Elasmobranch (sharks and rays) interaction with plastic pollution from global and local perspectives, via entanglement within anthropogenic debris and synthetic fibre ingestion.

Submitted by Kristian Parton, to the University of Exeter as a thesis for the degree of *Masters by Research* in Biological Sciences, December 2019.

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K.J Parton
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Abstract

Plastic pollution is a known threat to a host of marine organisms across the world. Research in recent years has exposed numerous negative impacts on some of the world's most threatened marine species, including turtles, cetaceans and pinnipeds. The impact of plastic pollution on elasmobranchs, however, has been relatively understudied. Sharks and rays are widely accepted to be two of the most threatened marine species in the oceans, most notably due to anthropogenic impacts including direct fisheries and bycatch. Their relationship with plastic pollution is only now being investigated in further detail. Previous studies have alluded to damaging effects on sharks and rays as a result of plastic pollution but have lacked in wide synthesis of existing information and empirical evidence. In this thesis, the impact of entanglement within and ingestion of plastic is highlighted for sharks and rays both globally and locally in the North-East Atlantic. Chapter one aimed to collect existing information on the occurrence and distribution of elasmobranch entanglement events, using a systematic literature review and novel data collection from social media site "Twitter". Our results highlighted ghost fishing gear to be the most common entangling material for sharks and rays globally, consistent with previous studies on other marine species. The review also highlighted the lack of standardised reporting for elasmobranch entanglement and therefore resulted in the creation of an online entanglement report form for sharks and rays (ShaREN), allowing citizen scientists across the world to report entanglement incidents quickly and efficiently. Chapter two investigated the presence of microplastics and synthetic contaminant particles in four species of demersal shark found in the North-East Atlantic. Almost 70% of sharks analysed contained at least one contaminant particle,

however no significant relationship between size/weight and number of contaminants was identified, although further analysis was recommended. The study highlighted the ubiquity of synthetic fibres such as rayon and viscose, commonly found in clothing items, as contaminants in the marine environment. Chapter two presents the first empirical evidence of microplastic ingestion by UK shark species and highlights the pervasive nature of microplastic pollution off the English coast. While these two threats are unlikely to have significant population impacts on sharks and rays globally, similar to that of direct fisheries and bycatch, they are identified to be of clear animal welfare concern for these species. Entanglement within and ingestion of plastic is symptomatic of a degraded marine environment and highlights the need for policy-makers, scientists and stakeholders to work together to mitigate this issue for all marine species.

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Authors declaration

All aspects of this research were conducted under the regulations and ethical considerations of the University of Exeter, under the guidance of Professor Tamara Galloway and Professor Brendan Godley.

Between October 2017 and January 2019, all shark samples were collected from either Captain Alan Dwan in Penzance or David Seabourne of Seabourne Fish in Penryn. All individuals were collected according to ethical approval from the University of Exeter. Processing of these samples were conducted under the guidance of Professor Tamara Galloway at the University of Exeter, Cornwall Campus and were further processed under FT-IR spectrometry at the University of Exeter, Streatham Campus under the supervision of David Santillo.

All literature searches and collation were conducted by Kristian Parton, as well as manuscript/thesis structuring, writing and formatting. Professor Brendan Godley and Professor Tamara Galloway contributed significantly to these steps, advising and guiding throughout.

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"I think the tide's with us."

-Chief Martin Brody (*Jaws*, 1975)

General Introduction

Plastic Pollution & Anthropogenic Marine Debris

There is no denying the pervasive nature of plastic pollution and anthropogenic debris in the worlds' oceans (Ryan et al. 2009, Cole et al. 2011, Vegter et al. 2014, Jambeck et al. 2015, Galloway et al. 2017). Since its invention in the mid 20th century, plastic has become an integral part of human life (Andrady & Neal 2009, Singh & Sharma 2016). Its wide range of uses, efficiency and durability, paired with its low production cost has resulted in plastic becoming an unavoidable material in everyday life (Ryan et al. 2009). However, it is unfortunately these raw properties that has led to its persistence within the environment, both in terrestrial and marine biomes (Jambeck et al. 2015).

It is estimated that between 4.8-12.7 million tonnes of plastic enter the oceans every year and without adequate mitigation strategies this number is only set to increase in the coming years (Jambeck et al. 2015). Anthropogenic debris in the form of either macro (>5mm) or microplastics (<5mm) has been found in each of the worlds' oceans, from the uninhabitable waters around the Southern Ocean and Antarctic peninsula to the most remote islands of the Pacific (Cincinelli et al. 2017, Naranjo-Elizondo & Cortés 2018, Forrest & Hindell 2018, Lacerda et al. 2019). It has also been discovered in all sections of the water column, from the shallow waters of the epipelagic zone, to the deep-sea trenches of the hadopelagic zone (Derraik 2002, Cole et al. 2011, Fischer et al. 2015, Bond et al. 2018). With no area of water safe from its reach, the scope for its impact in the environment is vast and research is now beginning to decipher a host of negative impacts on vulnerable marine species. These impacts include injury and death via entanglement, internal damage from ingestion, such as choking and injuries to internal organs and finally physiological changes, including suppression of development and alterations to endocrine and immune systems. (Laist 1997, Page et al. 2004, Wilcox et al. 2013, Gall & Thompson 2015, Cole et al. 2015, Gandara e Silva et al. 2016, Botterell et al. 2019).

Plastic pollution & marine organisms

Plastic and anthropogenic debris engages with a plethora of marine organisms across the globe, including the smallest planktons and the largest cetaceans (Fossi et al. 2012, Botterell et al. 2019), there are very few species safe from its reach. Our knowledge on how plastic impacts marine creatures has increased significantly in the last 10 years and advancements in sampling and detection techniques have enabled us to discover marine plastics in species we had previously thought to be relatively unaffected (Barboza & Gimenez 2015, Araujo et al. 2018, Botterell et al. 2019, Markic et al. 2019). There is no doubt that certain marine species are worst affected when compared to others. For example, marine turtles are regularly highlighted in regard to their negative relationship with plastic and other forms of marine debris, with research showing direct consequences on all life stages leading to detrimental population effects via both entanglement and ingestion (Tomás et al. 2002, Chanrachkij et al. 2009, Wilcox et al. 2013, Schuyler et al. 2014, Wilcox et al. 2015, Ryan et al. 2016, Duncan et al. 2017, 2018, 2019). Cetaceans are also widely reported to suffer from both the smallest and largest plastics, particularly through build-up of toxic persistent organic pollutants attached to plastics ingested into their bodies, impacting vital internal body systems such as endocrine and immune response. (Johnson et al. 2005, Fossi et al. 2012, 2014, Germanov et al. 2018, Moore et al. 2019).

These impacts on charismatic megafauna species appear to have come to the forefront of global issues as a result of widespread media reporting of ground-breaking scientific research and education from wildlife documentary programmes – most notably Sir David Attenborough's Blue Planet II series which aired in 2017 (Thompson & Pahl 2018, Schnurr et al. 2018, Jones et al. 2019). Since the release of this documentary series, the general public have begun to put increasing pressure on governments (particularly in the UK) to provide solutions to solve the plastic epidemic and in recent years change is underfoot. In 2015, the 'Carrier bag tax' was rolled out across the UK, with plans to increase the tax from 5p to 10p as early as 2020 (Schnurr et al. 2018, Thomas et al. 2019). In 2018, the ban on manufacture and sale of products containing 'microbeads' came into place in the UK, preventing the deluge of plastics in cosmetic products entering the oceans through our showers and bathroom sinks (Dauvergne 2018, Kentin & Kaarto 2018, Schnurr et al. 2018). This year (2019), has started to see the removal of plastic straws from restaurants and fast-food chains, with an official ban on plastic straws and cotton buds taking place in 2020 (Schnurr et al. 2018, Godfrey 2019, Nagarajan et al. 2019) .

Regardless of these changes, there are still calls for policy makers to tackle the biggest 'plastic polluters' such as single-use plastic packaging and, most notably, fishing gear. It is estimated that at least 640,000 tonnes of fishing gear enters the oceans each year, and this number is likely significantly higher (Macfadyen et al. 2009, Wilcox et al. 2015, Stelfox et al. 2016). It was discovered that at least 46% of the plastic in the "Great Pacific Garbage Patch" was comprised of abandoned or lost fishing equipment (Lebreton et al. 2018), although global estimates report that approximately 10% of all ocean plastics are fishing debris related (Spiritus et al. 2019). When analysing the threat of fishing gear to marine wildlife, it appears to be a double-edged sword. Large swathes of lost gear, coined as "Ghost fishing gear" has the capacity to directly entangle thousands of marine organisms across the world, causing immobility, suffocation and often death (Sazima et al. 2002, Macfadyen et al. 2009, Barreiros & Raykov 2014, Wilcox et al. 2015, Stelfox et al. 2016, Duncan et al. 2017, Parton et al. 2019). While the second threat involves a gradual breakdown of the fibres that comprise the netting into smaller microfibres as a result of wave action, UV radiation and physical abrasion (Joseph et al. 2002, Welden & Cowie 2017). Lost fishing equipment therefore also has the potential to release thousands of fibres over weeks and months (Montarsolo et al. 2018). These fibres, along with their associated toxins can be directly ingested by marine organisms leading to a host of negative physiological impacts (Ivar do Sul & Costa 2014, Cole et al. 2015, Ryan et al. 2016, Gandara e Silva et al. 2016, Pham et al. 2017, Botterell et al. 2019). Our understanding of these physiological impacts on fish species is fairly limited, with most studies broadly reviewing their presence in wild marine fish species

(Possatto et al. 2011, Lusher et al. 2013, Wang et al. 2019). Some studies suggest biological changes including hepatic and oxidative stress, endocrine disruption, changes in metabolism and deteriorations of intestinal structure and function, albeit under laboratory conditions and with varying results (Rochman et al. 2013, 2014, Lu et al. 2016, Pedà et al. 2016, Yazdani et al. 2016, Alomar et al. 2017).

It is not only micro fibres from fishing gear that are a growing issue in the oceans. Anthropogenic synthetic fibres are now commonly found in a wide range of items used by humans, typically textiles and clothing (Napper & Thompson 2016, De Falco et al. 2018). Research has revealed synthetic fibres are released in their thousands when cleaned in washing machines and due to their microscopic size are easily transported from waste water treatment facilities out into the oceans (Napper & Thompson 2016). These fibres are now being identified in numerous water samples around the world and are therefore consequently being ingested by marine species that inhabit these waters (Moore 2008, Woodall et al. 2014, Duncan et al. 2018, Stanton et al. 2019). It is relatively unknown what impacts these fibres may have on species at a cellular level, however if these fibres have associated inorganic pollutants or toxins attached to them (like many true microplastics), similar detrimental physiological impacts may occur (Fossi et al. 2014, Rochman et al. 2014, Germanov et al. 2018).

Overlooked marine species

There are multiple marine species that may often be overlooked in regard to their relationship with plastic pollution. Smaller species such as plankton, crustaceans and molluscs have all been revealed to be negatively affected by plastic pollution (Murray & Cowie 2011, Setälä et al. 2014, Devriese et al. 2015, Naji et al. 2018), many of which have helped pave the way in our understanding of the cellular impacts of microplastics and how these particles transfer between species in the marine food web (Setälä et al. 2014). One species group, that has perhaps surprisingly been overlooked are elasmobranchs. Research on the impact of plastic on elasmobranchs (sharks, skates and rays) is fairly scarce in the scientific literature, with only a handful of studies looking at their relationship with microplastics (Neves et al. 2015, Alomar & Deudero 2017, Fossi et al. 2017, Bernardini et al. 2018, Germanov et al. 2018, Smith 2018, Valente et al. 2019), and even less on their susceptibility towards entanglement in larger plastics and other forms of anthropogenic debris (Laist 1997, Seitz & Poulakis 2006, Wegner & Cartamil 2012, Stelfox et al. 2016, Parton et al. 2019).

Elasmobranchs: Class Chondrichthyes

Elasmobranchs (sharks, skates and rays) are a diverse subclass of Chondrichthyes consisting of around 1200 cartilaginous fishes (Weigmann 2016), although this number is consistently changing as further species are discovered. This diverse group of marine species have a variety of anatomical and behavioural differences, occupying numerous habitat niches in the majority of the worlds' oceans, whilst also having important ecological significance (Knip et al. 2010, Heupel et al. 2014, Roff et al. 2016).

Life histories & Ecological role

Elasmobranchs are generally thought to display K-selected life history strategies, due to their slow growth rate, late maturation and low fecundity (Smith et al. 1998, Lessa et al. 1999, Pardini et al. 2001, Forrest & Walters 2009), although some species do vary from this. They are incredibly diverse in body plan, with rays and skates generally displaying dorsoventral compression (Compagno 1999), whereas sharks take on a fusiform shape, each form specifically adapts them to their habitat and unique ecology (Compagno 1999). Elasmobranchs can be found in a multitude of different habitats including deep-sea demersal habitats, coral reefs, estuaries and pelagic zones (Baum & Myers 2004, Ulrich et al. 2007, Simpfendorfer & Kyne 2009, Heupel et al. 2010). Elasmobranchs are a vital component of healthy marine eco-systems. As apex predators they play an important role in keeping food webs in balance via top-down control (Myers et al. 2007, Baum & Worm 2009), whilst also helping keep their prey populations healthy (Bornatowski et al. 2014, Roff et al. 2016), resulting in knock-on benefits for fisheries. These charismatic megafauna species also play an important role in the tourism industry, with millions of dollars brought in each year across the globe via eco-tourism activities such as diving, sight-seeing and snorkelling excursions (Gallagher & Hammerschlag 2011, Cisneros-Montemayor et al. 2013).

Threats to elasmobranchs

Despite their pivotal role in the worlds' oceans, elasmobranchs are some of the most threatened species in the marine realm, with research suggesting ¼ of all elasmobranch species are threatened with extinction (Dulvy et al. 2008), whilst 25% are listed as data deficient (Camhi et al. 2007). Due to their life history strategies they are generally predisposed to population declines as a result of anthropogenic activities (Stevens et al. 2000, Pearson et al. 2014). Sharks and rays have a wide range of threats, with the two greatest thought to be direct fisheries and bycatch, in which millions of sharks are killed every year (Clarke et al. 2006, Molina & Cooke 2012, Davidson et al. 2016). It is likely that focus on these two major issues has rightly taken research priority, however this consequently may have masked research on other threats including their relationship with plastic pollution and anthropogenic debris.

Plastic pollution two broad threats: Entanglement & Ingestion

Plastic pollution and its relationship with elasmobranchs (as well as most marine species) can broadly fall into two categories: entanglement and ingestion. Entanglement usually consists of an individual becoming trapped or entwined in some form of anthropogenic debris, usually hampering their ability to move and feed or in extreme cases results in their complete immobilisation (Laist 1997, Gall & Thompson 2015, Parton et al. 2019). In some shark species, complete immobilisation can often lead to suffocation due to their inability to pass water across the gills to breathe (Roberts 1975). Entanglement can also lead to severe cuts, abrasions and loss of limbs, further disabling them from performing their usual behaviours (Laist 1997, Gall & Thompson 2015).

The second threat: ingestion, refers to the consumption of plastic debris, either directly from their biotic environment, or indirectly through their prey species. The debris ingested can consist of larger macroplastics, resulting in damage to internal organs, blockages of passageways or false satiation (Farrell & Nelson 2013, Welden & Cowie 2016, Duncan et al. 2018, Nelms et al. 2018). Smaller pieces such as microplastics or microfibres (<5mm in diameter) may also be ingested. These smaller debris items can have a variety of associated toxins attached such as persistent organic pollutants (POPs) which are capable of causing disruptions to the endocrine and reproductive systems, as well as immunosuppression (Geyer et al. 2000, Anselmo et al. 2011, Cole et al. 2015, Gandara e Silva et al. 2016, Botterell et al. 2019).

Population declines of sharks and rays due to entanglement or ingestion could have severe detrimental impacts on the health and well-being of marine organisms and eco-systems (Myers et al. 2007, Ferretti et al. 2008), as well as humans in coastal communities who heavily depend on the oceans for sustenance and survival. It is therefore imperative that we, as scientists, understand the variety of different threats these animals face and the consequences these issues may have on elasmobranch populations, not only locally – but globally. By understanding some of the “lesser-known” threats to elasmobranchs, scientists may be able to put forth mitigation strategies to governments in an attempt to help deal with these issues and prevent further population declines in already threatened elasmobranch species.

Thesis content

Given the current gaps in the scientific knowledge on the impacts of plastic pollution and anthropogenic debris on elasmobranchs, it is an important line of questioning to assess this threat to sharks and rays, not only locally here in the UK, but also globally. This may help facilitate conservation measures for these particularly vulnerable marine fishes.

In **Chapter One** the first systematic literature review on the susceptibility of sharks and rays to entanglement within anthropogenic debris globally was conducted. By using data from the scientific literature and novel data collection from social media site Twitter, it was deciphered to what extent sharks and rays are impacted by entanglement, highlighting particularly vulnerable species and areas in which this threat may be highest, whilst also determining the most common debris types responsible for entanglement.

