxicological findings to broader environmental health (Colbourne et al. 2022; Hardiman 2020; Huff et al. 2019). This enables the use of comparative genomics for non-model species that do not have detailed genome annotations, like many wild birds. By leveraging orthologous gene sequences from closely related model species, transcripts may be identified in these non-model organisms (Richards et al. 2018). The Adverse Outcome Pathway (AOP) framework provides a structured approach to understanding the specific biological events linking chemical exposure to adverse effects (Ankley et al. 2010; Biamis et al. 2021; Huff et al. 2019). The AOP framework can be used to design more effective sampling programs by linking plastic exposure to specific biological responses and targeting measurable key events (KEs) along that pathway. This approach prioritizes biomarkers closely tied to exposure, such as indicators of gut injury, oxidative stress, endocrine disruption, immune changes, and particle translocation, and guides the choice of appropriate tissues or matrices, including feces, blood, feathers, and microbiome samples. By aligning sampling with biologically sensitive periods (e.g., breeding, chick-rearing, molt) and incorporating repeated measures and molecular endpoints where possible, researchers can reduce the limitations of “snapshot” sampling and better capture temporal patterns. Pairing particle metrics with biological markers in the same individuals and applying weight-of-evidence criteria across multiple KEs strengthens inference of plastic impacts. In practice, this could involve seasonal sampling of marked individuals using non-lethal sampling and standardized particle assessments to track both exposure and biological effects over time. Overall, this facilitates improved prediction and understanding of the health and ecological risks associated with environmental stressors, which is essential for informing regulatory decisions and conducting risk assessments (Verga et al. 2022).

Characteristics of northern fulmars and their utility as a systems biology sentinel of plastic exposure

The biological consequences of exposure to plastic chemicals and the role of sentinels in understanding the complex consequences have been discussed, and now our attention is focused on the role of fulmars in this capacity. Since 2002, the stomach contents of bycaught and beached fulmars were utilized by Oslo-Paris Convention (OSPAR) and European Union Marine Strategy and Framework Directive (MSFD) for biomonitoring plastic pollution in the North Sea, later extending to the Northeast Atlantic. The biomonitoring capacity of fulmars is attributed to plastic stomach contents being representative of environmental plastic contamination levels in their marine feeding areas (OSPAR Commission 2008; van Franeker et al. 2011, van Franeker et al. 2022, van Franeker and Meijboom 2002). Various parameters including (1) history of fulmars as a marine plastic bioindicator, (2) characteristics and biology, (3) distribution, (4) breeding ecology, (5) behavior, (6) conservation, and (7) interaction with plastics are reviewed here to understand the potential of use of these birds in a system biology context. In addition, recent efforts to characterize fulmar plastic ingestion and EDC bioaccumulation are summarized.

The fulmar's history as a bioindicator of marine plastic pollution

Catalyzed by a major oil spill, OSPAR was formed in 1972 by multiple governments and the EU to provide continual assessment of the Northeast Atlantic marine environment. Following a pilot study carried out by van Franeker and Meijboom (2002), OSPAR accepted beached fulmars stomach contents as a reliable and sensitive monitoring tool of marine plastic levels in Northeastern Atlantic areas. Investigators also confirmed that levels of ingested plastics in dead beached fulmars were markedly similar to that of living birds at any given time (van Franeker and Meijboom 2002; J. van Franeker et al. 2005). Outside the North Sea, studies on plastic ingestion by fulmars are more incidental, as recently summarized by de Bruin et al. (2025).

Due to the success of this risk assessment tool, OSPAR implemented a quantifiable OSPAR Ecological Quality Objective (EcoQO) as a policy instrument, utilizing beached fulmars to inform governmental bodies regarding regional marine pollution. The EcoQO stated that < 10% of beached fulmars should contain > 0.1 g of plastic in their stomach. Recently, at the request of the Marine Strategy Framework Directive, the EcoQO was replaced by the “Fulmar Threshold Value” (Fulmar-TV) which utilizes the same methodology and target definition (van Franeker et al. 2021). This threshold remains regularly exceeded within the North Sea region (Kühn et al. 2022). Geographical patterns in fulmar plastic ingestion reveal varying rates across different locations, with more pristine areas like High-Arctic Canada showing reduced rates (Fulmar-TV% of 10%) compared to lower latitudes like California (86%) (van Franeker et al. 2022). These patterns might be related to ocean currents, transporting plastic litter over large areas (de Bruin et al. 2025).

Due to shared characteristics within the Procellariiformes order, to which the fulmar belongs, other common Procellariiformes such as petrels, albatrosses, and shearwaters have also been proposed as plastic bioindicators globally (Baes et al. 2024; Perold et al. 2025; Phillips and Waluda 2020; Puskic et al. 2025; Rodríguez et al. 2024; Thibault et al. 2024).

Fulmar characteristics for marine plastic biomonitoring

Some characteristics of fulmars make them particularly useful for marine plastic biomonitoring (Figure 1). Their exclusive foraging at sea (in contrast to gulls, *Laridae*), and their anatomy resulting in plastic retention within their stomach, reflects relative site fidelity and litter pollution in their immediate marine environment (OSPAR Commission 2008). The surface feeding behavior of fulmars, combined with their widespread distribution across Arctic and boreal seas, and their ability to retain ingested plastics for extended periods, indicates that their stomach contents provide a comprehensive “snapshot” of environmental conditions over broad geographic areas and timeframes (Kühn et al. 2022; OSPAR Commission