In **Chapter Two**, for the first time microplastic/microfibre ingestion was investigated in four species of demersal sharks found around the U.K and North-East Atlantic. Differences in the quantity and type of debris ingested between the species was deciphered and whether their diet influences this. If successful, the study will be the first of its kind to empirically show that U.K shark species have ingested microscopic anthropogenic materials.

Chapter One: A global review of shark and ray entanglement in anthropogenic marine debris.

Kristian J. Parton[1*], Tamara S. Galloway[2] , Brendan J. Godley[1]

Addresses

[1] Centre for Ecology & Conservation, School of Biosciences, University of Exeter, Penryn Campus TR10 9EZ, United Kingdom

[2] Biosciences, College of Life and Environmental Sciences, Geoffrey Pope Building, University of Exeter, Stocker Road, Exeter, Devon EX4 4QD, United Kingdom

*Corresponding author: kp336@exeter.ac.uk

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Abstract

Numerous marine taxa become entangled in anthropogenic marine debris, including cartilaginous fishes (Class Chondrichthyes e.g. Elasmobranchs: sharks, skates and rays, Holocephalans: chimaeras). Research that has been conducted on the susceptibility of these taxa to entanglement in marine debris is here reviewed by conducting a systematic literature review complemented by novel data collection from the social media site “Twitter”. The literature review yielded 47 published elasmobranch entanglement events (N = 557 animals) in 26 scientific papers, with 16 different families and 34 species in all three major ocean basins affected. The most commonly reported entangled species were *Scyliorhinus canicula* (lesser spotted dogfish)*, Hydrolagus colliei* (spotted ratfish) and *Squalus acanthias* (spiny dogfish) comprising nearly 60% of total reports (N = 332 animals). The most common entangling objects were ghost fishing gear (74% of animals), followed by polypropylene strapping bands (11% of animals) with other entangling materials such as: circular plastic debris, polythene bags and rubber tyres comprising 1% of total entangled animals. Most cases were from the Pacific and Atlantic Oceans (49% & 46%, respectively) with a bias towards the United States of America (44% of animals), the United Kingdom (30% of animals) and South Africa (10% of animals). While investigating the social media site “Twitter”, 74 cases of elasmobranch entanglement were found, representing 14 families and 26 species with the following species presenting 3 or more tweets regarding entanglement : whale shark (*Rhincodon typus;* 25.3%), great white shark (*Carcharodon carcharias;* 9.8%), lesser spotted dogfish (*Scyliorhinus canicular*; 7%), tiger shark (*Galeocerdo cuvier*; 5.6%), basking shark (*Cetorhinus maximus;* 4.2%) and grey nurse shark (*Carcharias*

Taurus; 4.2%). On Twitter, ghost fishing gear was again the most common entangling material (94.9% of animals), with the majority of entanglement records originating from the Atlantic Ocean (89.4% of total entangled animals). Entanglement in marine debris is symptomatic of a degraded marine environment and is a clear animal welfare issue. Our evidence suggests, however, that this issue is likely a far lesser threat to this taxon than direct or indirect take in marine fisheries. A relative paucity of scientific data on this subject is highlighted and a standardisation of reporting is recommended in an attempt to accurately quantify elasmobranch entanglement risks and locate interaction hotspots.

1. Introduction

1.1 Plastic in the marine environment

Globally, anthropogenic debris in the marine environment is increasing (Derraik 2002), with the majority of debris consisting of plastic materials (Gregory & Ryan 1997, Derraik 2002, Galgani et al. 2015). Plastic is now being found in all sections of the water column, from the epipelagic zone at the surface to the deep sea trenches of the hadopelagic zone, in the all of the world's oceans (Gregory 1996, Derraik 2002, Cole et al. 2011, Fischer et al. 2015, Bond et al. 2018). Plastic is inexpensive to produce, lightweight, durable and efficient in its uses (Ryan et al. 2009). Unfortunately, it is these properties, in conjunction with its disposable nature, rapid consumption by humans and poor waste governance that leads to its presence and persistence in oceans, estimated as taking hundreds of years to degrade (Barnes et al. 2009). Between 4.8-12.7 million tonnes of plastic are estimated to enter the marine environment every year and without appropriate waste management strategies this number could increase substantially in the coming years (Jambeck et al. 2015) .

Marine life engages with plastic in numerous ways, with nearly 700 marine species interactions reported (Gall & Thompson 2015). The major threats of plastic to marine life revolve around ingestion and entanglement (Laist 1997, Cliff et al. 2002, Page et al. 2004, Votier et al. 2011, Barreiros & Raykov 2014, Vegter et al. 2014, Lawson et al. 2015, Nelms et al. 2018), alongside potential wider ecosystem effects such as habitat degradation (Islam & Tanaka 2004, Nelms et al. 2016).

The marine environment is littered with various types of debris that result in the entanglement of elasmobranchs. Broadly these can be categorised into two groups: fishing based sources of debris and other land-based sources of debris (Duncan et al. 2017). Fishing equipment is often lost at sea due to wear and tear over time, or during inclement weather conditions (Gilman, 2015). This equipment can be defined as abandoned, lost or discarded fishing gear (ALDFG) (Gilman 2015, Wilcox et al. 2015). It is also described by the term "ghost fishing gear" which has the potential to passively drift across oceans, often continuing to capture a variety of marine life (Macfadyen et al. 2009, Duncan et al. 2017). The second category encompasses other sources of debris, often items used by humans on land, these include: plastic packing straps, plastic bags and other packaging. These items enter oceans via a number of land based outputs, often as a result of poor waste management facilities (Jambeck et al. 2015)

Fish aggregating devices (FADs) also entangle marine species (Franco et al. 2009, Filmalter et al. 2013, Poisson et al. 2014). FADs are often created in conjunction with tuna purse-seine fisheries in an attempt to attract fish species into a confined area, before encircling them within the purse-seine nets (Fonteneau et al. 2000, Ménard et al. 2000). They are intentionally created to attract large numbers of target species and often indiscriminately entangle larger species of marine fauna including elasmobranchs (Filmalter et al. 2013, Poisson et al. 2014). FADs can be moored to the ocean floor or can be free drifting (DFADs – Drifting Fish Aggregating Devices), equipped with electronic buoys to allow remote monitoring across the ocean (Maufroy et al. 2015). Many DFADs have large quantities of netting hanging several metres below the surface to create drag, ensuring ocean currents determine its direction as opposed to wind (Filmalter et al. 2013).

1.2 Elasmobranchs and anthropogenic debris

Sharks and rays generally display life history traits such as: late maturation (Heppell *et al.* 1996), low reproductive output (Pardini *et al.* 2001) and long life-span (Cailliet *et al.* 2001), making them highly susceptible to overexploitation (Adams 1980; Stevens *et al.* 2000; Pearson *et al.* 2014). Elasmobranchs are one of the most threatened taxa in the marine environment, with 24% of elasmobranch populations considered as threatened with extinction from a variety of anthropogenic threats (Dulvy et al. 2014). Bycatch and targeted shark fisheries pose two of the greatest threats to shark populations across the globe and it is estimated that between 63-273 million sharks are killed annually through a variety of fishing practices, however fully quantifying shark decline and risk of extinction has been challenging, predominately due to a lack of scientific data (Worm et al. 2013).

Entanglement of elasmobranchs in marine debris is relatively understudied within the scientific literature (Stelfox et al. 2016), with only a handful of studies investigating the problems elasmobranchs face with regards to entanglement within plastic pollution (Laist 1997, Seitz & Poulakis 2006, Wegner & Cartamil 2012, Stelfox et al. 2016). Elasmobranchs are suggested to be less vulnerable to plastic pollution than other large marine species, however this could be a consequence of a lack of studies rather than an inherently lower susceptibility (Stelfox et al. 2016). There have, however, been studies investigating the vulnerability of sharks and rays to plastic ingestion, highlighting that large filter-feeding elasmobranchs may be particularly vulnerable to this threat (Fossi et al. 2014, 2017, Germanov et al. 2018). Only a few studies have touched upon the categories of anthropogenic debris that may be most entangling elasmobranchs, with a particular focus on ghost fishing gear (Gilman 2015; Stelfox *et al.* 2016). If elasmobranchs are susceptible to entanglement in anthropogenic debris, this could have potential negative implications on rapidly declining populations. Entanglement can lead to starvation, suffocation, immobilisation and ultimately death (Laist 1997, Gall & Thompson 2015), making this unequivocally an animal welfare issue, if not of conservation relevance.

1.3 Social Media site "Twitter"

A rise in the use of social media in the last 10 years has transformed the ability of participants to document and share information about the natural world. Social media websites have begun to open the eyes of many in regards to some of the threats animals face in the environment, with certain "viral" messages, photos and videos reaching audiences of millions. Websites such as Twitter, Youtube and Facebook have become potential digital scientific databases and researchers are now beginning to use this data to aid their own scientific research (Davies et al. 2012). The social media site Twitter allows users to post messages or "tweets" of up to 280 characters, as well as other accompanying photographs or videos. Those working in the marine sector, alongside members of the public often tweet about marine conservation issues that they experience day to day. This can provide real-time data on issues such as entanglement, strandings, beach debris and bycatch that can be accessed quickly and easily by anyone registered to the website (Shiffman 2018).

In this review "*Elasmobranch entanglement*" is defined as "*the process by which any cartilaginous fish (including sharks, rays and chimaera) becomes entwined or trapped within anthropogenic debris – excluding those bycaught in active fishing gear*". The aim of this study was to: (1) Assess to what extent elasmobranchs are impacted by marine debris by reviewing existing and obtaining new reports of the occurrence and global spatial distribution of elasmobranch entanglement; (2) Gain insights into which families are most at risk whilst also highlighting the ocean basins where elasmobranch entanglement is most prevalent; and (3) Determine the categories of plastic debris that are most impacting elasmobranchs via entanglement.

2. Methods

2.1 Literature review

Between November 2017-May 2018, and again in March 2019, scientific literature was reviewed for records of elasmobranch entanglement in marine anthropogenic debris. ISI Web of Science was searched for the terms: “plastic”, “macroplastic”, “marine debris”, “entanglement”, “entrapment” “ghost nets”, “ghost fishing” and “Fish Aggregating Device”. Each of these terms were paired with: “Chimaera”, “Elasmobranch”, “Shark”, “Ray”, “Stingray”, “Mobula”, “Manta”, “Sawfish” and “Guitarfish”. Most search terms returned with fewer than 30 results, many returned with no results. In total, after filtering for erroneous entries, this resulted in 20 publications. Additionally, the top 200 search results for these terms on google scholar (for each decade between 1940 and 2019) were scanned to locate any papers that may have been missed in the initial search process; this yielded an additional 6 papers to add to the review. Information on species, location and entangling debris were recorded where possible. The authors note that sawfish are not by definition a marine species of elasmobranch, but are included due to their high susceptibility to entanglement in anthropogenic debris.

Duncan et al (2017) highlight the clear need to differentiate between “entanglement” and “bycatch”. Bycatch is known to be defined as “the unselective catch of either unused or unmanaged species during fishing, with a particular focus on “active” gear. Whereas “ghost gear” can be defined as “when the fisher has lost operational control of the equipment”(Davies *et al.* 2009; Duncan *et al.* 2017; Smolowitz, 1978).

Similarly in this study, only elasmobranchs caught in passive ghost fishing gear were considered to be "entangled" animals, "bycaught" animals were not considered here.

2.2 Twitter search

To complement published work, social media site "Twitter" was searched between 2009 and 2019 (from the first recorded tweet about elasmobranch entanglement) featuring the same terms used in our literature search. 74 Relevant tweets were recorded and investigated further. Again, information on species, location and entangling debris were recorded where possible, directly through the tweet itself or through any other associated images and URL links.

Certain publications reported shark entanglement in DFADs/FADs, however it is unknown whether the shark became passively entangled in the netting as the DFAD was drifting, or whether the sharks became entangled after being encircled in the purse seine nets. Some papers were therefore omitted from this review.

3. Results

3.1 Extent of impact

Research on sharks and rays has steadily been increasing over time. Sharks in particular have become a topic of intense research in the last 30 years, with thousands of papers released yearly (Figure 1A.). Entanglement papers, as a proportion of overall papers on these taxa however, remain relatively low (Figure 1B.).

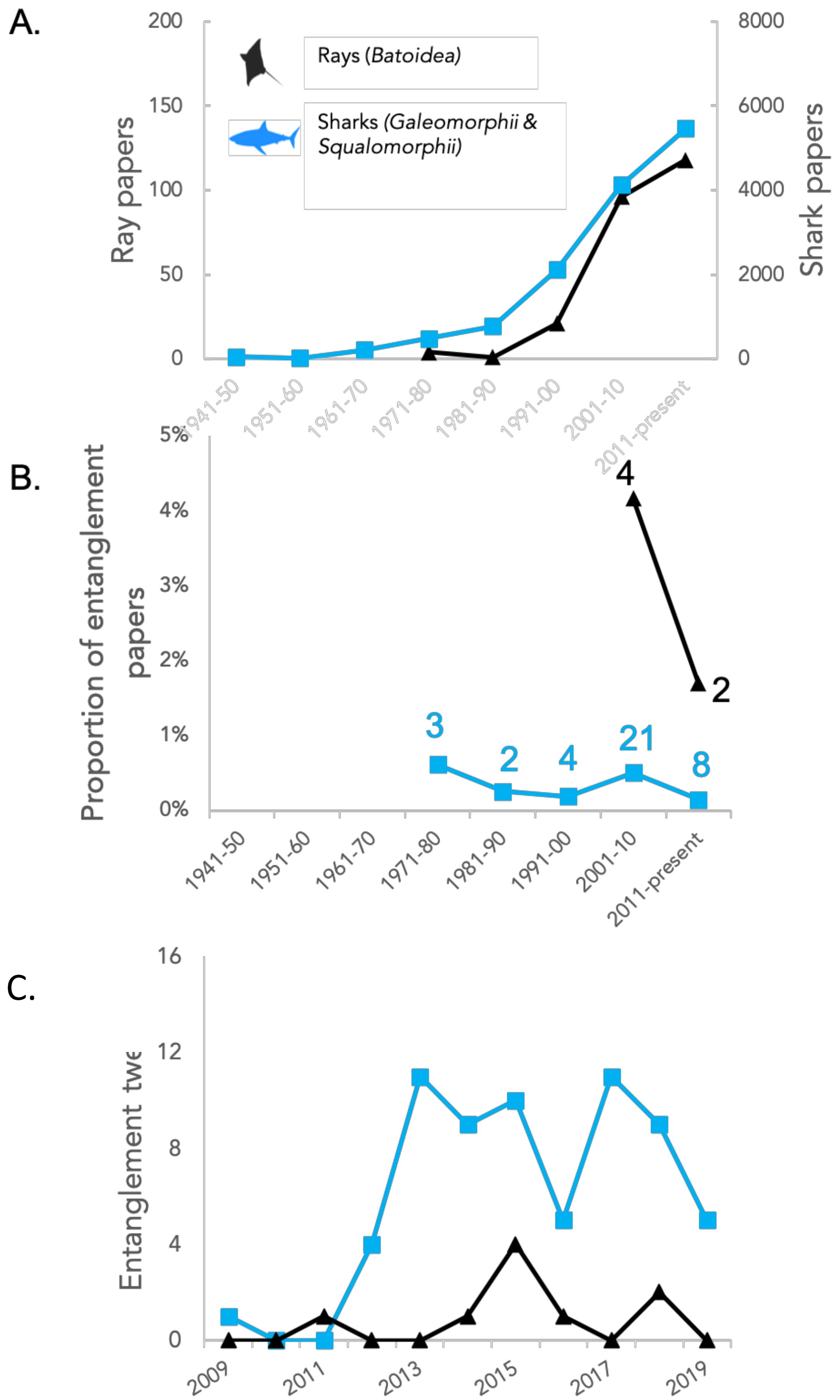

Figure 1: Publication trends. A.) Total number of peer reviewed articles on sharks (*Galeomorphii* & *Squalomorphii*) and rays (*Batoidea*)) from 1941 to present day. Based on web of science searches. B.) Entanglement papers as a proportion of total number of papers on sharks and rays with numbers of publications annotated. C.) Total tweets featuring elasmobranch entanglement from the first recorded elasmobranch entanglement tweet 2009-2019.

Not shown: zero data points and chimaera papers (14 papers from 1981-present, 2 featuring chimaera entanglement).