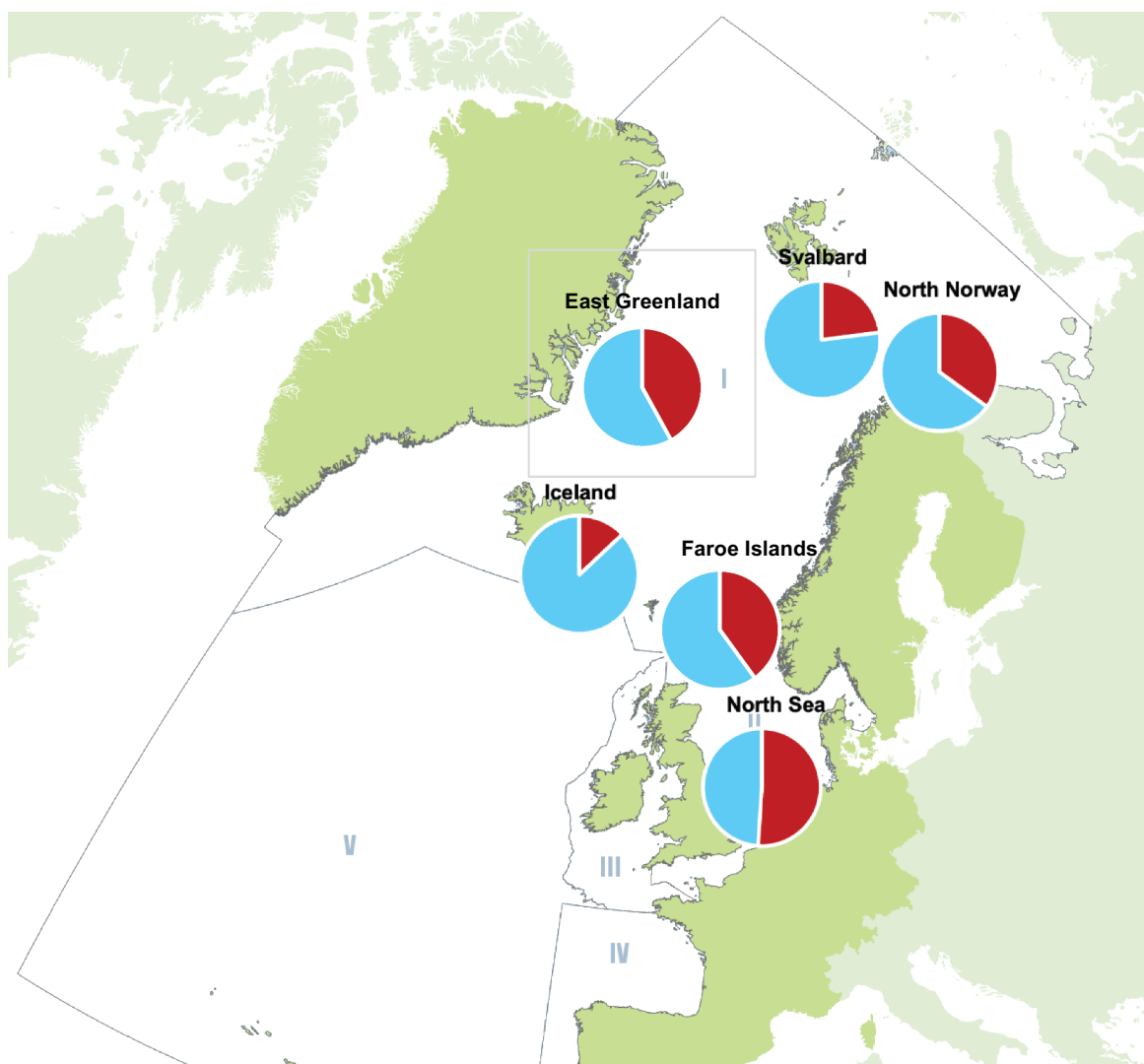


Figure 1. Geographical differences in plastic pollution measured using fulmar stomach contents. Data from van Franeker et al. was plotted in pie charts (van Franeker et al. 2022). Red represents the Fulmar-TV%, i.e. the percentage of fulmars with more than 0.1 g of plastic in their stomach. Blue represents the percentage that do not exceed the limit. All data is from this century in the Northeast Atlantic region. The map backdrop (OSPAR 2024) focuses on OSPAR management regions, where fulmars are mainly used as ecological quality indicators. For measurements outside this region, see van Franeker et al. (2022). As a sample size of 40 provides more reliable averages, measures below this sample number were excluded for this demonstration (Ireland, NE Greenland). Where multiple studies exceeded 40 samples for one location, the most recent measurement was utilized. East Greenland $n = 147$, Iceland $n = 121$, Faroe Islands $n = 699$, North Sea $n = 393$, North Norway $n = 72$, and Svalbard $n = 40$.

2015). These biomonitoring characteristics coalesce with others, such as their long lifespan, to make them ideal sentinels for the adverse environmental health effects of plastic and their associated chemicals.

Anatomical and gastrointestinal characteristics

Fulmars are (1) abundant generalist seabirds, ranging between 45 cm and 50 cm long, (2) possess a typical wingspan of approximately 110 cm, (3)

an average body mass of 767 g, and (4) are sexually monomorphic (Hatch and Nettleship 1998; Mallory et al. 2020a, 2020b). Body colors vary from mostly white to dark grey. The characteristic “tubenose” as well as their stiff winged flight pattern makes them readily identifiable by observers (van Franeker and Wattel 1982). The fulmar stomach is divided into two units, firstly, the proventriculus (fore-stomach) is where food is stored and digestion begins, and secondly the ventriculus or gizzard which is smaller and more muscular for

the grinding of hard foods before passage into the gut (van Franeker and Meijboom 2002). Between the gizzard and intestines, food as well as plastics need to pass the narrow and acutely angled pyloric sphincter, which may trap plastics present for some time (Furness 1985). Compounding this, fulmars do not regurgitate indigestible prey items as regularly as other birds due to the narrow connection between their proventriculus and gizzard, which extends the retention time of plastics in the gizzard (Acampora et al. 2016). This results in long plastic retention times of 2–3 weeks, providing more potential for EDC leaching from plastics and subsequent bioaccumulation (van Franeker and Law 2015; van Franeker et al. 2011). Tanaka et al. (2020) in an *in vivo* semi-field feeding study confirmed that marine plastic ingestion in seabirds directly results in high levels of plastic-associated EDC accumulation. Similarly, stomach oils in fulmars were shown to promote the leaching of EDCs from ingested plastics (Kühn and van Franeker 2020). Of the ingested plastic that may be regurgitated, parents often transfer this to their chicks as chicks are fed by parental regurgitation (Tulatz et al. 2023) which initiates a decline in accumulated plastics in adults over the breeding season, while simultaneously exposing the chick to higher loads of plastics and chemicals that may leach out of the plastics in stomachs and intestines (van Franeker et al. 2022). This results in high plastic ingestion in younger birds during sensitive windows of development.

Distribution, breeding and feeding ecology

Fulmars form monogamous pairs, and are colonial, indicating that these birds return to the same nesting site to lay just one egg per pair annually, permitting annual sampling from live birds in summer at their colonies, as well as health checks and gathering of survival and reproductive metrics (Dunnet and Ollason 1978). However, since adults do not necessarily breed every year, it is challenging to reliably sample the same individuals on an annual basis. Fulmars have a widespread distribution over Arctic and boreal zones of the Northern Hemisphere, breeding on cliffs of the Arctic,

Europe, North America, and Asia (Mallory et al. 2020b). This contrasts with 250 years ago, when colonies in the temperate climate zones were rare. Their southern spread to boreal regions such as the UK, Ireland and even France, has been attributed to increasing availability of discarded fish and offal from commercial fisheries in these areas (Thompson 2006). The large distance over which fulmars disperse during their wintering period provides a vast marine area over which large-scale differences in marine plastic pollution levels may be examined to understand its health effects (Edwards et al. 2016; Grissot et al. 2020). It is widely accepted that wide-ranging marine predators such as the fulmar might encounter relatively large spatial variation in EDC distribution (Quinn et al. 2016). This indicates that health risk may vary across geographical locations in line with exposure patterns.

During their breeding period, from April to August, fulmars largely remain near their colony. Although geolocation tracking efforts at a colony on Eynhallow island, in Scotland, revealed fulmars' ability to travel 3000 km from their colony between courtship and egg laying (Edwards 2015; Edwards et al. 2016), such distances are unusual during the breeding period. Egg laying occurs at the end of May, and incubation occurs for approximately 53 days until the chick hatches, until approximately 45 days when fledging occurs (Robinson 2005).

The non-breeding period is from September to March, which is when fulmars spend most of their time at sea. Historically, this period of the year is more difficult to characterize, due to the remote oceanic locations in which these birds spend this period. Fulmars are sea surface feeders that utilize open sea and ocean feeding areas. These birds feed at a variety of levels of the marine food chain and their diet consists of pelagic fish, squid and zooplankton, such as euphausiids, amphipods, and even small copepods (Mallory et al. 2020b).

Fulmars are positioned at high trophic levels rendering exposure to relatively high levels of these chemicals, attributed to bioaccumulation of these compounds. With their ingestion of marine plastics, which are particularly

contaminated with EDCs, these birds are exposed to high levels of EDCs, ranging from sub $\mu\text{g/kg}$ to mg/kg levels (Koelmans et al. 2014; Teuten et al. 2009). In addition, fulmars prefer fatty prey or fatty parts of prey, often selecting to feed on oil-rich livers from offal and jelly fish gonadal tissue, which is where hydrophobic EDCs are most concentrated (Garthe et al. 1996; Monosson et al. 2003). The high levels of exposure and resulting conditions coupled with longevity in arctic and boreal seas suggest that this species might exhibit significant biological responses (J. van Franeker et al. 2005).

Behavior

Fulmars ingest some of the highest levels of marine plastics among marine megafauna (Kühn and van Franeker 2020). This ingestion is in the size ranges from micro- to meso-plastic (Kühn et al. 2022). Long-term studies in the Netherlands indicate that the average mass of plastic particles ingested has decreased since 1980, indicating a trend toward the presence of smaller, yet more numerous, plastic particles (Kühn et al. 2024; van Franeker et al. 2011). Nanoplastic size ranges are apparently, currently unexplored in the species.

Behavioral factors that increase fulmar propensity for plastic ingestion raise their likelihood of mistaking of plastic debris for prey. First, the fulmar surface feeding habits coincide with positively buoyant plastic debris such as polyethylene (PE) and polypropylene (PP), which are the most frequent polymers found in the marine environment (Enders et al. 2015; Geyer et al. 2017; Wessel et al. 2016). These are concentrated in ocean areas called gyres, where ocean currents trap floating debris allowing these areas of open ocean to become hotspots of plastic contamination and ingestion by marine organisms (Høiberg et al. 2022). Further, PE tends to bind more organic contaminants than other plastic polymers (Teuten et al. 2009). Due to limited exchange of marine debris between the hemispheres and higher populations in the Northern Hemisphere, most of the world's marine debris accumulates there. It is noteworthy that this is also where fulmars are found (Lebreton et al. 2012).

Conservation

Fulmars have an average lifespan of 35 years and typically begin breeding when the birds are approximately 9 years old (Dunnet and Ollason 1978). This extended lifespan enables potential buildup of epigenetic changes in response to exposure to plastics. Further, because of their K-strategy for reproduction, fulmar populations recover slowly from declines (Stienen et al. 2007). Following the expansion of fulmar range and population, bird numbers began to decline at colonies such as in Ireland and Scotland from 1984 to 2008, due to diminished reproduction probability and decreased adult survival (Cordes et al. 2015; Lewis et al. 2009). Declines in fulmar populations were also observed in most other colonies across their North Atlantic breeding range, with a 40% fall in breeding populations in the Northeastern Atlantic, although the reason for this reduction is not yet fully understood. These declines were attributed to large-scale processes, such as reduced prey availability from commercial fisheries, bycatch, interaction with anthropogenic stressors, and sea surface temperature changes (Cordes et al. 2015). The species is currently not globally threatened, with a world-wide population of approximately 20 million individuals; however, in Europe fulmars are considered vulnerable, and in Ireland they were identified as a 2020–2026 “Bird of Conservation Concern” (BirdLife International 2018; Gilbert et al. 2021). This highlights the urgency of understanding plastic exposure and its health effects in fulmars.

Characterizing plastic ingestion and EDC bioaccumulation patterns in fulmars

Fulmar-TV% measurements are the key element in the OSPAR and EU-MSFD monitoring approach. The Fulmar-TV% describes the % fulmars with ingested plastic levels exceeding 0.1 g, of which the formal policy target is that it should be reduced to under 10% of the birds. Recently, North Atlantic Fulmar-TV% measurements have been determined. The regions in the Northeastern Atlantic most affected by plastic pollution were found to be at more southern latitudes but seem mostly related to ocean currents (de Bruin et al. 2025; van

Franeker et al. 2022). This pattern was confirmed by other marine pollution monitoring studies (Lebreton et al. 2012; Tekman et al. 2022). However, recent data that more explicitly include the western parts of the North Atlantic show stronger correlations to water currents in relation to source areas of pollution. Currents are reflected in the sea surface Temperatures (de Bruin et al. 2025).

It is important to note that plastic stomach content measurements at the time of death may not necessarily reflect longer term exposure patterns (Kühn et al. 2022). This is important in the context of EDC bioaccumulation, which is a longer-term process. Fulmars tend to have preferred overwintering locations from year to year, which is postulated to indicate that some individuals may obtain higher lifetime exposures than others when feeding in more contaminated areas (Bond 2023).