A total of 47 entanglement events of sharks, rays and chimaera were recorded, encompassing 34 different species (82.9% sharks, 12.7% rays and 4.2% chimaera) from 16 families in 26 scientific publications between 1971 and present day (Table 1.).The most affected species featuring in 3 or more publications were silky sharks (*Carcharhinus falciformis*; 12%) and dusky sharks (*Carcharhinus obscurus*; 12%). Bull sharks (*Carcharhinus leucas*), bluntnose sixgill sharks (*Hexanchus griseus*), great white sharks (*Carcharodon carcharias*), Greenland sharks (*Somniosus microcephalus*), lesser spotted dogfish (*Scyliorhinus canicula*), shortfin mako sharks (*Isurus oxyrinchus*), spiny dogfish (*Squalus acanthias*) and tiger sharks (G*aleocerdo cuvier*) featured in the top 10 entangled shark species each comprising 8% of all entanglement records. A total of 557 animals were found to be entangled with lesser spotted dogfish (21.6%), spotted ratfish (19.1%) and spiny dogfish (19.1%) in the top 3 for most individuals entangled. Leafscale gulper shark (*Centrophorus squamosus*) and Greenland shark (*Somniosus microcephalus*) were reported as tonnes in their respective publications and therefore were omitted from this analysis.

On Twitter, although no incidences of chimaera entanglement were found, a total of 74 different incidences of entangled sharks and rays were recorded, encompassing 26 species, between 2009 and 2019 (Figure 1C, Table 2.). The most reported species with 3 or more records of entanglement included: whale sharks (*Rhincodon typus;* 25.3%), great white sharks (*Carcharodon carcharias;* 9.8%), lesser spotted dogfish (*Scyliorhinus canicular*; 7%), tiger sharks (*Galeocerdo cuvier*; 5.6%), basking sharks (*Cetorhinus maximus;* 4.2%) and grey nurse sharks (*Carcharias Taurus;* 4.2%).

3.2 Entangling materials

Our review found that ghost fishing gear was responsible for over two thirds of all the entanglement records in the published literature for sharks and rays (74% of total animals, N = 412 animals, Figure 2A.). Alongside this, 60% of total entangled animals had their entire body trapped (N = 334 animals), as more often than not when animals are entangled in ghost fishing gear, they become twisted in the material, trapping their entire bodies in the process. Four publications reported elasmobranchs entangled in polypropylene strapping bands (11% of total animals, N = 62 animals), referred to henceforth as PSBs. Our review also revealed the gill region was a common area for sharks to become entangled (Figure 2C.), making up 12% of all entangled animals in the published literature (N = 68 animals).

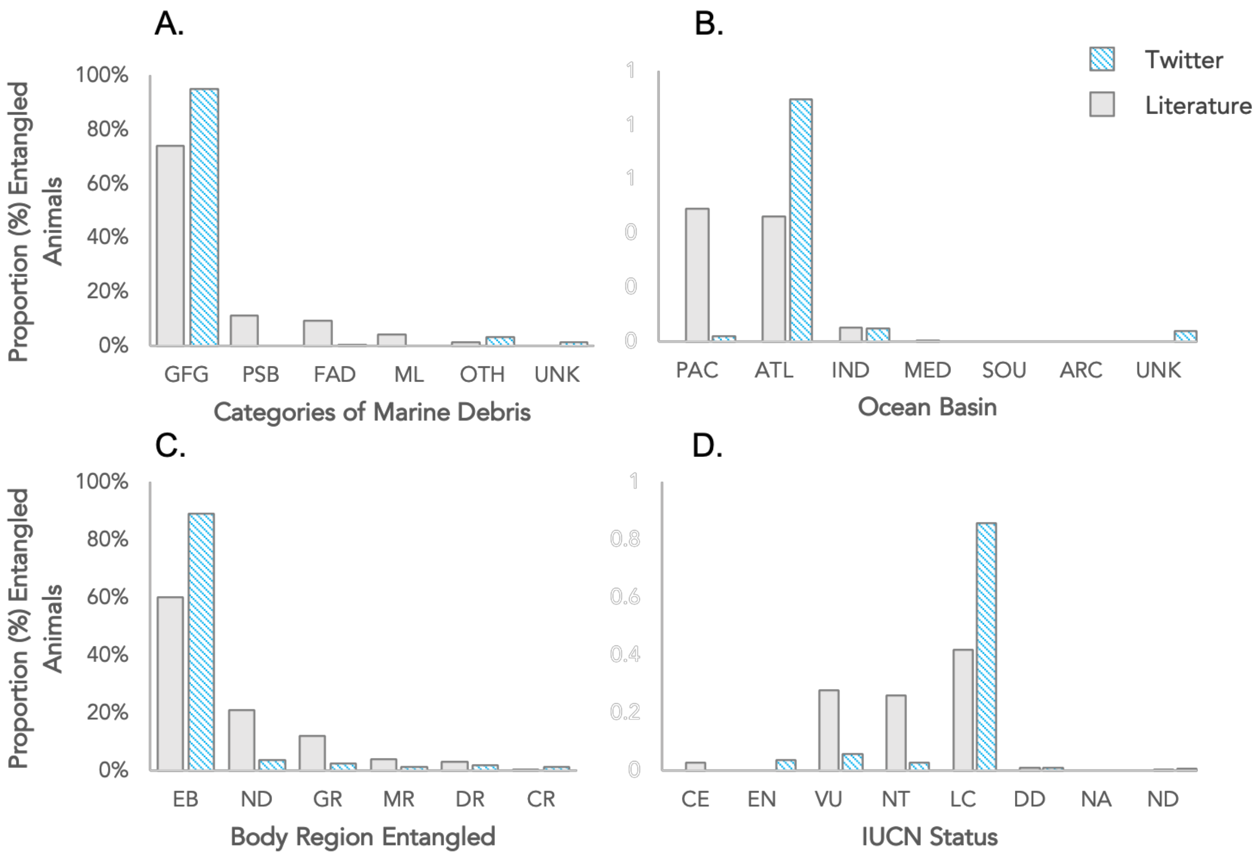

Figure 2: Breakdown of published studies (grey) and Twitter reports (blue) as a proportion of total entangled animals: A.) Categories of marine debris (GFG: Ghost fishing gear, PSB: Polypropylene strapping bands, FADs: Fish aggregating devices, ML: Monofilament line, OTH = Other, UNK = Unknown). B.) Ocean basins (PAC = Pacific, ATL = Atlantic, IND = Indian, MED = Mediterranean, SOU: Southern, ARC = Arctic, UNK = Unknown). Zero cases found in the Southern and Arctic oceans. C.) Region of the body entangled (EB = Entire body, ND = No data, GR = Gill region, MR = Mouth region, DR = Dorsal region, CR = Caudal region). D.) IUCN status of species (CE = Critically endangered, EN = Endangered, VU + Vulnerable, NT = Near threatened, LC = Least concern, DD = Data deficient, NA = Not assessed, ND = No data). Published studies: N=557 animals, Twitter: N=559 animals.

Other land based debris was reported in 6 publications (1% of total animals, N = 8 animals), including that of circular plastic debris (see supplementary Figure 1) which are commonly now found on packs of canned beverages.

On the social media site Twitter, again, ghost fishing gear was responsible for the majority of entanglement records (94.9% N = 531 animals, Figure 2A.). Other forms of debris, including: polythene bags, elastic cords, clothing and SCUBA-diving equipment made up 3.4% of total entangled animals (N = 19 animals). However, in 8 tweets the item causing entanglement was not described, therefore unknown entangling materials made up 1.4% of Twitter entanglement records (N = 8 animals).

3.3 Geographic distribution

Our review found records of elasmobranch entanglement in all but two of the world's oceans: the Arctic and Antarctic/Southern Oceans, of which have only a few reports of elasmobranch species (Long 1992, Campana et al. 2015). The majority of entangled animals in the published literature were found in the Pacific Ocean (Figure 2B. 49% N = 275 animals), with 46% (N = 253 animals) and 5% (N = 28 animals) of entangled animals originating from the Atlantic and Indian Oceans respectively. Areas where large populations of sharks that have been the subject of long-term scientific study appear to feature regularly, particularly in the U.S.A (44% of animals, N = 242). The U.K. (30% of animals, N = 168) and South Africa (10% of animals, N = 53) also feature numerous entanglement reports, albeit from single published papers. Other publications also originated from nations such as Canada and Australia (combined 4% of animals, N = 26) (Figure 3A.).

The majority of entangled animals highlighted from Twitter originated from the Atlantic Ocean (89.4% N = 500 animals), with the Indian and Pacific Oceans featuring significantly less reports of entanglement at 4.8% (N = 27 animals) and 1.9% (N =11 animals), respectively. A small proportion (3.7%) of entanglement records were of unknown origin (N = 21 animals). (Figure 3B.).

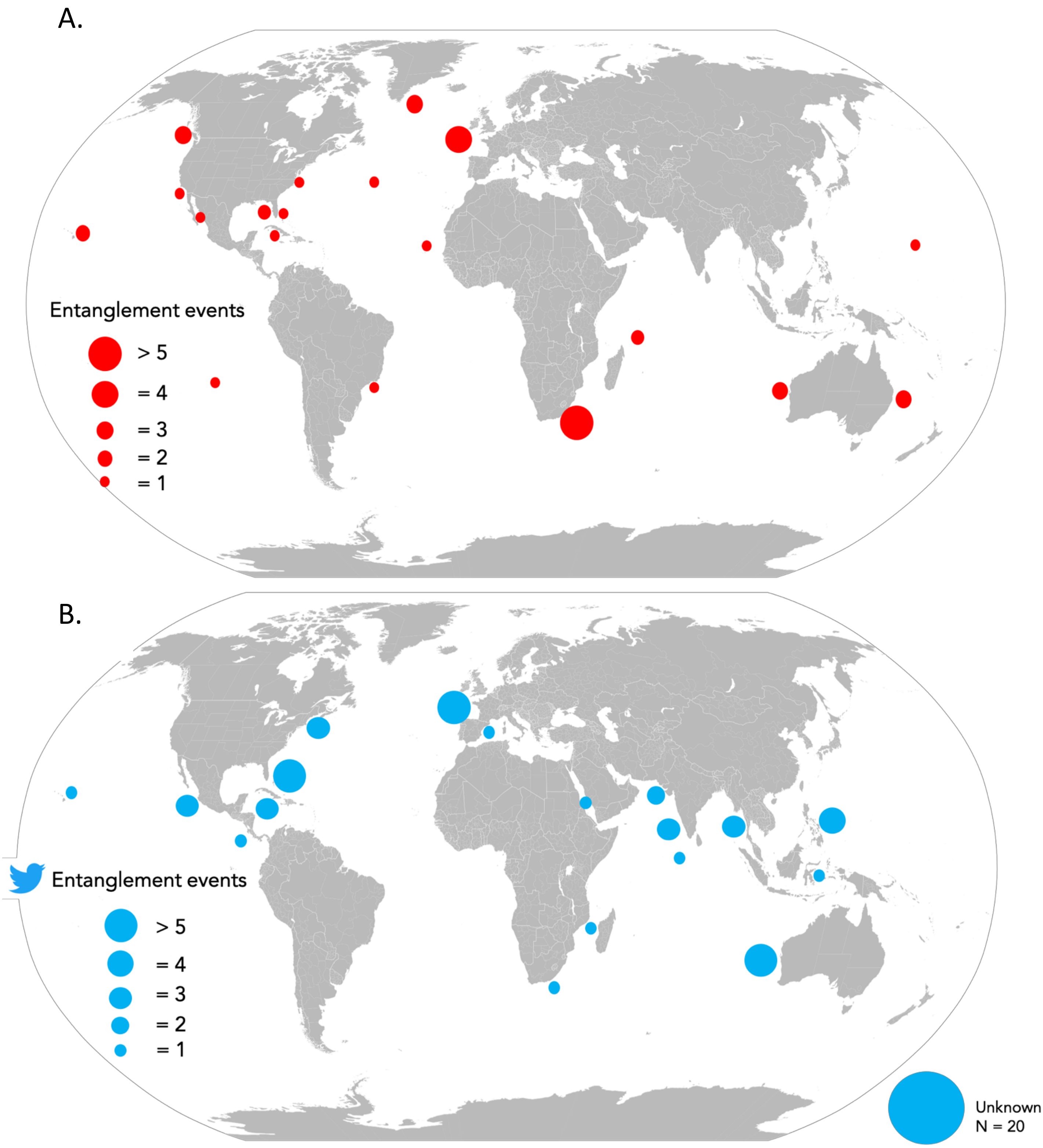

Figure 3: Global distribution of entanglement events from A) published scientific literature. B) distinct tweets from the social media site “Twitter” from 2009-2019. Circles are proportional to magnitude.

Our review found 15 elasmobranch (and 1 chimera) families were impacted by entanglement in anthropogenic debris based on the scientific literature (See Table 1.). 14 elasmobranch families were impacted from social media site Twitter. Combining the published literature with the results from Twitter, a total of 22 different families were impacted: Alopiidae, Carcharhinidae, Centrophoridae, Cetorhinidae, Chimaeridae, Dasyatidae, Ginglymosomatidae, Heterodontidae, Hexanchidae, Lamnidae, Megachasmidae, Mobulidae, Odontaspididae, Orectolobidae, Pristidae, Rajidae, Rhincodontidae, Scyliorhinidae, Somniosidae, Sphyrnidae, Squalidae and Triakidae. The families more commonly impacted by entanglement are: the houndsharks (Triakidae 2/46 species, 467 individuals entangled), the catsharks (Scyliorhinidae 2/148 species, 180 individuals), the requiem sharks (Carcharhinidae, 19/59 species, 143 individuals), the chimaeras (Chimaeridae, 1/38 species, 106 individuals), the dogfish sharks (Squalidae, 1/28 species, 106 individuals), the whale sharks (Rhincodontidae, 1/1 species, 21 individuals), the sawfish (Pristidae, 2/5 species, 17 individuals), the mobulas (Mobulidae, 2/8 species, 16 individuals), the cow sharks (Hexanchidae, 1/5 species, 14 individuals) and the mackerel / white sharks (Lamnidae, 2/5 species, 13 individuals). For proportional breakdown of entangled species in both the scientific literature and from Twitter, see Figure 4.

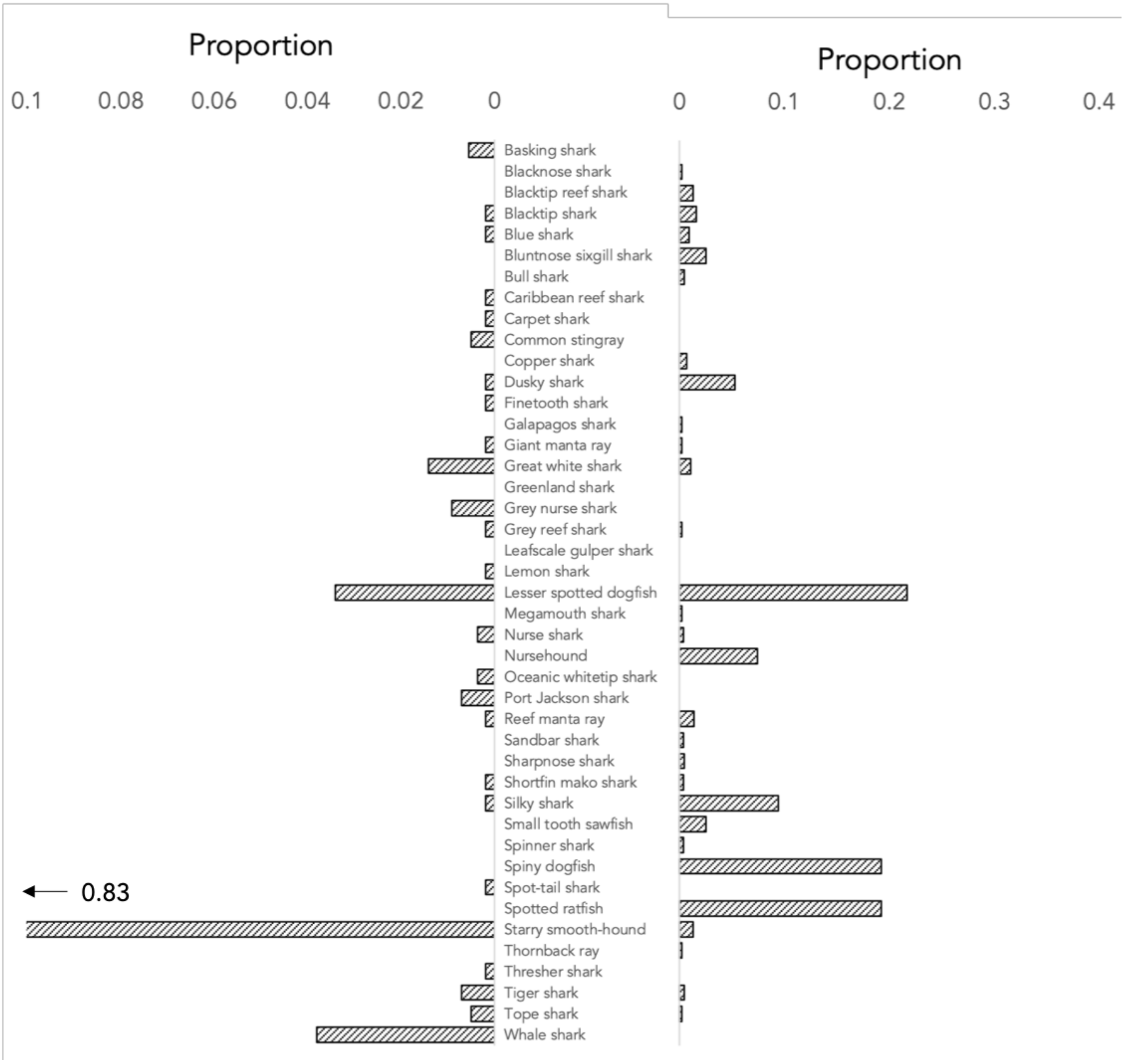

Figure 4: Breakdown of entangled species by: Left: Number of entangled sharks as a proportion of total entangled animals on twitter (N=544) on social media site “Twitter” and Right: Number of entangled animals as a proportion of total individuals entangled in the peer reviewed literature (N=552). Unknown species removed. Greenland shark and Leafscale gulper shark no data points as reported in published paper as “tonnes”. Proportion for “Starry smooth-hound” on Twitter annotated on figure.