Both PP and PE occur predominately in stomach contents, but polyvinyl chloride, polystyrene and other plastics are also common (Kühn et al. 2021). Ingested plastics are of consumer and industrial origin, and their shapes vary from sheets to threads, foam, fragments and other formats (van Franeker et al. 2022).

There are some variations in ingestion according to sex and age. Younger fulmars tend to exhibit higher plastic ingestion rates (van Franeker and Meijboom 2002; van Franeker et al. 2011) and females display significantly higher ingestion rates although this difference between the sexes is less consistent across locations (van Franeker et al. 2022). On a positive note, between 2009 and 2018, there was a significant reduction in the amount of plastic found in the stomachs of fulmars from the North Sea. This is considered a consequence of the efforts to reduce plastic mismanagement in recent years by raising stakeholder awareness (van Franeker et al. 2021). Nonetheless, the Fulmar-TV is still being exceeded in all areas of the Northeast Atlantic.

While the link between fulmar and seabird plastic ingestion and bodily contaminant load has been investigated, findings remain inconclusive (Table 1). For instance, Canadian fulmars exposed to plastic did not present with elevated hepatic PCB load (Provencher et al. 2018), and Norwegian studies suggest that plastics may act more as passive samplers of environmental pollutants rather than

direct EDC sources, as evidenced by similar PCB profiles in plastics and fulmar tissues (Herzke et al. 2016). However, Yamashita et al. (2011) noted in short-tailed shearwaters (*Puffinus tenuirostris*), related to fulmars, a positive relationship between plastic ingestion and specific lower-chlorinated PCB congeners ingested plastic. There is a high level of heterogeneity in tissue and compound focus as well as in units reported in investigations (Table 1) (Collard et al. 2022; Herzke et al. 2016; Neumann et al. 2021; Provencher et al. 2018, 2019; Sühling et al. 2022; Trevail et al. 2014). Further, most studies concentrated on the accumulation of persistent organic pollutants (POPs) in fulmar tissues. The accumulation of other plastic-associated and derived EDCs, such as PFAS, phthalates and alkylphenols are less characterized and understood in the species, especially in the context of plastic ingestion (Sühling et al. 2022). In a recent study, although several plastic-related chemicals were detected in fulmar tissues, there was generally no significant relationship between plastic burden and concentrations, highlighting the complexity of chemical exposure pathways in this species (Collard et al. 2024). This leaves a gap in our understanding of how more emerging contaminants are accumulating in fulmar tissues, particularly in relation to plastic exposure. Addressing this knowledge gap may offer new insights into the implications of plastic ingestion.

Evidence of adverse health effects from plastics and their chemicals in fulmars

Although the relationship between plastic-associated EDC bioaccumulation and plastic ingestion is complex, there are significant indicators that plastic exposure is associated with adverse health effects in fulmars. There have been a few indications of ill health in fulmar populations, attributable to plastics (van Franeker and Fulmar Study Group 2011). There was an exceptional mortality event of adult females in 2004 because of a winter-long shortage of food with persistent poor body condition, resulting in long periods of elevated levels of pollutants circulating in the blood, including plastic chemicals (van Franeker and Fulmar Study Group 2011). Females with fully developed internal eggs died, indicating endocrine or

Table 1. Summary of studies quantifying plastic-associated EDCs in wild fulmar tissues and the relationship with ingested plastics. These studies aim to compare momentary “snapshots” of ingested plastic, with the longer-term accumulation of chemicals from a long history of plastic ingestion, often resulting in poor or absent correlations. PCBs: polychlorinated biphenyls; PBDEs: polybrominated diphenyl ethers; HBCDD: hexabromocyclododecane; aBFR: alternative brominated flame retardants; Cl-OPEs: chlorinated organophosphate esters; non-Cl-OPEs: non-chlorinated organophosphate esters and PFAS: per- and polyfluoroalkyl substances. Only samples collected after 2011 are included. Ranges with maximums outside the limit of detection (LOD) are not included. Wet weight (ww) and lipid weight (lw) EDC concentrations are included. Ordered by sample type.

Plastic-associated EDC type	Sample type (s)	Location	Sampling year (s)	Plastic-associated EDC concentrations (by tissue type)	Relationship with plastic	Sources
Phthalates	Preen oil	Canada (Newfoundland and Labrador)	2015	All phthalates were undetectable.	No relationship.	Provencher et al. (2019)
PBDEs, HBCDD, aBFR, Cl-OPEs, non-Cl-OPEs, PFAS	Fat, muscle, liver, brain	High and Sub Arctic Navanut and Labrador Sea, (Canada)	2015	Cl-OPEs in brain: 0.14–0.98 ng g ⁻¹ ww, fat: 0.62–6.30 ng g ⁻¹ ww, muscle: 0.12–0.80 ng g ⁻¹ ww. Non-Cl-OPEs in brain: <LOD – 2.70 ng g ⁻¹ ww, fat: 1.60–34.0 ng g ⁻¹ ww, muscle: <LOD – 1.2 ng g ⁻¹ ww. PBDEs in brain: LOD – 0.22 ng g ⁻¹ ww, fat: <LOD – 32.0 ng g ⁻¹ ww, muscle: <LOD – 0.65 ng g ⁻¹ ww. HBCDD in muscle: <LOD – 2.20 ng g ⁻¹ ww. aBFR in muscle: <LOD – 0.14 ng g ⁻¹ ww. PFAS in liver: 40.0–206 ng g ⁻¹ ww. PCB in muscle: 495–914 pg g ⁻¹ ww, liver: 763–999 pg g ⁻¹ ww.	A significant positive correlation was found between both Cl-OPE and non-Cl-OPE concentrations and the mass of ingested plastics. No significant relationship with plastic was found for other compounds.	Sühling et al. (2022)
PCBs, PBDEs	Muscle and liver	Norway	2012–2013	PBDE in muscle: 0.59–1.10 pg g ⁻¹ ww, liver: 0.93–1.17 pg g ⁻¹ ww. Younger chicks: 1.30 ± 0.69 ng g ⁻¹ ww. Older chicks: 0.76 ± 0.61 ng g ⁻¹ ww.	No significant relationships, suggesting that plastic acts as passive vector for contaminants.	(Herzke et al. 2016)
PBDEs	Liver (chicks-old and young)	The Faroe Islands	2020		No significant relationship.	(Collard, et al. 2022)
PBDEs	Liver (bycaught (with and without ingested plastic) and beached fulmars (with ingested plastic))	Norway (North and South)	2013–2016	Bycaught without ingested plastics: 0.50–8.90 ng g ⁻¹ lw. Bycaught with ingested plastics: 10.0–41.6 ng g ⁻¹ lw. Beached: 145–2913 ng g ⁻¹ lw.	There were higher PBDE concentrations in birds that had ingested plastics compared to those that had not (positive relationship). BDE209 was found in ingested plastic and livers of birds with ingested plastic only. Although contaminant burden increased with plastic load, no significant relationships were found.	Neumann et al. (2021)
PCBs, PBDEs	Liver (with and without ingested plastic)	Svalbard and Iceland	2013–2014	PCBs without ingested plastics: 674 ± 81 ng g ⁻¹ , and with high levels of ingested plastic: 753 ± 194 ng g ⁻¹ . PBDEs in fulmars with no plastic: 1.4 ± 0.3 ng g ⁻¹ , and with high levels of ingested plastic: 2.1 ± 0.6 ng g ⁻¹ . 2427.13 – 27,806.48 ng g ⁻¹ lw.		Treval et al. (2014)
PCBs	Liver	Newfoundland and Labrador Sea (Canada)	2015	PBDE209 in liver: 1.36 ± 0.65 SE ng g ⁻¹ ww. Dechloranes in liver: 0.38 ± 0.12 ng g ⁻¹ ww. Regarding phthalates measured in plasma, some were undetectable, but MEP was at the highest concentration (6.99 ng mL ⁻¹).	No significant relationship but suggests there are interactions between ingested plastics and PCB exposure.	Provencher et al. (2018)
PBDEs, phthalates, dechloranes	Liver and blood plasma	Svalbard	2020		There was no correlation between contaminants and plastic burden.	Collard et al. (2024)

hormonal disruption was involved, as birds in poor health tend not to reproduce. More recent necropsies of beached Procellariiformes showed that fulmars are more likely to exhibit neoplasia (abnormal and excessive growth of tissue) in the digestive tract than other seabird species, with emaciation being the main cause of death (Daoust et al. 2021). Emaciation might occur as a byproduct of high plastic ingestion, as plastics may induce a false sensation of satiety. Neumann et al. (2021) reported that body condition significantly influenced elevated PBDE levels, with beached birds exhibiting higher levels than bycaught birds (Table 1). These limited studies suggest that plastic exposure is leading to adverse health effects in fulmars. More recently, *in vitro* analyses demonstrated that BPA, BPS, 4-OP, PBDEs, and PCBs activated both fulmar and human estrogen receptors (ESR1 and ESR2), highlighting the similar responsiveness of these receptors to plastic-associated chemicals across species (van Hassel et al. 2025). This indicates fulmar's potential as a sentinel species, albeit further work is needed to assess similarity in responses to other marine species. Further, chemical leachate was produced from some fulmars' plastic stomach contents, showing diverse effects on estrogen receptors. This investigation indicates the importance of consideration of the complex mixture of chemicals in the toxicity posed by plastics, further emphasizing the value ecotoxicological approaches in plastic toxicology.

Practical considerations in utilizing fulmars as sentinels for systemic plastic toxicity

Systems biology approaches are beneficial for understanding the molecular and genomic impacts of plastic-related exposures and bioaccumulation. These approaches leverage large data methodologies to analyze gene regulation and expression changes, which may indicate disturbances in critical biological pathways and cellular signaling systems (Hardiman 2020; Huff et al. 2019; Renaud et al. 2017; Richards et al. 2018). Techniques such as transcriptomic and DNA-m sequencing require well-preserved nucleic acids and a thoroughly annotated genome for accurate gene identification through bioinformatic methods. Oxford Nanopore sequencing enables direct

detection of DNA methylation by measuring base-specific electrical signal shifts as DNA passes through the pore, eliminating the need for bisulfite conversion and allowing long-read, single-molecule methylation profiling (Simpson et al. 2017). This approach is especially powerful for resolving methylation patterns across repetitive regions, haplotypes, and large genomic contexts. A challenge with environmental samples is that they frequently encounter the issue of nucleic acid degradation, particularly with RNA. RNA's instability stems from its single-stranded structure, making it more prone to enzymatic degradation and chemical breakdown than its double-stranded DNA counterpart. Challenging sampling conditions such as fluctuations in temperature, exposure to nuclease enzymes, and presence of contaminants in the environment might rapidly degrade RNA, leading to significant challenges in obtaining high-quality samples necessary for accurate molecular analysis. Consequently, investigators need to employ meticulous collection and preservation methods to mitigate RNA degradation and ensure reliable data for studies involving environmental samples.

In systems biology models, the integration of various routine health bioindicators might vastly enhance our understanding of organismal responses to environmental challenges. Hormonal screening tools in feather tissue serve as an exemplary case of such bioindicators. These tools enable researchers to detect and quantify hormone levels that are crucial for understanding stress responses, reproductive health, and metabolic processes in birds. Metabolomic profiling, for instance, offers insights into the biochemical activities within an organism, reflecting immediate responses to environmental changes. Similarly, microbiome analyses reveal how alterations in external environments impact the internal microbial ecosystems, influencing overall health and disease resistance. Proteomic analysis further complements these models by tracing protein expressions and modifications, which are pivotal in adaptive and stress responses. In addition, examining genetic and epigenetic markers provides understanding of long-term adaptations and heritable changes induced by environmental pressures. Immune system

indicators, including specific antibody levels, offer direct evidence of pathogen exposure or stress, critical for assessing population health in changing habitats. Finally, markers of oxidative stress, such as antioxidants and biomarkers of oxidative damage, indicate the physiological stress levels and may be crucial in evaluating the impact of environmental toxins. Taken together, these bioindicators provide a comprehensive toolkit for researchers to dissect complex biological responses, aiding in conservation efforts and ecological management by predicting how species might respond to future environmental scenarios (Ma et al. 2021; Millar-Wilson et al. 2022).

Procedural considerations for systems biology research in fulmars

Procedural considerations for systems biology ecotoxicological research in bird species such as the fulmar demand careful planning and adherence to specific protocols to ensure the integrity and reliability of data collected. Once a species with a sequenced genome and suitable exposure profile and characteristics is selected, ethical considerations need to be prioritized, with all research activities requiring approval from relevant wildlife and ethical review boards to minimize distress and harm to the birds. Sampling methods, particularly for collecting feathers, blood, or tissue, need to be noninvasive or minimally invasive whenever possible to avoid impacting the birds' health or stress levels. When handling birds, investigators need to be trained in proper capture and restraint techniques to ensure safety and reduce stress for the animals. The preservation and processing of samples, especially those susceptible to rapid degradation like RNA, need to be conducted swiftly and under controlled conditions to maintain sample quality. In addition, the choice of analytical techniques, whether genomics, proteomics, or metabolomics, needs to be aligned with the research goals and specific characteristics of the bird species under study. Finally, data integration and analysis need to consider complex interactions within biological systems, employing robust statistical and computational models to derive meaningful insights from multi-omics datasets. These procedural considerations are crucial when expanding the role of plastic bioindicators to sentinels.