4. Discussion

Entanglement in anthropogenic debris is symptomatic of a degraded marine environment. Entanglement of sharks and rays is likely underreported in the scientific literature and has here been identified as a clear animal welfare issue. In conjunction with other threats to elasmobranchs, the issues surrounding entanglement within ghost fishing gear, if not mitigated, may contribute to population concerns for specific elasmobranch families across multiple ocean basins highlighted below.

4.1 Primary drivers of elasmobranch entanglement

The primary drivers for entanglement appear here to be: habitat use, migratory species and body shape/form.

The greatest number of entangled individuals stemmed from the houndsharks (Triakidae) and the catsharks (Scyliorhinidae). These families of sharks are demersal in nature, often feeding on crustaceans and small teleost fishes in benthic habitats up to 200-300m (Ellis et al. 2009, Bengil et al. 2019). In our study, these species were generally entangled in large quantities of ghost fishing gear. Ghost gear when lost at sea can drift for large periods of time, until the weight of entangled species causes it to sink (Phillips 2017, Richardson et al. 2018). Once on the seabed a number of other scavenging marine species become entangled in the netting, consequently attracting predatory demersal elasmobranchs (Kaiser et al. 1996). The use of a demersal habitat, may predispose these sharks to entanglement.

The Carcharhinid sharks were one of the worst affected families, this is likely due to their high abundance, habitat use and mobile nature. (Simpfendorfer & Milward 1993) with many species travelling large distances (100s-1000s kilometres) to feed, breed and give birth (Bonfil et al. 2005, Lea et al. 2015). Although not in the Carcharhinid family of sharks, the same can be applied to whale sharks, basking sharks, white sharks and manta rays. Plastic pollution drifts passively across oceans worldwide (Barnes & Milner 2005, Katsanevakis 2008, Wabnitz & Nichols 2010, Eriksen et al. 2014), therefore species that occupy these oceanic/pelagic habitats may be more likely to become entangled in debris through chance encounters. This could be particularly apparent if they congregate in convergence zones which aggregate large quantities of marine litter (Donohue et al. 2001, Martinez et al. 2009, Law et al. 2014)

The migratory pathways of multiple shark and ray species are now being mapped (Bonfil et al. 2005, Skomal et al. 2009, 2017, Block et al. 2011, Campana et al. 2011, Carlisle et al. 2012, Werry et al. 2014, Jaine et al. 2014, Thorrold et al. 2014, Braun et al. 2015, Queiroz et al. 2016, Braccini et al. 2016, Omori & Fisher 2017, Doherty et al. 2017, Gaube et al. 2018). These pathways may overlap with large aggregations of debris, particularly for individuals displaying offshore migratory movements. This overlap is likely as studies have recently highlighted crossover between filter-feeding megafauna habitat use and microplastic hotspots (Germanov et al. 2018). Sharks are also highly inquisitive in their nature (Laist 1997) and often bite objects to determine if they are palatable or not (Hammerschlag et al. 2012, West 2014). Carson (2013) noted 16% of plastic debris items beached in Hawaii showed bite marks from sharks or predatory fish indicating “testing” of materials. Floating patches of plastic would undoubtedly be novel objects in a sharks’ environment and this exploratory behaviour may often be the cause of initial entanglement in anthropogenic debris.

Species with specific body shapes and anatomically protruding appendages also appear to be prone to entanglement. Elasmobranchs that display an elongated body shape may be more prone to entanglement than those that are dorso-ventrally flattened, due to their swimming kinematics and need for continuous forward motion (Lowe 1996, Lauder & Di Santo 2015) This may explain the low number of rays found entangled across both the scientific literature and social media site Twitter. Other species with morphological differences such as the basking sharks’ elongated snout and mobulid rays’ cephalic fins, can easily become encircled or caught by marine debris such as monofilament line or polypropylene strapping bands (Whalley 2012, Stewart et al. 2018). Other species like the sawfishes (Pristidae), have elongated rostrum lined with saw-like teeth which can easily become entwined in monofilament fishing lines and netting. Although not primarily marine in nature, sawfish populations have declined at alarming rates in recent years (Moore 2017, Jabado et al. 2017, White et al. 2017, Leeney et al. 2018), mostly due to direct and indirect fishing pressures. High habitat specificity, morphology and foraging strategies predispose them to entanglement in river and estuarine habitats which are known to be major entrances of marine debris into oceans (Barnes et al. 2009, Smith & Edgar 2014, Rech et al. 2014).

Despite our review being global in view, the relatively low numbers of incidences of entanglement are likely, at least in part, due to under-reporting. This is evidenced by additional species and locations being highlighted on the social media site Twitter, that were not featured in published reports. This could be due to the ease and instant nature of reporting such incidents via Twitter at the click of a button, often directly through a smartphone. Reports from the literature were often anecdotal, therefore providing further in-depth information may not have been at the forefront of the authors' minds. Future efforts in the peer-reviewed literature should aim at providing as much information as possible when entangled elasmobranchs are encountered. Likewise, entanglement reports by members of the public via social media are inconsistent in nature and therefore could benefit from a citizen science platform via a website or smartphone app to aid in the collection, standardisation and organisation of data.

4.2 Types of marine debris leading to entanglement

4.2.1 Ghost fishing gear

Each year approximately 640,000 tons of fishing gear is lost in the world's oceans (Macfadyen et al. 2009, Wilcox et al. 2015), this ghost gear is a well-known threat to numerous marine taxa (Wilcox et al. 2013, Stelfox et al. 2016). Ghost fishing gear commonly consists of synthetic nylon nets that are non-biodegradable and can persist in the ocean for many years (Saldanha et al. 2003, Nelms et al. 2016). It is evident that ghost fishing presents a threat to elasmobranchs in regards to entanglement, with the majority of animals identified from both the scientific literature and Twitter being entangled within ghost nets.

4.2.2 Fish Aggregating Devices

In our review, 3 publications (Filmalter et al. 2013, Poisson et al. 2014, Hutchinson et al. 2015) reported elasmobranchs becoming entangled in DFADs in the Indian Ocean. Within these studies the silky shark (*Carcharhinus falciformis*) was the only shark reported to have been entangled. The silky shark makes up 90% of the elasmobranch bycatch in the tuna purse seine fishery in the Indian Ocean (Gilman 2011), with estimates of between 480,000 and 960,000 silky sharks killed per year by FADs in this ocean (Filmalter et al. 2013). Of these individuals killed, large numbers are in the first three years of their life, indicating juveniles may be significantly impacted (Filmalter et al. 2013). The redesigning of FADs to minimise the use of large quantities of mesh netting is an emerging method in an attempt to reduce entanglement of shark species (Franco et al. 2009, Dagorn et al. 2013), as well as the use of sisal ropes and biodegradable materials (Delgado de Molina et al. 2006, Franco et al. 2012, Filmalter et al. 2013).

One of the difficulties when reviewing publications concerning shark entanglement in DFADs is attempting to ascertain at which point in the process the shark became entangled. As stated in the methods, some papers were omitted from this review as it was not possible to determine whether the sharks were passively entangled, or caught as bycatch. Consequently, the numbers of elasmobranchs reported as entangled in DFADs is highly conservative and may, with clearer data collection, be the major source of entanglement in anthropogenic debris.

4.2.3 *Land based debris*

Polypropylene strapping bands (PSBs) made up 13 of the 19 land-based debris entanglement events in the scientific literature and are commonly used in parcel packaging, or with crates and pallets (Donaldson 1964). They are a rigid form of plastic that can often form a loop capable of encircling marine organisms, particularly around the gill region of sharks. This can have severe impacts on their ability to pass oxygen over the gills and can ultimately lead to suffocation (see supplementary Figure 1). Naturally, it is difficult to ascertain the exact entry of PSBs into oceans, although possible entry points could include rivers, beaches or container ships transporting large boxed goods. Other land-based debris items entangling elasmobranchs included clothing, SCUBA equipment (regulator hose), and plastic packaging.

4.3 Entanglement Hotspots

Plastic pollution has been found in all of the world’s oceans, with many having their own plastic “garbage patch” (Eriksen et al. 2014). The most famous of these is located in the North Pacific gyre (Lebreton et al. 2018). Similar gyres can be found in the South Pacific, as well as the North and South Atlantic. (Eriksen et al. 2014). While the Pacific and Atlantic Oceans contained the greatest numbers of entangled elasmobranchs across both the scientific literature and social media site Twitter, more research is needed to ascertain high risk ocean areas. Mapping debris hotspots alongside elasmobranch migration routes may provide further clarification on species that are expected to be severely impacted.

Although the Indian Ocean did not contain the highest numbers of entangled animals, it is known to suffer from heavy levels of plastic pollution, particularly in coastal areas (Jambeck et al. 2015) and is estimated to have more plastic than the South Atlantic and South Pacific combined (Eriksen et al. 2014). Paired with this problem, it is one of the most biodiverse oceans in the world for marine species, although data on elasmobranchs is somewhat lacking (Dulvy et al. 2008, Romanov et al. 2010, Tittensor et al. 2010, Wafar et al. 2011, Bowen et al. 2013). More research on this topic, alongside a greater understanding of entanglement in DFADs could well reveal the Indian Ocean to be one of the major risk areas for elasmobranch entanglement.

There are several caveats associated with mapping the geographic locations of elasmobranch entanglement in the scientific literature as well as reports from social media site Twitter. There is a known scientific sampling bias towards wealthier nations including the U.S.A. , Canada and the U.K. (May 1997, Momigliano & Harcourt 2014). This may explain the large numbers of entanglement reports originating in the Atlantic Ocean as wealthy countries have the resources to conduct more scientific research. There will also, undoubtedly, be more reports concentrated in areas where there are known elasmobranch populations that feature heavily in the scientific literature, including Australia, South Africa and Florida (Clark & Von Schmidt 1965, Dudley & Simpfendorfer 2006, Heithaus et al. 2007, Reid et al. 2011, Naylor et al. 2012).

4.4 Social media

The use of social media in acquiring data for the natural sciences is yet to be fully explored. Within 140 (and more recently 280) characters, it was usually possible to ascertain the species, location, and type of debris responsible for entanglement. This was aided by the occasional use of photographs uploaded alongside the tweet, or URL links provided within the tweet to enable us to locate information that may not have been provided within the character limit. There were however, several tweets where we were unable to garner all of the information required, the most notable of which being geographic location. Despite this, our searches highlighted 11 different elasmobranch species that had no records of entanglement in peer-reviewed articles. Alarmingly, numerous tweets were found regarding whale shark entanglement, compared to none in the published literature. This emphasizes that entanglement is more than likely impacting a significantly greater number of species on a vastly larger scale than this review has presented.

In using social media as a tool to document the geographical locations of elasmobranch entanglement, it becomes difficult to control for factors such as tourists travelling to diving hotspots in tropically biodiverse coastal areas (Gössling 1999). This may explain the large numbers of entanglement records in the northern Indian Ocean and Indonesia. It is also difficult to control for biases towards more “popular” flagship species which are commonly encountered by members of the public in tourism hotspots, this again may explain the large number of distinct tweets featuring whale sharks and great white sharks.

Overall, the datasets found among social media sites can, at the least, be used to anecdotally document records of entanglement among elasmobranch species. The authors do not suggest the use of social media to be equivalent to that of a systematic literature review, however by investigating the use of specific keywords and hashtags on Twitter, scientists can obtain real-time data on entanglement events for a variety of marine species. To those working in the marine sector, or people who may encounter entangled elasmobranchs it will be important to provide as much information as possible when deciding to post about these issues on social media. The uploading of photos of the entanglement is reccomended if possible, whilst clearly stating the location, species entangled and the debris causing the entanglement. It is also recommend to use relevant hashtags such as #Entanglement, #Elasmobranch and #MarineDebris to allow scientists to locate these posts quickly and efficiently. Social media remains a novel tool for identifying the threat of entanglement and can, if used correctly, provide valuable insights into marine conservation issues (Abreo et al. 2019).

4.5 Future Directions

4.5.1 Differentiation between entanglement and bycatch

To ensure accurate reporting it will be important to distinguish between entangled individuals and bycaught individuals. The low numbers reported in this review could be indicative that entanglement incidents may have been included under the category of bycatch. Bycatch is well understood in regards to threats to elasmobranchs and it remains one of the most frequent threats to sharks globally accounting for 66.9% of shark species reported by the IUCN (Molina & Cooke 2012).

4.5.2 Standardisation of data collection

Our review found a distinct lack of standardisation in the reporting of entanglement of elasmobranchs in anthropogenic debris. Of the available scientific data in the literature, there is no standardisation in the reporting of entanglement incidents. Many incidents are only anecdotally available within studies, usually as an anecdote from a separate study (Bird 1978, Berra & Hutchins 1990, Flores-ramírez et al. 2015). There are examples in studies listing “x” amount of “sharks” or “rays” as entangled, however various data were missing on the species and consequently some of these accounts were not included in the review (Loog-on & Chanrachkij 2003, Ceccarelli 2009) . A standardised method of reporting entanglement incidents would provide valuable scientific data in an attempt to qualitatively and quantitively assess the entanglement of sharks and rays.

Due to this lack of data standardisation it is also currently difficult to assess at what life stages elasmobranchs are most likely to become entangled. There are a handful of accounts of juvenile elasmobranchs being entangled in anthropogenic debris (Sazima et al. 2002, Colmenero et al. 2017). In most incidences no information was available on life stage. Scientists have highlighted the importance of identifying vulnerable life stages of various marine taxa, with juvenile turtles, seals and whales commonly referred to as the most at risk from entanglement (Henderson 2001, Johnson et al. 2005, Mazaris et al. 2005, Duncan et al. 2017). If juvenile elasmobranch species are more susceptible to entanglement in marine anthropogenic debris, this could have important consequences for elasmobranch species at a population level due to lower recruitment rates, particularly those already threatened with extinction (Stevens et al. 2000). As a result of this, when collecting data on entangled elasmobranchs it is recommended that the following information should be included: species, size, sex, ontogenetic phase, number of individuals entangled, debris type causing entanglement and location of entanglement. With this information it will be more likely that scientists can understand the extent of impact on shark and rays worldwide.

Citizen science has grown rapidly in the last two decades leading to an increase in its use in numerous peer-reviewed articles (Bonney et al. 2009, McKinley et al. 2016) and its impact on science cannot be ignored. Therefore, there is the potential for the creation of an online global database of elasmobranch entanglement, possibly run by an NGO, which allows for citizen scientists to upload information on entangled sharks and rays that they have encountered, thus enabling scientists to gather data quickly and efficiently. Alongside this, demographic studies in which rates of entanglement are calculated will aid scientists in implementing mitigation strategies for particularly threatened species of elasmobranch or within problematic areas.

5. Conclusions

The numbers of entangled elasmobranchs reported here are minimal in comparison to the numbers of elasmobranchs caught directly in targeted fisheries or indirectly as bycatch. Nonetheless, there is no doubt that entanglement in anthropogenic debris is an additional threat to sharks and rays. Further research may reveal this threat to be simply an animal welfare issue, rather than having wide ranging population level effects that have conservation implications. It is apparent however, that entanglement within anthropogenic debris from land-based pollution and discarded fishing gear, is a severely underreported threat to sharks and further research will help fill in existing knowledge gaps. The scientific community should work together with the fisheries sector and the general public in an attempt to quantify and understand this threat to a greater extent. Mitigating strategies that target the issues of ghost fishing, land-based pollution and problematic areas within oceans may aid in reducing the risks for declining elasmobranch species.

Table 1: Entanglement records for elasmobranchs from scientific literature. IUCN: CE = Critically endangered, E = Endangered, NT = Near threatened, VU = Vulnerable, LC = Least concern, DD = Data deficient, NA = Not assessed, N/A = Not applicable. Ocean Basin: ATL = Atlantic Ocean, IND = Indian Ocean, PAC = Pacific Ocean, MED = Mediterranean. Debris type: GFG = Ghost fishing gear, ML = Monofilament line, FAD = Fish aggregating device, PSB = Polypropylene strapping bands, OTH = Other entangling materials. Body region: GR = Gill region, EB = Entire body, MR = Mouth region, DR = Dorsal region, CR = Caudal region ND = No data.
- indicates no information available from scientific paper.
" indicates same as above.
ton indicates unit of measurement provided in scientific paper.
N = Number of entangled individuals

Family	*Genus species*	Common Name	IUCN	Ocean Basin	Location of Study	Year of Study	Debris Type	N	Body region	Reference
Carcharhinidae	*Carcharhinus acronotus*	blacknose shark	NT	ATL	North Carolina	1984	ML	1	GR	Schwartz 1984
	Carcharhinus amblyrhynchos	grey reef shark	"	IND	Australian waters	1994-2008	GFG	1	ND	Ceccarelli 2009
	Carcharhinus brachyurus	copper shark	"	ATL	KwaZulu-Natal, South Africa	1978-2000	PSB	4	GR	Cliff et al 2002
	Carcharhinus brevipinna	spinner shark	"	"	KwaZulu-Natal, South Africa	1978-2000	"	2	"	Cliff et al 2002
	Carcharhinus falciformis	silky shark	"	PAC	Western/ Central Pacific Ocean	2012	FAD	37	EB	Hutchinson et al 2015
	"	"	"	IND	Western Indian Ocean	2011-2012	"	11	"	Poisson et al 2014
	"	"	"	"	"	2010-2012	"	4	"	Filmalter et al 2013
	Carcharhinus galapagensis	galapagos shark	"	PAC	Rapa Nui, Easter Island	2017	OTH	1	GR	Thiel et al 2018
	Carcharhinus leucas	bull shark	"	"	KwaZulu-Natal, South Africa	1978-2000	PSB	2	"	Cliff et al 2002
	"	"	"	"	Sarasota, Florida	1975	"	1	"	Bird 1978
	Carcharhinus limbatus	blacktip shark	"	ATL	KwaZulu-Natal, South Africa	1978-2000	"	9	GR	Cliff et al 2002
	Carcharhinus melanopterus	blacktip reef shark	"	IND	Australian waters	1994-2008	GFG	7	ND	Ceccarelli 2009
	Carcharhinus obscurus	dusky shark	VU	ATL	KwaZulu-Natal, South Africa	1978-2000	PSB	27	GR	Cliff et al 2002

	"	"	"	"	North Carolina	1991	"	1	"	Lombardi & Morton 1993
	"	"	"	"	Sarasota, Florida	1975	"	1	"	Bird 1978
	Carcharhinus plumbeus	sandbar shark	VU	"	KwaZulu-Natal, South Africa	1978-2000	PSB	2	GR	Cliff et al 2002
	Galeocerdo cuvier	tiger shark	NT	"	"	"	"	2	"	Cliff et al 2002
	"	"	"	"	Sarasota, Florida	1975	"	1	"	Bird 1978
	Prionace glauca	blue shark	NT	"	Atlantic & Mediterranean	2016	"	5	"	Colmenero et al 2017
	Rhizoprionodon lalandii	sharpnose shark	DD	"	Sao Paulo, Brazil	1999-2001	OTH	3	"	Sazima et al 2002
Centrophoridae	*Centrophorus squamosus*	leafscale gulper shark	NA	ATL	Rockall and Porucupine Banks, Greenland	2005	GFG	6.2 to n	EB	Large et al 2009
Chimaeridae										
	Hydrolagus colliei	spotted ratfish	LC	PAC	Puget Sound, Washington	2008	GFG	3	EB	NS Initiative 2008
	"	"	"	"	"	"	"	103	"	Good et al 2010
Dasyatidae	*Dasyatidae sp.*	stingray sp.	N/A	IND	Australian waters	1994-2008	GFG	1	ND	Ceccarelli 2009
Ginglymostomatidae	*Ginglymostoma cirratum*	nurse shark	DD	ATL	Boa Vista, Cape Verde Islands	2001	GFG	2	EB	Lopez-Jurado et al 2003
Hexanchidae										
	Hexanchus griseus	bluntnose sixgill shark	NT	PAC	Puget Sound, Washington	2008	GFG	1	ND	Good et al 2010
	"	"	"	"	Flora Islets, British Columbia	2001-2002	"	13	DR	Dunbrack & Zielinksii 2005
Lamnidae										
	Carcharodon carcharias	great white shark	VU	ATL	KwaZulu-Natal, South Africa	1978-2000	PSB	5	GR	Cliff et al 2002
	"	"	"	PAC	Santa Maria, Gulf of California	2014	GFG	1	EB	Flores Ramírez et al 2015
	Isurus oxyrinchus	shortfin mako shark	VU	"	San Diego, California	2012	OTH	1	GR	Wegner & Cartamil 2012
	"	"	"	ATL	Cojimar bay, Cuba	1931	"	1	DR	Gudger & Hoffman 1931
Megachasmidae	*Megachasma pelagios*	megamouth shark	LC	PAC	Oahu, Hawaii	1976	OTH	1	EB	Berra & Hutchins 1990

Mobulidae	*Mobula alfredi*	reef manta Ray	"	PAC	Maui, Hawaii	2005-2009	ML	8	MR	Deakos et al 2011
	Mobula birostris	giant manta Ray	VU	IND	Australian waters	1994-2008	OTH	1	ND	Ceccarelli 2009
Pristidae	*Pristis pectinata*	small tooth sawfish	CE	ATL	Florida, USA	1980-2005	ML	14	MR	Seitz & Poulakis 2006
	Pristidae sp.	sawfish	E / CE	IND	Australian waters	1994-2008	GFG	2	ND	Ceccarelli 2009
Rajidae	*Raja clavata*	thornback ray	NT	MED	Turkey	2016	GFG	1	CR	Akyol & Aydin 2018
Scyliorhinidae	*Scyliorhinus canicula*	lesser spotted dogfish	LC	ATL	St. Bride's Bay, Southwest Wales	1995-1996	GFG	120	EB	Kaiser et al 1996
	"	"	"	"	Southwest Wales	1995	"	-	"	Bullimore et al 2001
	Scyliorhinus stellaris	nursehound	NT	"	St. Bride's Bay, Southwest Wales	1995-1996	"	41	"	Kaiser et al 1996
Somniosidae	*Somniosus microcephalus*	Greenland shark	NT	ATL	Greenland	2012	GFG	1	MR	Nielsen et al 2014
	"	"	"	"	Rockall and Porucupine Banks, Greenland	2005	"	1 to n	EB	Large et al 2009
Sphyrnidae	*Sphyrna sp.*	hammerhead shark sp.	N/A	IND	Australian waters	1994-2008	GFG	1	ND	Ceccarelli 2009
Squalidae	*Squalus acanthias*	spiny Dogfish	VU	PAC	Puget Sound, Washington	2008	GFG	3	EB	NS Initiative 2008
	"	"	"	"	"	"	"	103	ND	Good et al 2010
Triakidae	*Galeorhinus galeus*	tope shark	VU	ATL	Boa Vista, Cape Verde Islands	2001	GFG	1	EB	Lopez-Jurado et al 2003
	Mustelus asterias	starry Smooth-hound	LC	"	St. Bride's Bay, Southwest Wales	1995-1996	"	7	"	Kaiser et al 1996

Table 2: Entanglement records for elasmobranchs from social media site Twitter. IUCN: CE = Critically endangered, E = Endangered, NT = Near threatened, VU = Vulnerable, LC = Least concern, DD = Data deficient, NA = Not assessed, N/A = Not applicable. Ocean Basin: ATL = Atlantic Ocean, IND = Indian Ocean, PAC = Pacific Ocean, UNK = Unknown. Debris type: GFG = Ghost fishing gear, ML = Monofilament line, FAD = Fish aggregating device, PSB = Polypropylene strapping bands, OTH = Other entangling materials, UNK = Unknown.
" indicates same as above.
N = Number of entangled individuals

Family	Genus species	Common Name	IUCN	Ocean Basin	Location	Date of Tweet	Debris Type	N	Reference	Date of Access
Alopiidae										
	Alopias pelagicus	pelagic thresher shark	VU	PAC	Philippines	27/02/2019	GFG	1	https://twitter.com	07/03/2019
	Alopias sp.	thresher shark	UNK	UNK	Unknown	15/10/2017	"	1	"	07/03/2019
Carcharhinidae	Carcharhinus amblyrhynchos	grey reef shark	NT	IND	Maldives	01/12/2014	GFG	1	"	07/03/2019
	Carcharhinus falciformis	silky shark	VU	ATL	Cayman Islands	03/03/2019	"	1	"	07/03/2019
	Carcharhinus isodon	finetooth shark	LC	"	Florida, USA	16/10/2018	OTH	1	"	07/03/2019
	Carcharhinus limbatus	blacktip shark	NT	IND	South Africa	07/08/2013	"	1	"	07/01/2018
	"	"	"	ATL	Florida, USA	19/05/2015	GFG	4	"	07/03/2019
	Carcharhinus longimanus	oceanic whitetip shark	VU	"	Cayman Isands	18/04/2018	"	1	"	07/03/2019
	"	"	"	IND	Red Sea	30/10/2018	OTH	1	"	07/03/2019
	Carcharhinus obscurus	dusky shark	"	ATL	Maryland, USA	15/07/2014	"	1	"	07/01/2018
	Carcharhinus perezii	Caribbean reef shark	NT	"	Bahamas	15/04/2015	GFG	1	"	07/01/2018
	"	"	"	"	Cayman Islands	01/03/2019	"	1	"	07/03/2019
	Carcharhinus sorrah	spot-tail shark	"	IND	Pakistan	16/10/2017	OTH	1	"	07/01/2018
	Galeocerdo cuvier	tiger shark	"	PAC	Hawaii, USA	23/05/2016	GFG	1	"	07/01/2018
	"	"	"	IND	Australia	10/03/2016	"	1	"	07/01/2018
	"	"	"	UNK	Unknown	22/12/2018	OTH	1	"	07/03/2019
	"	"	"	IND	Western Australia	23/08/2016	"	1	"	07/03/2019
	Negaprion brevirostris	lemon shark	"	ATL	Bahamas	14/04/2014	GFG	1	"	07/01/2018
	"	"	"	"	Florida, USA	31/12/2017	"	1	"	07/03/2019
	Prionace glauca	blue shark	"	"	United Kingdom	17/07/2015	"	1	"	07/03/2019

Cetorhinidae	*Cetorhinus maximus*	basking shark	VU	ATL	Rhode Island, USA	14/05/2013	GFG	1	"	07/01/2018
	"	"	"	"	Spain	31/05/2015	"	1	"	07/01/2018
	"	"	"	"	Massachusettes, USA	09/06/2017	"	1	"	07/01/2018
Dasyatidae	Dasyatis pastinaca	common stingray	DD	ATL	France	21/06/2018	GFG	3	"	07/03/2019
Ginglymostomatidae	*Ginglymostoma cirratum*	nurse shark	DD	UNK	Unknown	15/08/2013	GFG	1	"	07/01/2018
	"	"	"	ATL	Florida, USA	09/06/2016	"	1	"	07/03/2019
Heterodontidae	Heterodontus portusjacksoni	Port Jackson shark	LC	PAC	NSW, Australia	27/10/2014	GFG	4	"	07/03/2019
Lamnidae	*Carcharodon carcharias*	great white shark	VU	UNK	Unknown	12/03/2012	UNK	1	"	07/01/2018
	"	"	"	"	"	01/03/2013	"	1	"	07/01/2018
	"	"	"	"	"	27/10/2015	GFG	1	"	07/01/2018
	"	"	"	"	"	15/12/2017	OTH	1	"	07/01/2018
	"	"	"	"	"	14/08/2009	"	1	"	07/03/2019
	"	"	"	"	"	06/02/2019	"	2	"	07/03/2019
	"	"	"	ATL	Mexico	30/11/2016	"	1	"	07/01/2018
	Isurus oxyrinchus	shortfin mako shark	"	IND	Australia	17/09/2013	GFG	1	"	07/01/2018
Mobulidae	Mobula alfredi	reef manta ray	VU	IND	Australia	13/01/2015	OTH	1	"	07/03/2019
	Mobula birostris	oceanic manta ray	"	ATL	Mexico	28/09/2018	GFG	1	"	07/03/2019
	Mobula sp.	manta sp.	"	UNK	Unknown	22/12/2014	OTH	1	"	07/03/2019
	"	"	"	PAC	Costa Rica	29/06/2015	GFG	1	"	07/03/2019
	"	"	"	"	Philippines	04/03/2016	"	1	"	07/03/2019
	"	"	"	IND	Australia	26/04/2015	"	1	"	07/03/2019
	"	"	"	UNK	Unknown	15/03/2015	"	1	"	07/03/2019
Odontaspididae	*Carcharias taurus*	grey nurse shark	VU	IND	Australia	06/11/2012	OTH	3	"	07/01/2018
	"	"	"	"	"	26/02/2014	"	1	"	07/01/2018

	″	"	"	"	"	13/10/2014	GFG	1	"	07/01/2018
Orectolobidae	*Cirrhoscyllium japonicum*	carpet shark	DD	UNK	Unknown	30/12/2017	GFG	1	"	07/01/2018
Pristidae	*Pristis sp.*	sawfish sp.	UNK	ATL	Florida, USA	06/06/2011	GFG	1	"	07/03/2019
Rhincodontidae	*Rhincodon typus*	whale shark	EN	IND	India	10/08/2012	GFG	1	"	07/01/2018
	″	"	"	UNK	Unknown	16/11/2012	UNK	1	"	07/01/2018
	″	"	"	IND	India	30/01/2013	GFG	1	"	07/01/2018
	″	"	"	UNK	Unknown	14/02/2013	UNK	1	"	07/01/2018
	″	"	"	IND	Australia	12/06/2013	GFG	1	"	07/01/2018
	″	"	"	PAC	Mexico	08/08/2013	"	1	"	07/01/2018
	″	"	"	IND	Mozambique	13/09/2013	"	1	"	07/01/2018
	″	"	"	"	Thailand	09/03/2014	"	1	"	07/01/2018
	″	"	"	"	"	19/07/2014	"	1	"	07/01/2018
	″	"	"	UNK	Unknown	08/06/2015	"	1	"	07/01/2018
	″	"	"	"	"	22/07/2015	UNK	1	"	07/01/2018
	″	"	"	IND	Thailand	29/01/2017	"	1	"	07/01/2018
	″	"	"	"	India	16/03/2017	GFG	1	"	07/01/2018
	″	"	"	"	Pakistan	21/08/2017	"	1	"	07/01/2018
	″	"	"	"	Indonesia	21/08/2017	"	4	"	07/01/2018
	″	"	"	PAC	Philippines	01/03/2019	FAD	1	"	07/03/2019
	″	"	"	UNK	Unknown	01/12/2017	GFG	1	"	07/01/2018
	″	"	"	PAC	Philippines	12/06/2015	"	1	"	07/03/2019
Scyliorhinidae	*Scyliorhinus canicula*	lesser spotted dogfish	LC	ATL	United Kingdom	16/12/2015	GFG	2	"	07/01/2018
	″	"	"	"	"	14/05/2018	"	14	"	07/03/2019
	″	"	"	"	France	21/06/2018	"	1	"	07/03/2019
	″	"	"	"	United Kingdom	02/09/2018	OTH	1	"	07/03/2019
	″	"	"	UNK	Unknown	01/08/2015	GFG	1	"	07/01/2018
Triakidae	Galeorhinus galeus	tope shark	VU	ATL	France	21/06/2018	GFG	3	"	07/03/2019

	Mustelus asterias	starry smooth-hound	LC	"	"	21/06/2018	"	4 5 6	"		07/03/2019
Unknown	*Unknown*	Unknown	UNK	UNK	Unknown	24/08/2013	UNK	1	"		07/01/2018
	"	"	"	"	"	28/02/2014	"	1	"		07/01/2018

Chapter Two: Investigating the presence of synthetic particles in four demersal sharks species found in the North-East Atlantic.