Genome completeness: assembly and annotation

Gene annotation involves assigning locations and functions to genes within sequenced genomes. The Avian Phylogenomics Project assembled genomic data from 48 species, spanning most bird orders. It represents the largest vertebrate phylogenomic project to date and has provided valuable insights into avian evolution (Zhang et al. 2014). Despite fragmented low-coverage genomes, the project predicted 70–80% of avian genes. Prior to this, annotated avian genomes were largely limited to the domestic chicken (International Chicken Genome Sequencing Consortium 2004), turkey (Dalloul et al. 2010), and zebrafinch (Warren et al. 2010), making genomic investigations in other birds challenging. Investigators successfully sequenced and assembled the fulmar genome of *Fulmarus glacialis* (Zhang et al. 2014). Fulmar DNA extracted from a voucher male sample (#137838) at the Natural History Museum of Denmark provided the source material. The genome was sequenced using Illumina short read technology to a depth of approximately 33X. Genome assembly was achieved using the SOAP (Short Oligonucleotide Analysis Package) *de novo* assembler developed at the Beijing Genomics Institute (BGI) and designed to assemble high-quality genomes from short-read sequencing data (Hardiman 2008).

De novo genome assembly involves reconstructing the complete genome of an organism without the need for a reference genome. Table 2 summarizes the statistics that describe the fulmar genome assembly (i.e., gene and feature statistics). The contig N50 and scaffold N50 values are metrics employed to assess the quality of a DNA sequence assembly. A contig is a continuous sequence of DNA, and the Contig N50 is defined as the length of the shortest contig at which the sum of all contigs of that length or longer makes up at least 50% of the total genome. Similarly, the scaffold N50 applies to scaffolds, which are collections of contigs linked together with gaps representing unknown sequences. The scaffold N50 measures the length of the shortest scaffold required to cover 50% of the genome assembly. Higher N50 values generally indicate more complete and contiguous genome assemblies. The current fulmar genome assembly, as reported in 2014 by BGI, includes 57,389 scaffolds but no assembled chromosomes. The total

Table 2. National center for biotechnology information (NCBI) *Fulmarus glacialis* annotation release 100 assembly statistics. Detailed overview of the genome's structure and quality in both NCBI reference sequence database (RefSeq) and NIH genetic sequence database (GenBank). Key metrics include genome size, total ungapped length, number of scaffolds and contigs, scaffold and contig N50 and L50 values, GC content, genome coverage, and assembly level, kb (kilobase) Gb (gigabase).

Assembly statistics	RefSeq	GenBank
Genome size	1.1 Gb	1.1 Gb
Total ungapped length	1.1 Gb	1.1 Gb
Number of scaffolds	57,389	57,389
Scaffold N50	47.2 kb	47.2 kb
Scaffold L50	7,016	7,016
Number of contigs	112,214	112,214
Contig N50	25.9 kb	25.9 kb
Contig L50	13,102	13,102
GC percent	41	41
Genome coverage	33.0x	33.0x
Assembly level	Scaffold	Scaffold

size of these high-quality sequences is approximately 1.14 Gb, with contig and scaffold N50 values of 17 kb and 46 kb, respectively. These metrics demonstrate the assembly's limited contiguity and completeness compared to more refined genomic builds found in model bird species such as the domestic chicken (*Gallus gallus*), which is provided for comparative purposes (Figure 2).

The genome sequence submitted by BGI in September 2014 predicted 13,926 genes, of which 12,056 were protein coding. In October, this was updated by the National Center for Biotechnology Information (NCBI) to 15,409 predicted genes and 15,108 protein coding genes (Table 3). Benchmarking Universal Single-Copy Orthologs (BUSCO) is a tool used to assess the completeness of *de novo* assemblies by comparing them to a set of highly conserved and single-copy genes that are expected to be present in all members of a particular lineage (Manni et al. 2021). A BUSCO completeness score above 90% is considered very robust. The fulmar genome assembly has a completeness of 92.1%, with 91.8% of the expected single-copy orthologs present, 0.3% duplicated, 5.4% fragmented, and 2.5% missing. Currently, the fulmar genome lacks its own dedicated genome assembly in the Ensembl database, which provides comprehensive and integrated access to genomic data for model organisms and species of interest in biomedical research, agriculture, and related fields. This presents several challenges, particularly due

to the lack of high-quality gene annotations, pre-computed orthologs, and compatibility with Ensembl-based bioinformatics such as BioMart. This absence limits access to curated genomic data and automated functional predictions which must be addressed using alternative resources. Despite current limitations in genome completeness, which need to be considered when designing bioinformatic approaches, the annotated fulmar genome remains a valuable resource for HTS-based, systems-level investigations into the biological effects of plastic exposure. By aligning transcriptomic and methylomic data to this reference genome, one can systematically assess gene regulation and expression changes, enabling the identification of plastic-induced alterations in key biological pathways.

Health biomarkers

Functional validation of sequencing data is often necessary and may be achieved utilizing quantitative polymerase chain reaction (qPCR), which measures RNA transcript abundance in biological samples. Potential targets for validation include oxidative stress markers, prolactin, and other biomarkers associated with plastic-derived EDCs. In fulmars, hormone levels may be assessed using feathers, which serve as stable matrices that retrospectively integrate circulating hormones over feather growth, providing insight into physiological responses over time. For adult breeding fulmars, molt of primaries and tail feathers is from September to December, in their post-breeding period (Grissot et al. 2020). Due to the stability of the hormones within the feather matrix, these compounds typically do not degrade over time, even in suboptimal storage conditions typical of ecological sampling. Corticosterone analyses are typically used to assess integrated stress levels over a 2–3 week period comprising feather growth (Abbasi et al. 2017; Eulaers et al. 2011). Feather corticosterone is frequently associated with body condition and environmental factors (Jenni-Eiermann et al. 2022). However, comparing hormone levels in feathers with stomach content data is challenging due to the temporal disconnect between the two measures. Feather hormone integration may occur well before the