Authors
Kristian J. Parton*[1], Brendan J. Godley[1], David Santillo[2], Muhammad Tausif[3], Lucy C.M. Omeyer[1], Tamara S. Galloway[4]

Addresses
[1] Centre for Ecology & Conservation, College of Life and Environmental Sciences University of Exeter, Penryn Campus TR10 9EZ, United Kingdom
[2]Greenpeace Research Laboratories, School of Biosciences, Innovation Centre Phase 2, University of Exeter, Exeter, United Kingdom
[3] Textile Technology Research Group, School of Design, University of Leeds, LS2 9JT, Leeds, United Kingdom
[4] Biosciences, College of Life and Environmental Sciences, Geoffrey Pope Building, University of Exeter, Stocker Road, Exeter, Devon EX4 4QD, United Kingdom

*Corresponding author: kp336@exeter.ac.uk

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Abstract

Microplastic pollution is ubiquitous in the marine environment and is ingested by numerous marine species. Sharks are an understudied group regarding their susceptibility to microplastic ingestion. Here the authors provide evidence of microplastic and other anthropogenic fibre ingestion in four demersal sharks species found in the waters of the United Kingdom and investigate whether body burdens of contamination vary according to species, sex or size. Sharks were collected from the North-East Atlantic, UK. Stomachs and digestive tracts of 46 sharks of 4 species were examined and 67% of samples contained at least one contaminant particle. Although the authors acknowledge modest sample size, estimated particle burden increased with body size but did not vary systematically with sex or species. A total of 379 particles were identified, leading to median estimates ranging from 2-7.5 ingested contaminants per animal for the 4 species. The majority were fibrous in nature (95%) and blue (88%) or black (9%) in colour. A subsample of contaminants (N = 62) were subject to FT-IR spectroscopy and polymers identified as: synthetic cellulose (33.3%), polypropylene (25%), polyacrylamides (10%) and polyester (8.3%). The level of risk posed to shark species by this level of contamination is unknown but is likely much less than that for fisheries mortality. This study presents the first empirical evidence and an important baseline for microplastic and anthropogenic fibre ingestion in native UK shark species and highlights the pervasive nature of plastic pollution.

1. Introduction

1.1 Plastics in the marine environment

Research on plastic in the marine environment has accelerated rapidly in the last decade, with numerous publications describing its impact on ecosystems and marine taxa (Ryan et al. 1989, Cole et al. 2011, Denuncio et al. 2011, Eriksen et al. 2014, Jambeck et al. 2015, Galloway & Lewis 2016, Bucci et al. 2019). It is estimated that between 4.8-12.7 million tonnes of plastic enter the oceans every year from a variety of sources (Jambeck et al. 2015). Plastic is a popular material due to its durability, low production cost and efficiency in its uses (Ryan et al. 2009). It is these properties, alongside its often disposable nature that leads to its prevalence in the environment for many years (Barnes et al. 2009).

Microplastics (defined as plastic particles <5mm) (Arthur et al. 2009) are ubiquitous in the marine environment (Koelmans et al. 2014, Sussarellu et al. 2016, Galloway et al. 2017). Despite this knowledge, quantitative assessments of their abundance are still fairly limited (Avio et al. 2017), although some estimates place their abundance at 5.25 trillion particles globally, weighing in at over 250,000 tonnes (Eriksen et al. 2014). Microplastics, in the form of fibres, fragments or beads/spheres, assimilate in the marine ecosystem via multiple avenues. Larger pieces of plastic can disintegrate over time due to UV radiation exposure, wave action and physical abrasion, eventually fragmenting into microscopic particles (Browne et al. 2007). Microplastics are also found in many everyday items used by humans including cosmetic products and can be produced by clothing wear (Napper & Thompson 2016, Hernandez et al. 2017, Carney Almroth et al. 2018, De Falco et al. 2018). These can then reach the oceans via wastewater treatment plants (Murphy et al. 2016).

1.2 Ingestion of microplastics in marine species

Ingestion of microplastics is reported in many marine species including turtles, marine mammals and fish (Denuncio et al. 2011, Schuyler et al. 2014, Neves et al. 2015, Nadal et al. 2016, Duncan et al. 2018, Nelms et al. 2018). Alongside these larger species, microplastics have been reported in invertebrates such as zooplankton and crustaceans (Murray & Cowie 2011, Setälä et al. 2014, Devriese et al. 2015). Our understanding of the impacts of microplastic ingestion is better understood in the latter group, with reports suggesting dose-dependent detrimental effects on feeding behaviour, development, reproduction and lifespan (Cole et al. 2015, Gandara e Silva et al. 2016, Botterell et al. 2019). There have however been some laboratory-based studies on fish species, suggesting oxidative and hepatic stress, as well as alterations to the intestinal and endocrine systems (Rochman et al. 2013, 2014, Lu et al. 2016, Yazdani et al. 2016, Alomar et al. 2017), although results have varied.

1.3 Microplastic ingestion in elasmobranchs

Elasmobranchs are relatively understudied in regards to threats from plastic pollution (Stelfox et al. 2016, Parton et al. 2019), nonetheless their susceptibility to microplastic ingestion has been reported in a handful of scientific publications (Neves et al. 2015, Alomar & Deudero 2017, Fossi et al. 2017, Bernardini et al. 2018, Germanov et al. 2018, Smith 2018, Valente et al. 2019). It is thought that some species of elasmobranch may be at higher risk of microplastic ingestion based on their feeding strategies or habitat use (Germanov et al. 2018). Filter feeding species (such as whale sharks and basking sharks) that occupy habitats which overlap areas with high densities of plastic pollution have been suggested to be at higher risk of microplastic ingestion (Fossi et al. 2014, Germanov et al. 2018, 2019). Many shark species, however, are non-filter feeders, instead feeding on a range of larger organisms such as fish, crustaceans, marine turtles and marine mammals, all of which have records of microplastic ingestion (Murray & Cowie 2011, Neves et al. 2015, Duncan et al. 2018, Nelms et al. 2018). With sharks classified as fish species, microplastic ingestion could be expected to cause physiological changes similar to those already described in bony fish species, although a current lack of knowledge in this area may restrict accurate comparisons. If these changes are transferable between bony fish and sharks, this could present inherent biological risks to already threatened shark species.

1.4 North-East Atlantic demersal elasmobranchs

The North-East Atlantic is home to numerous shark and ray species, including small to medium sized demersal sharks. These species can be found at varying depths from 5-900m (Sims et al. 2001, Sulikowski et al. 2010), most often residing in benthic habitats (Ellis et al. 2009, Fordham et al. 2016). They feed on a wide range of small teleost fishes, crustaceans and cephalopods (Domi et al. 2005, Ellis et al. 2009). Due to their habitat choice they are often caught in demersal fisheries as bycatch, however targeted fisheries for these species also exist (Hammond & Ellis 2004, Revill et al. 2005). The exposure of microplastics to demersal shark species globally, is currently poorly investigated, with only a few reports of plastic ingestion, mostly situated in and around the Mediterranean Sea (Anastasopoulou et al. 2013, Neves et al. 2015, Bellas et al. 2016, Alomar & Deudero 2017, Smith 2018, Valente et al. 2019). There have, however, been multiple studies of plastic ingestion in bony fish in the regions, with ingestion rates varying from 1-47% across the species (Foekema et al. 2013, Neves et al. 2015, Rummel et al. 2016, Lusher et al. 2016, Murphy et al. 2017).

Here, the first detailed comparative study of microplastic ingestion in four shark species in the North-East Atlantic was carried out (small-spotted catshark; *Scyliorhinus canicula*, starry smooth-hound; *Mustelus asterias*, spiny dogfish; *Squalus acanthias* and bull huss; *Scyliorhinus stellaris*). These species were chosen due to their availability as bycatch in local fisheries. Alongside this, all four species are primarily demersal in their habitat choice, therefore studying microplastic ingestion within them may provide insights into contaminant levels for this marine biome and as a result indicate whether these species would be good bio-indicators for marine pollution. The authors hypothesized that there would be differences in contaminant load among species, between sex and among size classes.

2. Materials and Methods

2.1 Collection and dissection of shark samples

The study was conducted in Cornwall, UK using sharks caught as bycatch in a demersal hake fishery, fishing in and around the North-East Atlantic and Celtic Sea (ICES rectangles: VIIg, VIIh and VIIf). Four species of sharks were investigated (Total N = 46), including: small-spotted catshark (*Scyliorhinus canicula*) (n = 12), spiny dogfish (*Squalus acanthias*) (n = 12), starry smooth-hound (*Mustelus asterias)* (n = 12) and bull huss (*Scyliorhinus stellaris*) (n = 10). Standard shark morphometric measurements were taken for each species (for full details see Supplementary Materials).

2.2 Necropsy and analysis

Upon dissection, the entire gastrointestinal tracts were removed (stomach and intestines) and 10ml (20-50% of total volume depending on species) of their contents were removed for analysis and visual inspection of gut contents (see Supplementary Fig. S1). Subsamples of gut contents were taken due to equipment restraints and to reduce work load. Samples were treated with 20% potassium hydroxide (KOH) as recent studies have highlighted its efficacy at digesting fish ingesta (Foekema et al. 2013, Dehaut et al. 2016, Kühn et al. 2017, Bessa et al. 2018) and heated for 48 hours at 60°C to aid digestion of biological materials. Digested samples were filtered and subsequently analysed under a digital stereo microscope (Leica M165C) and classified by type (fibre, fragment or bead) and colour, as well as measured (mm). A subsample of the contaminants identified (including potential fragments and fibres) underwent Fourier Transform Infrared spectroscopy (FT-IR) to gain insights into their polymer make-up and possible origins. Substantial measures were taken to reduce and control for contamination of shark samples throughout laboratory work, including the running of procedural blanks and air-borne contamination blanks at every stage of the necropsy and subsequent analysis (for full details, including quality control and contamination control measures see Supplementary Materials). All methods were carried out in accordance with relevant guidelines and regulations.

All statistical analyses were conducted on raw data. Data were tested for normality using Shapiro-Wilks test. A negative binomial generalised linear model (GLM) was used to investigate the influence of species, sex and individual length on the estimated number of ingested fibres, using the MASS package in R v3.5.1. All combinations of terms were examined and ranked by Akaike's Information Criteria (AIC) using subset selection of the maximal model using the MuMIn package v1.42.1. Top ranked models were defined as models ΔAIC ≤ 2 units of the best supported model, after excluding further models where a simpler model attained stronger weighting (for full details see Supplementary Materials).

3. Results

3.1 Descriptive Statistics

In total, 46 individual sharks were analysed, of which 56.5% were male, although proportion varied across species (Proportion male for individual species: small-spotted catshark 66.6%, starry smooth-hound 25%, spiny dogfish 83.3%, bull huss 50%). Overall, 67.4% of sharks were classified as adults although again, the proportion differed among species (Proportion adult for individual species: small-spotted catshark 75%, starry smooth-hound 66.6%, spiny dogfish 58.3%, bull huss 58.3%).

Almost all particles identified in sharks were classified as fibres, with only two fragments identified, and no beads/spheres found. Of the 46 sharks analysed in this study, samples from 67% (31/46) contained at least one contaminant particle and incidence was relatively consistent across species (small-spotted catshark 66.6%, starry smooth-hound 75%, spiny dogfish 58%, bull huss 70%).

Fibres ranged in length from 0.3mm to 14.4mm and had an average length of 2.7mm ± 2.6 SD (see Figure 1).

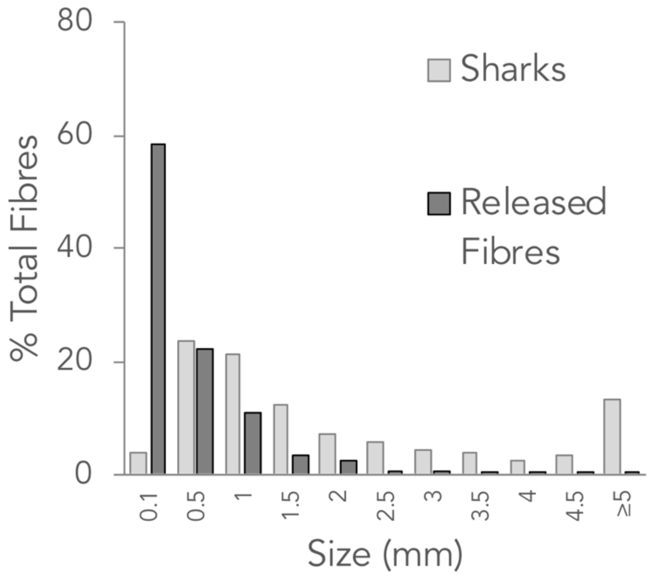


Figure 1: Fibre length distribution. Fibre lengths as a proportion of total fibres for fibres found in shark species (light grey) and fibres released in laboratory conditions after washing of various cotton and polyethylene terephthalate textiles. Palacios Marin AV, (2019) Release of microfibres from comparative common textile structures during laundering (Unpublished Masters dissertation). University of Leeds, UK

The vast majority (88%) of fibres were blue (88.0%) or black (8.8%) in colour, with the remaining colours including: red, yellow and other (clear, green and white) each making up 3.8% (see Figure 2 A-D). The two fragments identified were blue and white in colour. Fibres larger than 5mm (n = 50) were considered here as macroplastics and were excluded from the analysis, although can be found grouped together in the ≥5mm category on Figure 1.

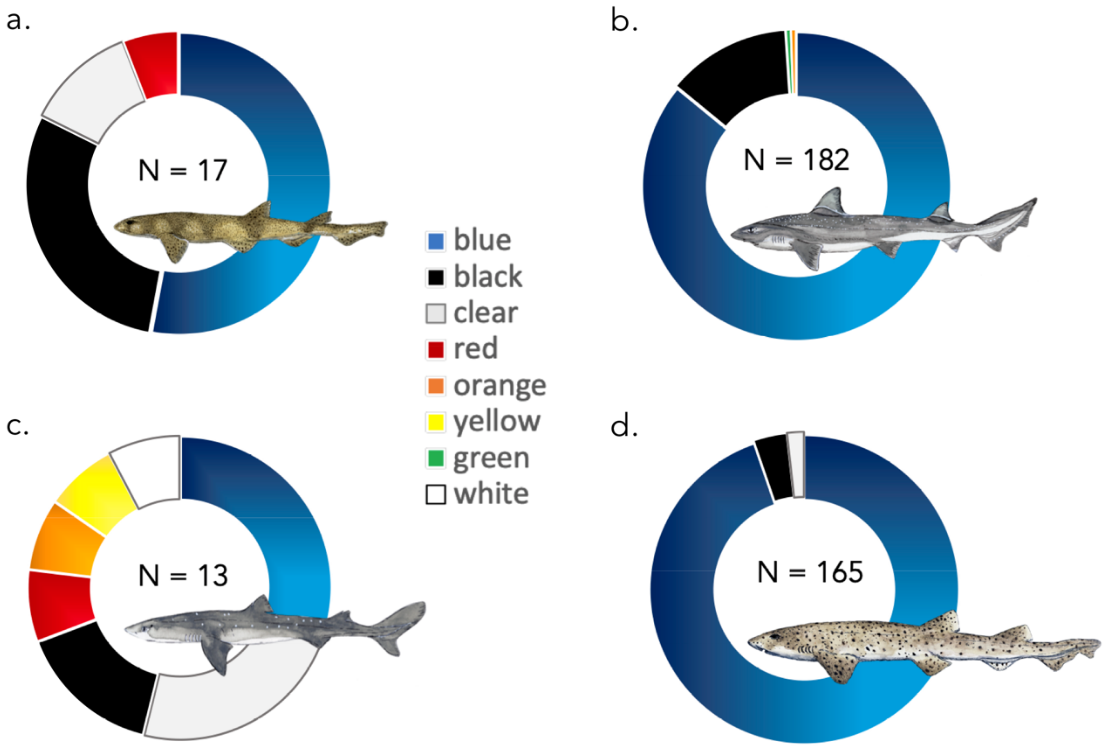


Figure 2: Composition of colours for ingested fibres, found across both the stomachs and intestines of four species of north-east Atlantic demersal sharks: a. small-spotted catshark (*Scyliorhinus canicula*), b. starry smooth-hound (*Mustelus asterias*), c. spiny dogfish (*Squalus acanthias*) and d. bull huss (*Scyliorhinus stellaris*). Total N of coloured fibres identified annotated within figure. Elasmobranch drawings by Lucie Jones.