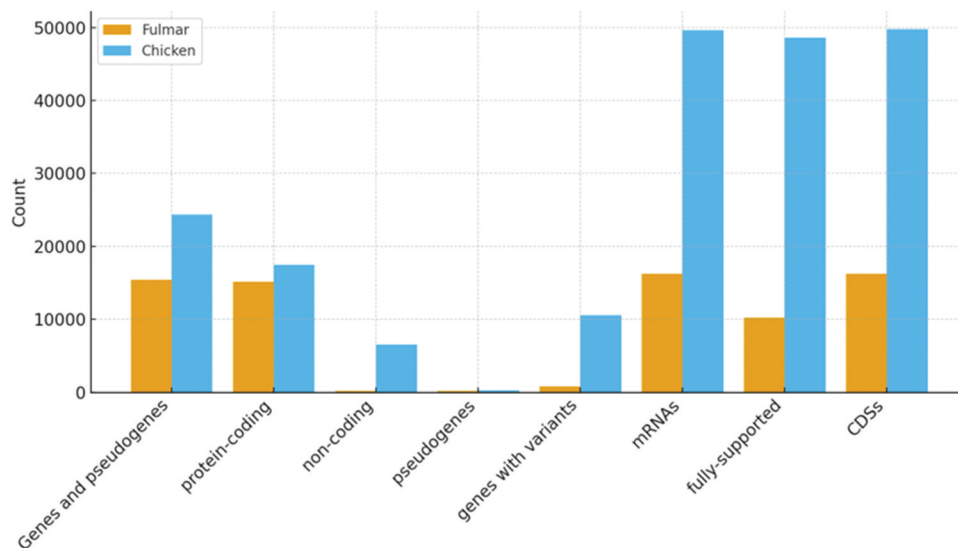


Figure 2. Comparative genome annotation statistics for northern fulmar and chicken. Bar plot comparing annotated genomic features between the northern fulmar (*Fulmarus glacialis*) and the domestic chicken (*Gallus gallus*). Categories include total genes and pseudogenes, protein-coding genes, non-coding genes, pseudogenes, genes with annotated variants, mRNAs, fully supported transcripts, and coding DNA sequences (CDSs). Chicken exhibits substantially higher counts across most categories, reflecting deeper annotation and available genomic resources, whereas the Fulmar genome contains fewer annotated non-coding genes and transcripts. CDSs represent the protein-coding regions of genes, excluding untranslated regions (UTRs). Data are based on genome annotation summaries for each species.

Table 3. Comparison of NCBI *Fulmarus glacialis* annotation release 100 and *Gallus gallus* annotation release 104. Genes and features statistics. includes data on the number and types of genes and pseudogenes, protein-coding genes, non-coding genes, pseudogenes, genes with variants, mRNAs, and CDSs.

Gene and feature statistics	Fulmar	Chicken
Genes and pseudogenes	15,409	24,373
protein-coding	15,119	17,477
non-coding	139	6,534
pseudogenes	151	262
genes with variants	774	10,544
mRNAs	16,240	49,661
fully-supported	10,201	48,584
CDSs	16,251	49,772

time of death, whereas stomach contents represent a “snapshot” of recent ingestion. This time is crucial to consider when interpreting the meaning underlying physiological biomarker values and quantities or types of ingested plastic.

Study population considerations

Different tissue types and associated sample metadata are available for assessment of systemic health impacts of plastic ingestion in fulmars, and these tissues are mostly either bycaught environmental samples from dead birds or samples obtained from live birds in

long-term study colonies. Fulmars from long-term studied colonies are protected and thus their bodies, tissues and eggs are generally not readily available for analysis. Regulatory approval and permits are required for the study of live, wild birds, and these prioritize noninvasive sampling so as not to interfere with bird health or behavior, potentially affecting the reliability of molecular analyses (Stansfield 2015). It is important to ensure that experimental groups are balanced where possible in terms of sex and age (Colominas-Ciuró et al. 2024), and that any sex differences exist in the overwintering location are considered when designing research questions.

Body feathers provide a relatively noninvasively obtained tissue matrix for retrospective health biomarker analyses, even from beached fulmars. However, the availability of blood and liver tissue in particular presents opportunities in regard to HTS applications, which enable gene expression and regulation to be explored in fulmars exposed to different levels of marine plastic pollution (Bortolotti et al. 2008; Quinn et al. 2016). Long-term study cohorts such as on Eynhallow island in Northern Scotland, contrasting with bycaught

fulmars from environmental monitoring initiatives such as the “Save the North Sea” project provide distinct yet complementary perspectives on effects of plastic exposure, and are reviewed below. Both populations represent valuable data resources that need to be coupled to better understand the consequences of plastic exposure.

Environmental monitoring program utilizing dead fulmars

Samples from fulmars have been archived as part of studies that measured their plastic stomach contents. The Netherlands serves as a central hub for fulmar research, largely driven by collaborations between van Franeker and Kühn at Wageningen University, boreal region researchers, and fishers across the Northeast Atlantic. This network originated from the “Save the North Sea” initiative, which focused on collecting fulmars that were either found dead along coastlines or unintentionally caught in longline fishing operations. These efforts provided a consistent and geographically broad source of specimens for investigating plastic ingestion and related ecological impacts (J. van Franeker 2004). As the whole body is collected and dissected, key tissues for toxicological assessments become available to researchers, including the liver.

A logistical hurdle inherent within this sample population is the variable time gap between discovery of a bycaught fulmar and the researcher's access to it, which might adversely affect nucleic acid preservation. This challenge has slowed the adoption of HTS techniques on environmental samples; however, the evolving landscape of sequencing and RNA preservation technologies is steadily overcoming these obstacles. Improved methods are now more adept at handling samples with fragmented or low-concentration nucleic acids, significantly boosting the potential for in-depth analysis of this unique species and offering a more comprehensive understanding of the adverse health effects of plastic exposure (Adiconis et al. 2013; Shao et al. 2023).

Importantly, samples obtained from bycaught fulmars facilitate a direct investigation into the impact of plastic ingestion on their health, attributed to plastic stomach contents enumeration as

part of environmental monitoring. As previously noted, the presence of plastic in stomach contents at the time of death may not accurately reflect long-term ingestion patterns. However, if individuals exhibit consistent, sex-related preferences for specific foraging areas, this may imply relatively stable foraging behaviors across years, potentially supporting more reliable interpretations of plastic exposure over time.

Long-term study cohorts

Investigations of epigenetic perturbations in sentinel wildlife populations benefit from samples obtained over a long timeframe with associated meta data to uncover chronic exposure effects and responses to environmental changes. An exemplary potential resource for this is a colony breeding on Eynhallow island, in Orkney, Scotland (Dunnet 1991; Pirotta et al. 2018). Since the 1950s, long-term monitoring at this site included data on population trends, breeding pair counts, individual sex, and breeding status, with the aim of interpreting these metrics in relation to changing environmental conditions (Thompson and Ollason 2001).

Geolocation sensor (GLS) loggers track the geolocation of fulmars during their non-breeding periods, found individual differences in average wintering locations (Edwards et al. 2013; Edwards et al. 2016; Pirotta et al. 2018). Wintering locations overlap with OSPAR areas used for fulmar biomonitoring and might result in differing individual plastic ingestion (de Bruin et al. 2025). Coastal areas and semi-enclosed seas are relatively high in plastic and pollution, with the North Sea being highly contaminated (Tekman et al. 2022). Thus, it is conceivable that Eynhallow fulmars experience elevated plastic ingestion risk according to latitude, as well as proximity to coastal areas.

Sex differences in preferred foraging areas were observed in the Eynhallow colony. Edwardset al. (2016) observed that males and females remained in different marine areas during the pre-laying exodus period, with 68% of females foraging within the Barents and Norwegian Seas (OSPAR Region I), and 60% of males foraging in the North Sea (OSPAR Region II). Further, females made longer foraging trips than males. Similarly, differential marine areas were utilized by females and males over the non-breeding period whereby females

Table 4. Summary of the current bottlenecks limiting the fulmar's utility as a sentinel for the systemic environmental health impacts of plastic exposure. This summary aims to focus efforts to bridge gaps and enable expansion of the fulmar's role.

Bottlenecks	Potential solutions
Incomplete genome annotation, lacking Ensemble genome assembly	More sequencing efforts for the species, or until then, adapted bioinformatic analyses pipelines.
Logistic concerns for sample integrity, such as the time gap between bycatch and freezing of tissues for RNA sequencing efforts.	Ideally, researcher involvement in bycatch events to act quickly, e.g. using RNA preservation measures, otherwise, bioinformatic techniques.
Sex-specific overwintering distributions may obscure the distinction between sex-related and location-related effects.	Increasing sampling efforts.
Unknowns such as the relationship between fulmar EDC responses versus responses in other species may render comparisons difficult.	Increasing research efforts.
The "snapshot" problem, such as in bycatch, which means that ingested plastic at time of death may not represent longer term exposure patterns.	Take into consideration retention time and integrate findings from long-term study cohorts to better understand long-term dynamics.

generally utilized locations further from the colony than males, tending to reside in oceanic locations, whereas males were more concentrated in the North Sea or shelf region (Grissot et al. 2020).

Another hotspot for long-term fulmar study site is in Svalbard, particularly on the island of Spitsbergen, where OSPAR conducts bioindicator sampling. While having less individual-level or longer-term consistent data in comparison to the Eynhallow colony, it provides a wealth of data for broad environmental health assessments (Collard et al. 2022; Tultat et al. 2023). Other fulmar long-term study colonies include Baffin Island, Canada and Little Saltee island, Ireland (Cordes et al. 2015; Mallory et al. 2020b).

In terms of tissue availability, body feathers and small blood volumes from the Eynhallow fulmar population were sampled from individuals during their breeding seasons at the colony over several decades. Both sample types possess the potential to shed light on dynamic DNAm modifications in response to differential long-term plastic exposures. This, along with historical GLS data and related stable isotope data represents vital resources in understanding fulmar ecology, and their health profiles in relation to exposures resulting from their disparate foraging areas across the Northeast Atlantic.

While direct assessment of plastic ingestion in live fulmars remains unfeasible, there are numerous opportunities for research using live individuals. Noninvasive biomonitoring tools applied across the lifespan offer a more comprehensive view of physiological effects of environmental change compared to single time-point analyses, while also providing access to fresh tissues. When integrated with data on survival, reproductive

success, distribution, and age, these samples become highly valuable for elucidating the long-term consequences of plastic ingestion in fulmars.

Conclusions

The strategic recommendations and perspectives presented in this review draw upon marine ecology, toxicology, and systems biology research. A case has been presented that provides an early framework for understanding the ecotoxicological impact of plastics on the fulmar, including practical considerations for systems biology research in this species. Due to northern fulmar's (1) high propensity for plastic ingestion, (2) established role as a bioindicator, (3) long lifespan, (4) useful behavioral and anatomical traits, (5) availability of a reference genome and (6) diverse biological samples, this bird species is uniquely suited for system-level investigations into biological effects attributed to exposure to plastic pollution. Advances in computational toxicology, comparative genomics, and bioinformatics, in conjunction with increasing capacity to sequence environmental samples, now enable molecular-level analyses of plastic-related impacts in this species. These findings inform cross-species risk assessment frameworks and contribute to broader evaluations of environmental health. This study is intended to serve as a practical resource for ecologists and ecotoxicologists aiming to advance the application of fulmars in environmental health research using integrative systems biology approaches. In Table 4, we have summarized bottlenecks currently challenging the use of fulmars as a sentinel for the environmental health impacts of plastics as well as potential

solutions for these bottlenecks, as discussed throughout this review.

A paradigm shift is urgently needed from traditional, passive monitoring focused on characterizing ingested plastics, to dynamic assessments of their biological consequences. Integrating fulmar-based studies with broader ecotoxicological models might enable a more comprehensive investigation into how chronic plastic exposure leads to bioaccumulation of plastic-associated EDCs and elicits molecular responses at epigenetic and transcriptomic levels. Such approaches are essential for clarifying the adverse health impacts attributed to plastic pollution and for enhancing our understanding of the complex interactions between environmental contaminants and biological systems. Ultimately, this approach may contribute to more informed risk assessments, improved conservation strategies, and support the development of regulatory instruments such as a potential International Plastics Treaty.

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ORCID

Christina Biamis  <http://orcid.org/0000-0002-6466-1735>
 Susanne Kühn  <http://orcid.org/0000-0003-1052-7634>
 Ana Payo-Payo  <http://orcid.org/0000-0001-5482-242X>
 Pierre Bize  <http://orcid.org/0000-0002-6759-4371>
 Jan van Franeker  <http://orcid.org/0000-0002-2035-5007>
 Gary Hardiman  <http://orcid.org/0000-0003-4558-0400>

Data availability statement

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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