The estimated number of ingested microfibres was positively influenced by individual shark body length, (see Figure 3 & Supplementary Table S2); (estimated median fibres (IQ range ; range): Overall: 4(0-9 ; 0-770), starry smooth-hound (7.5(3.8-28.75 ; 0-735), small spotted catshark (2(0-4 ; 0-6), spiny dogfish (4(0-4 ; 0-12), bull huss (5(1.3-13.8 ; 0-770). The number of microfibres did not, however, differ between species or sex (See Figure 3, Supplementary Table S2 and Supplementary Fig. S5). It should be noted two individuals in this study (one starry smooth-hound and one bull huss) had much higher levels, with the sample from the former individual containing 147 fibres and the sample from the latter containing 154 fibres. Upon visual examination, these fibres appeared to be strands of blue rope, subsequently confirmed as olefin polypropylene. (Supplementary Figures S4-S8 have been

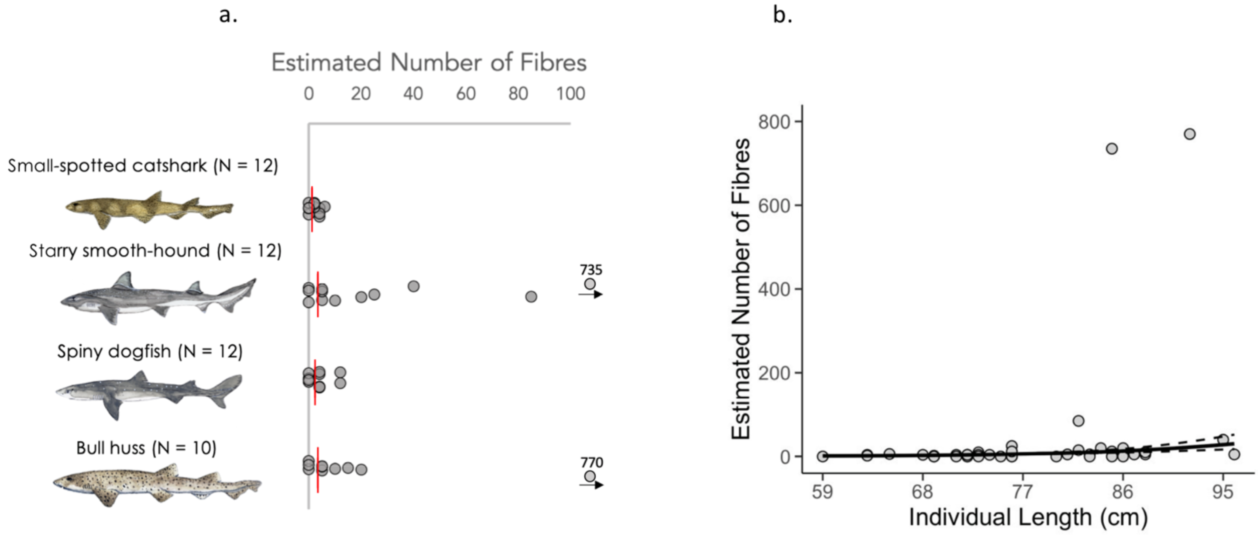


Figure 3: Estimated fibre ingestion & relationship with total length (cm). a.) Expected number of fibres based on extrapolation from full stomach/GI tract volumes. Medians marked by red line. N = annotated. Elasmobranch drawings by Lucie Jones. b.) Relationship between the estimated number of ingested fibres and individual length. Lines denote predictions from the top ranked model presented in Supp Table S2. Standard errors are shown by the dashed lines.

3.3 Polymer Identification.

A subsample of particles (n = 60 fibres, n = 2 fragments) were subject to FT-IR analysis (16% of total contaminants identified). However, when considering the sample set without the two outliers mentioned above which were olefin polypropylene fibres, the subsample of particles that underwent FT-IR spectroscopy equalled 78.9% of all particles isolated (n = 76).

Our analysis revealed 33.3% of fibres (n = 20) were cellulose derivatives (Alpha & Ecteola modified), however further analysis by light microscopy revealed these cellulose fibres were anthropogenic in nature due to their uniform diameter distribution across the fibre length and observation of convoluted structure of the fibre; a characteristic of cotton fibres (see Supplementary Fig. S2). Polyacrylamides made up 10% of fibres (n = 6), 8.3% of fibres were polyesters (n = 5) and 1.7% were cellophane (n = 1). Another 25% (n = 15) registered as Olefin polypropylene. Combined with the aforementioned microplastic contaminants (polyester and polyacrylamide), this results in a total of 43.3% of particles being true microplastics.

The remaining 21.6% of fibres (n = 13) were either unidentifiable due to low spectral match scores (n = 7) or returned as biological in nature (n = 6). Biological returns were excluded from broader statistical analysis. See Figure 4 & Supplementary Table S1.

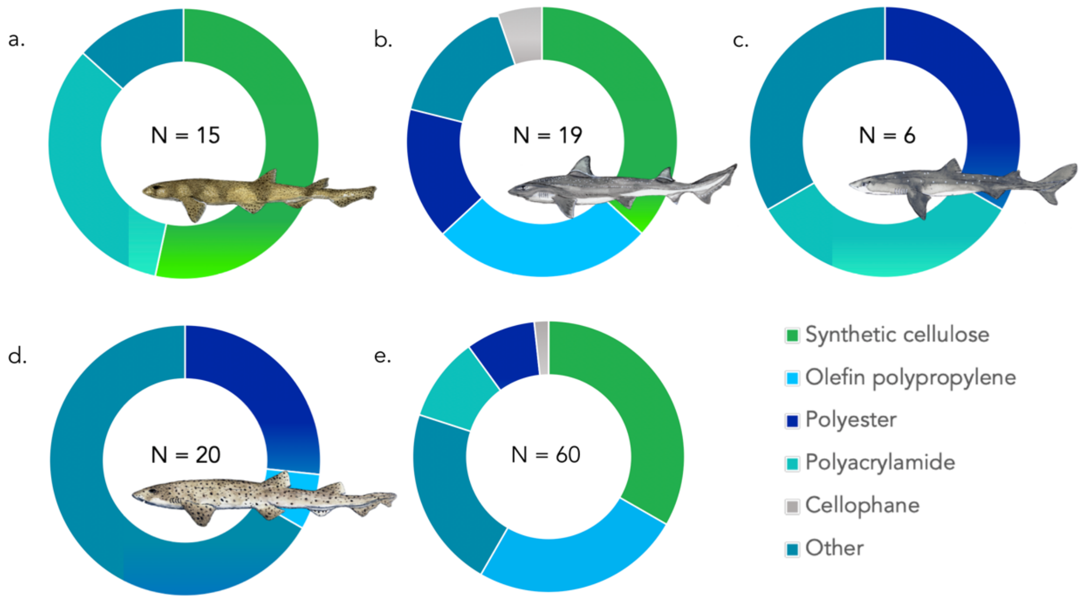


Figure 4: Composition of polymer make up of fibres between shark species. N of polymers identified in each species annotated on figure. a. small-spotted catshark. b. starry smooth-hound. c. spiny dogfish. d. bull huss. e. Total polymer percentages for all four species. Other = Biological materials and/or low spectral match scores. Elasmobranch drawings by Lucie Jones.

The two fragments identified returned as polyethylene and polypropylene (see Supplementary Fig. S3).

4. Discussion

Our study is the first of its kind to demonstrate the presence of microplastic and anthropogenic particle contaminants in resident UK shark species in the North-East Atlantic. Despite there being no substantial differences in microplastic uptake among the shark species studied here, the research provides an important empirical baseline for future work investigating contaminant levels in UK sharks. Greater levels of contamination might be expected in animals that inhabit other parts of the UK with lower water quality. Although no health impacts have been the demonstrated on the sharks, the presence of these particle contaminants indicates their pervasiveness in the marine environment. With increasing global plastic production and its prevalence in every day products, the abundance of such marine pollutants is likely set to increase.

4.1 Synthetic particle ingestion by species, sex and size

Nearly 70% of all sharks sampled in our study contained at least one contaminant particle in their digestive tracts. Although this is likely to be a conservative estimate of incidence, this number is significantly higher than many other reports for similar shark species around the world (Cliff et al. 2002, Anastasopoulou et al. 2013, Deudero & Alomar 2015, Neves et al. 2015, Bernardini et al. 2018, Smith 2018) see Table 1. Studies by Alomar & Deudero (2015) and Smith (2018) revealed ingestion rates of microplastics at 16.8% in blackmouth catsharks sampled in the Mediterranean, and 15% in small-spotted catsharks from the North sea, respectively (Deudero & Alomar 2015, Smith 2018) . Interestingly, the Mediterranean is considered by some to be one of the worst affected oceans with regards to plastic pollution (Eriksen et al. 2014, Cózar et al. 2015, Suaria et al. 2016), therefore ingestion of contaminant particles may have been expected to have been lower in North-East Atlantic. The only other study to have been conducted on similar species and within a similar ocean area is that of Neves (2015), which found microplastic ingestion rates of 20% in small-spotted catsharks collected from the North-East Atlantic coast of Portugal (Neves et al. 2015), with microplastics being mostly fibrous in nature.

The contaminants found within the sharks in this study is consistent with other studies investigating the presence of pollutants in the marine environment (Lusher et al. 2014, Duncan et al. 2018, Gago et al. 2018, Compa et al. 2018), and their colours (Lusher et al. 2014, Steer et al. 2017, Duncan et al. 2018, Gago et al. 2018). Fibres are quickly becoming the most ubiquitous contaminant type in many compartments of marine ecosystems, as well as in the gut contents of numerous marine species including turtles, seals and cetaceans (Lusher 2015, Duncan et al. 2018, Nelms et al. 2018, Zhu et al. 2019). Fibres have a number of potential sources, including break-off from fishing and maritime equipment such as nets and ropes (Welden & Cowie 2017), fibre shedding from automotive tyre wear and the washing of synthetic fabrics in clothing, as well as breakage and release from other textiles (Hartline et al. 2016, Napper & Thompson 2016, Salvador Cesa et al. 2017, De Falco et al. 2018, Wagner et al. 2018).

The authors hypothesised that there would be differences in estimated contaminant load among species, between sexes and across size classes. The expected number of ingested fibres was only influenced by individual length (TL cm) with more found in larger sharks. As location/habitat was unable to be controlled for in this study, this remains to be explored in further detail. While diet could be an additional influencing factor for these shark species, with the current presented data and relatively small sample size, factors influencing contaminant burden in these demersal sharks can only be speculated.

It is interesting to note the relatively high proportion of fibres found here that are described as cellulosic following analysis (approximately 1/3rd of fibres analysed). In previously published studies, these may sometimes be disregarded due to the similarity to naturally occurring cellulose (Lusher et al. 2014, Henry et al. 2019). Their inclusion is believed to be warranted, since stringent analysis by textile fibre experts led to the conclusion that these fibres were of anthropogenic origin, as opposed to naturally occurring cellulose. Microscopic analysis of the visual structure and appearance of these fibres indicated their origins to be from textiles or clothing, including fibre types such as viscose, rayon, cotton and lyocell.

Due to the strict quality control that was applied during the processing of the samples, the presence of these anthropogenic fibres through laboratory contamination is unlikely and hence they have been included in the statistical analysis. If the synthetic cellulosics counts were to have been removed, it would reduce the total fibre count by approximately 33% (just over 120 fibres). This does have the potential to alter some of the statistical relationships found in the results and this must be considered within the broader context of the study and when making comparisons to previous research. Regardless, inclusion of these fibres in the results is important to enable future researchers in their understanding of the multitude of anthropogenic fibres found in the oceans, not just traditional petrochemical-derived polymer compounds. This is likely to become more important as alternative biopolymers (Klemm et al. 2005), derived from waste cellulose become more common. By reporting them here, scientists can now compare these findings to their own, which may aid in quantifying the presence of these synthetic cellulose fibres in animal and ocean samples moving forwards.

4.2 Ingestion pathways.

There are at least two potential ingestion pathways for contaminant particles by demersal shark species. Firstly, via the presence of contaminants directly in their food source. Microplastics and other synthetic materials have been reported in several prey species for these sharks, including crustaceans and molluscs (Watts et al. 2014, Van Cauwenberghe & Janssen 2014, Devriese et al. 2015, Rummel et al. 2016, Murphy et al. 2017). Some of these prey items have also been to shown to take-up and translocate microplastics around their bodies in laboratory conditions (Browne et al. 2008, Gandara e Silva et al. 2016), as well as transfer microplastics up the food-web (Farrell & Nelson 2013). The species in this study show some variation in their published dietary strategies with starry smooth-hounds and spiny dogfish having fairly specialist diets, compared to small-spotted catsharks and bull huss which are more generalist (Saldanha et al. 1995, Domi et al. 2005, Ellis et al. 2009, Martinho et al. 2012). It may have expected for the generalist feeding sharks to have more contaminants due to feeding on a wider range of prey items, however this was not evident.

The second pathway for exposure to these contaminants could be through direct engulfment alongside target prey species. Habitat use has been identified as a potential driver of plastic ingestion for other elasmobranch species, including whale sharks and manta rays (Germanov et al. 2019), as well as bony fish species (Ogonowski et al. 2019).The sharks analysed in this study all display similar strategies while feeding in their demersal habitat, in that to swallow their prey, they engulf it whole using suction feeding (Wilga & Motta 1998, Huber & Motta 2004). In doing so, many of these species will ingest large quantities of sediment alongside their prey. Although the majority of this is immediately expelled from the mouth, some makes its way to the gut (Kalmijn 1971, Smith & Merriner 1985). Numerous studies have revealed that microplastics eventually sink to the seafloor and rest in the sediment (Woodall et al. 2014, Maes et al. 2017, Ling et al. 2017, Martin et al. 2017). While sediment particles do occur quite regularly in these shark species, it is highly likely that many of the ingested microfibres would be excreted alongside these natural sediment particles. The potential for these particles to cause internal damage before excretion remains to be tested. The two individuals in this study containing comparatively high loads of microfibres had most likely fed on prey species close to anthropogenic materials. It is not uncommon for these shark species to raid inshore lobster/crab pots, and as the microfibres found in these individuals were almost all exclusively olefin polypropylene, this may have been what these pots were made of. Consequently, during feeding, a piece of material from the pots may have been ingested, which then broke down into multiple fibres as a result of natural stomach acid and/or the potassium hydroxide used in this study.

Existing studies have attempted to analyse environmental microplastic contamination in the North-East Atlantic, both on the sea surface and the sediment (Lusher et al. 2014, Maes et al. 2017). Lusher et al (2014) (Lusher et al. 2014) found that 94% of samples from surface waters in the North-East Atlantic contained what they believed to be potential microplastics, although after further analysis 63% of these appeared to be matt black anthropogenic fibres and not true microplastics. These matt black fibres are similar in description to many of the fibres found in our current study. When analysed under Raman FT-IR Lusher et al (2014) found they were matched closely with cellulose and rayon, again similar to the cellulose fibres found in this study. In a separate study, Maes et al (2017) identified microplastic particles in 89% of sediment samples from the North Sea and English channel, with most of the plastics considered spheres (microbeads) and fibres (Maes et al. 2017), however these authors do not allude to regenerated cellulose fibres in their samples, which may have been present, but not recorded. Given these environmental levels, it should, therefore, be no surprise that approximately 70% of the sharks in this study contained at least 1 synthetic particle.

4.3 Polymer Identification

Analysing the polymer make-up of marine plastics can reveal potential sources, fate and causes for ingestion (Duncan et al. 2018, Nelms et al. 2018). The use of FT-IR spectrometry to analyse environmental samples is a reliable method of determining their polymer make-up (Hidalgo-Ruz et al. 2012, Shim et al. 2017, Jung et al. 2018) and should be fundamental to any future study. The polymers identified largely reflect the results of similar studies in the marine environment (Remy et al. 2015, Comnea-Stancu et al. 2017, Cai et al. 2017, Duncan et al. 2018) and are also similar to polymer diversity of microplastics globally (Duncan et al. 2018, Gago et al. 2018, White et al. 2018), with polypropylene being one of the most widely abundant polymers identified worldwide (Duncan et al. 2018).

Scientists are, however, now seeing synthetic cellulose fibres being recorded in environmental samples across multiple studies (Lusher et al. 2013, Woodall et al. 2014, Duncan et al. 2018, Gago et al. 2018), although currently their diverse origins remains somewhat understudied. Such anthropogenic fibres made up a third of the analysed contaminants and were identified as regenerated cellulose, such as viscose and rayon, as well as lyocell and cotton, with the likely source of such fibres being textiles or personal hygiene items (Hartline et al. 2016, Napper & Thompson 2016, Salvador Cesa et al. 2017). Spectral libraries for FT-IR set-ups must continue to expand moving forwards, in order to develop reliable databases that are capable of accurately identifying regenerated cellulose fibres within environmental samples.

Estimates show that an average clothes wash of 6kg can release more than 700,000 fibres into waste water facilities and from there into the marine environment (Napper & Thompson 2016) and fibres such as polyester and cotton are globally in-demand between 24-46 million tonnes per year (Ladewig et al. 2015). Interestingly, the fibre lengths identified in the digestive tracts of sharks were relatively similar to that of fibres released upon washing of various textiles under laboratory conditions (see Figure 1). It is likely that the washing of clothes is a major fibre entrance route.

4.5 Potential implications

As only a sub-sample of gut content for each animal (20ml)) was tested, the proportional incidence of anthropogenic contaminants reported is a conservative estimate. Due to the microscopic size of these synthetic fibres, direct internal organ damage is unlikely, when compared to ingestion of larger macro-plastics, although the ability of small fibres to cause inflammatory damage is acknowledged in other contexts (Ivar do Sul & Costa 2014, Ryan et al. 2016, Pham et al. 2017). Translocation of relatively large (150 μm) particles can occur across the vertebrate gut via persorption (the passage of particles through the epithelial layers of the gastro-intestinal tract), whilst smaller particles are taken up through normal digestive processes such as pinocytosis and phagocytosis, circulating through the blood and lymph vessels. Thus, there is the opportunity for such circulating particles to enter cells and induce inflammatory damage before being excreted (Galloway 2015). Fibres of 100-1000 μm will most likely pass straight through the digestive tract and be excreted with other waste products (Duncan et al. 2018).

Future research could aim to assess whether certain fibres present exposure risks of associated contaminants and/or persistent organic pollutants (Duncan et al. 2018, Nelms et al. 2018). There is suggestion that certain textiles and clothing may contain toxic chemicals such as BPA (bisphenol A) and BPS (bisphenol S) (Xue et al. 2017), with both chemicals capable of causing disruption to reproductive and endocrine systems as well as growth suppression in marine taxa, at relatively low doses (Aluru et al. 2010, Huang et al. 2012, 2018, Park et al. 2019). Other studies have shown different associated contaminants can present inherent biological risks to various species, including elasmobranchs (Fossi et al. 2014, 2017, Rochman et al. 2014, Germanov et al. 2018).

Research has revealed that spiny dogfish and small-spotted catshark are regularly sold in fish and chip shops under pseudonyms such as “Rock”, “Rock salmon” and “Murgey” (Hobbs et al. 2019). If contaminants are able to pass from the digestive tract to the muscle tissue of these shark species, then humans may inadvertently be consuming these pollutants. Although, as of currently, there is no conclusive evidence to suggest these pollutants present inherent health risks to humans, further research is recommended to investigate the presence or absence of these particles in the muscle tissues of these shark species and other fish consumed across the world.

5. Conclusions

This study presents the first evidence of microplastics and anthropogenic fibre contaminants in a range of native UK demersal shark species. Although not occurring in as high levels as in other marine megafauna, the presence of anthropogenic particles in yet more marine species highlights the ubiquitous nature of these contaminants. Contamination at these levels is highly unlikely to cause any detrimental population effects. Due to these low levels of ingestion, these species are perhaps not ideal candidates to be used as bio-indicators for marine pollution in demersal habitats when compared to other bony fish species. Nonetheless, if inorganic pollutants can attach to these microfibres, alongside a future increase in their prevalence throughout the marine environment, biological side-effects may occur. Further research on the sources and pathways of these fibres may inform policy to reduce their overall prevalence in the environment. By limiting their production in everyday products (through supporting reduction, reuse and replacement of fibre-generating materials from the resource flow) and implementing strategies to prevent their initial entry into the oceans there lies the potential to dramatically reduce the occurrence of microfibres in the marine environment and across food webs.

Table 1: Breakdown of publications on elasmobranchs and microplastics, featuring species examined, location of samples, methodology for extraction and identification of microplastics/fibres and percentage of ingestion for species studied. Some figures presented here as reported in their respective study.

Ocean Basin	Species Examined	N	Methodology	% Ingestion	Reference
Atlantic					
North-East	*Scyliorhinus canicula* *Scyliorhinus stellaris* *Mustelus asterias* *Squalus acanthias*	46	Dissection, 20% KOH digestion, FT-IR	67%	Parton et al In Press
	Scyliorhinus canicula *Raja asterias*	20 7	Dissection, FT-IR	20% 40%	Neves et al 2015
North sea	*Scyliorhinus canicula*	20	Dissection, Visual inspection	15%	Smith 2018
Mediterranean					
Balearic Islands	*Galeus melastomus*	125	Dissection, FT-IR	17%	Alomar & Deudero 2017
Western Ligurian Sea	*Prionace glauca*	95	Dissection, FT-IR	25%	Bernardini et al 2018
Tyrrenhenian Sea	*Galeus melastomus* *Scyliorhinus canicula* *Etmopterus spinax*	96	Dissection, 10% KOH digestion, FT-IR	69%	Valente et al 2019
Ionian Sea	*Pteroplatytrygon violacea* *Galeus melastomus* *Squalus blainville* *Etmopterus spinax*	2 741 75 16	Dissection, Visual inspection	50% 3% 1% 6%	Anastasopoulou et al 2013
Pacific					
Gulf of California	*Rhincodon typus*	12	Skin biopsy (used to infer contaminant levels)	8.42 ng/g w.w. PCBs 1.31 ng/g w.w. DDTs 0.29 ng/g w.w. PBDEs 0.19 ng/g w.w. HCB	Fossi et al 2017
Indian					
KwaZulu-Natal, South Africa	*14 species, see study for details*	15,666	Dissection, Visual inspection	0.38% (macroplastics)	Cliff et al 2002

General Discussion

Plastic pollution is a clear threat to wildlife that can cause pain, suffering and often death for multiple marine species, including elasmobranchs (Gall & Thompson 2015, Duncan et al. 2017, Parton et al. 2019). Although it may not have severe implications for global shark and ray populations, it is unequivocally an animal welfare issue for these species (Parton et al. 2019). Elasmobranchs now join the ever-growing list of marine species that are negatively impacted by plastic pollution, either via entanglement or ingestion (Laist 1997, Cliff et al. 2002, Page et al. 2004, Seitz & Poulakis 2006, Murray & Cowie 2011, Votier et al. 2011, Wegner & Cartamil 2012, Barreiros & Raykov 2014, Vegter et al. 2014, Schuyler et al. 2014, Setälä et al. 2014, Devriese et al. 2015, Lawson et al. 2015, Neves et al. 2015, Stelfox et al. 2016, Duncan et al. 2017, 2018, Nelms et al. 2018). Further data collection could reveal elasmobranchs to be far worse impacted than what has been reported above, particularly if juvenile life stages, or certain species already displaying severe population declines are negatively affected (Duncan et al. 2017, Parton et al. 2019).

In chapter one, the first systematic literature review of global elasmobranch entanglement in anthropogenic debris was conducted. This literature review, paired with novel data collection from social media site Twitter, highlighted that elasmobranch entanglement is severely under-reported in the scientific literature. By directly comparing the number of entanglement incidents found over 80 years of scientific literature, and the number of entanglement incidents over 10 years of data from Twitter, it was shown that entanglement for elasmobranchs is likely happening at far greater levels than scientists currently believe. The use of Twitter to collect this environmental data has revealed that social media websites contain a wealth of information that is worth investigating. Careful use of social media websites in this way, whilst ensuring to take into account various biases, may help provide us with real-time data on a host of environmental issues (Parton et al. 2019).

‘Ghost fishing gear’ was identified to be the most common entangling material for sharks and rays, which is consist with findings in other marine species (Macfadyen et al. 2009, Wilcox et al. 2013, 2015, Nelms et al. 2016, Duncan et al. 2017). When comparing entanglement of elasmobranchs to other marine species such as marine turtles and cetaceans, it may appear the latter two species are more at risk. However, it is felt that this is likely due to more focus being placed on these species and their relationship with marine debris entanglement in recent years. The lack of data for this issue on elasmobranchs prevents us from making accurate comparisons between other marine fauna, however with the current available data it would suggest shark and ray species are less impacted than that of marine turtles and/or cetaceans. The Indian and Pacific Oceans were highlighted as data-deficient areas for entanglement and more research was recommended to investigate the threat of Fish Aggregating Devices to sharks and rays in these ocean basins (Parton et al. 2019). Various features that may pre-dispose certain species to entanglement were identified including habitat use, migratory nature and body shape/form. There is the potential for detrimental population effects to occur as a result of entanglement.

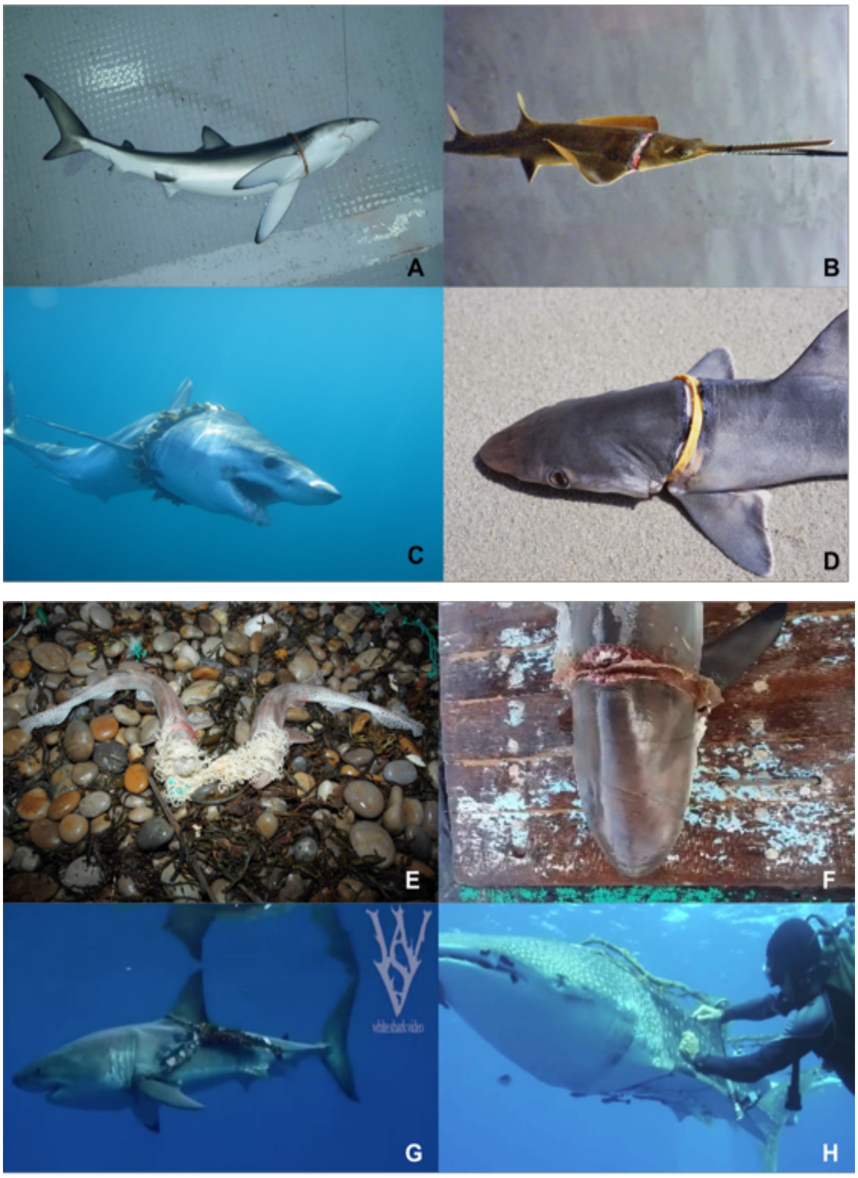
However, this may only be apparent for particular species, such as those who may already be listed as vulnerable or endangered. If juvenile individuals for these threatened species are prone to entanglement (as is the case among other marine species), then the knock-on effect for threatened elasmobranch species could be damaging. With further data collection on this topic, more accurate conclusions could be reached. A standardised method of reporting future shark and ray entanglements was outlined and subsequently an online entanglement report form for citizen scientists was created (in collaboration with the Shark Trust UK) known as ShaREN (The Shark and Ray Entanglement Network). To date, ShaREN has already collected over 100 entanglement reports for sharks and rays across the world. Future data collection via this citizen science project will enable us to highlight specific ocean areas where the risk of entanglement for sharks and rays is higher, whilst also further clarifying the most at-risk species and life stages. With this additional data scientists will be able to implement mitigation strategies for struggling elasmobranch species and reduce the potential for further population declines.

In chapter two, for the first time, empirical evidence of microplastics and anthropogenic fibres in UK demersal shark species was identified. By conducting laboratory dissections, microscope analysis and FT-IR spectrometry, it was revealed that all four species analysed contained either microplastics or anthropogenic microfibres. 67% of all individuals contained these contaminants, which is significantly higher than other reports for similar species around the world (Anastasopoulou et al. 2013, Neves et al. 2015, Alomar & Deudero 2017, Bernardini et al. 2018, Smith 2018). Consistent with other scientific findings, blue and black fibres were the most commonly identified contaminant colours (Lusher et al. 2014, Steer et al. 2017, Duncan et al. 2018, Gago et al. 2018). There appeared to be no difference in contamination levels between males and females and although diet and feeding strategies may play a role in the quantity of fibres ingested, further investigation is needed to ascertain this. The study highlights the ubiquity of anthropogenic fibres, particularly that of which from human clothing and/or textiles, in the marine environment (Lusher et al. 2013, Woodall et al. 2014, Gago et al. 2018). At the presented current levels of contamination, ingestion of these fibres is unlikely to have severe detrimental population effects on these species. However, if further research reveals these fibres to contain persistent organic pollutants, ingestion of these fibres may lead to detrimental physiological effects on North-east Atlantic demersal sharks.

This thesis has furthered our knowledge on the extent of the impact of plastic pollution on elasmobranch species. It is concluded that both entanglement within and ingestion of plastic or anthropogenic debris is unequivocally an animal welfare issue for sharks and rays. With increasing levels of debris in our oceans and with accelerated decline in elasmobranch populations, further research could reveal this threat to be of significant conservation concern for shark and ray populations around the world. Future research on elasmobranch entanglement could involve mapping known areas of ocean debris alongside shark or ray migration routes, allowing scientists to decipher areas and species that are more prone to entanglement incidents. Future research on microfibre ingestion in sharks should aim to decipher if anthropogenic synthetic fibres contain associated toxins capable of causing physiological changes, as well as investigating whether these microscopic fibres are able to pass from the digestive tract to other parts of the body. Mitigation strategies to reduce production of plastic, remove debris from oceans and prevention measures to stop existing plastic from initially entering our oceans should be implemented immediately to reduce negative impacts on struggling marine species, including elasmobranchs.

Supplementary Material

Chapter One:



Supp Figure 1 A-H: Images of shark species entangled in marine anthropogenic debris from the scientific literature. A) Juvenile blue shark (*Prionace glauca*) entangled in the gill region with strapping bands (Miguel Cayuela Padilla from Colmenero et al 2017). B) Smalltooth sawfish (*Pristis pectinate*) entangled around the gill region with an elastic band (Seitz & Poulakis 2006). C) Shortfin Mako (*Isurus oxyrinchus*) entangled with fishing rope around the gill region (Wegner & Cartamil 2012). D) Brazilian sharpnose shark (*Rhizoprionodon lalandii*) entangled in the gill region with plastic debris rings (Sazima et al 2002). Alongside images of shark species entangled in marine anthropogenic debris found on social media site "Twitter". E) Two lesser spotted dogfish (*Scyliorhinus canicula*) entangled in netting, Chesil beach, Dorset (Steve Trewhella). F) Spot-tail shark (*Carcharhinus sorrah*) entangled in the gill region with a polythene bag, south of Astola Island (WWF-Pakistan). G) Great white shark (*Carcharodon carcharias*) entangled in fishing rope, Guadeloupe, Mexico (Mike Bolton & Skyler Thomas). H) Whale shark (*Rhincodon typus*) entangled in fishing rope (Caters News Agency). All images used with permission.

Chapter Two:

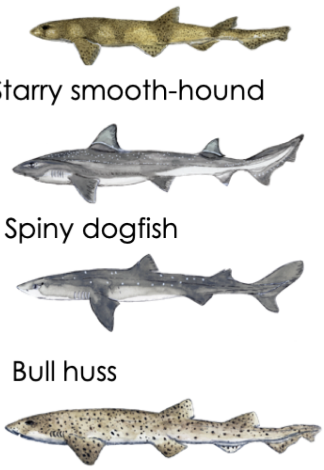
Supp Table 1: Results from the subsample of isolated particles (N = 62) analysed using Fourier transform infrared spectroscopy (FT-IR) to determine their polymer make up from gut content residue samples of UK demersal sharks. SSC: small spotted catshark, SS: starry smooth-hound, SD: spiny dogfish, BH: bull huss. Percentage of synthetic contaminants annotated in table.

Origin	**Group**	**FT-IR Identification**	**SSC**	**SS**	**SD**	**BH**
Synthetics	Plastics	Olefin Polypropylene fibres	-	5	-	10
		Polypropylene fragment	-	-	-	1
		Polyacrylamide	5	-	1	-
		Polyester fibres	-	3	1	1
		Polyethylene fragment	-	-	1	-
	Regenerated Cellulose	Rayon or Viscose	8	7	1	4
		Cellophane	-	1	-	-
			86.6%	**84.2%**	**57.1%**	**76.2%**
Non-synthetics / Low spectral match scores	Other	Hexocyclium	1	-	-	-
		Thiobis	1	-	-	-
		Acetyl triethyl citrate	-	1	-	-
		Ethylene (Low match score)	-	1	-	-
		Poly(film) (Low match score)	-	1	-	-
		Ethyl cellulose (Low match score)	-	-	1	-
		D-biotin	-	-	2	-
		Polyacrylonitrile (Low match score)	-	-	-	1
		Erthryose	-	-	-	1
		Cyanide (Low match score)	-	-	-	1
		Mercuric (Low match score)	-	-	-	1
		Human umbilical cords (Low match score)	-	-	-	1
Total:			**15**	**19**	**7**	**21**

Supp Table S2: Summary results of negative binomial generalised linear model. Top ranked model and adjusted weight after selection for ΔAIC ≤ 2 and applying the nesting rule. Top set model highlighted in bold.

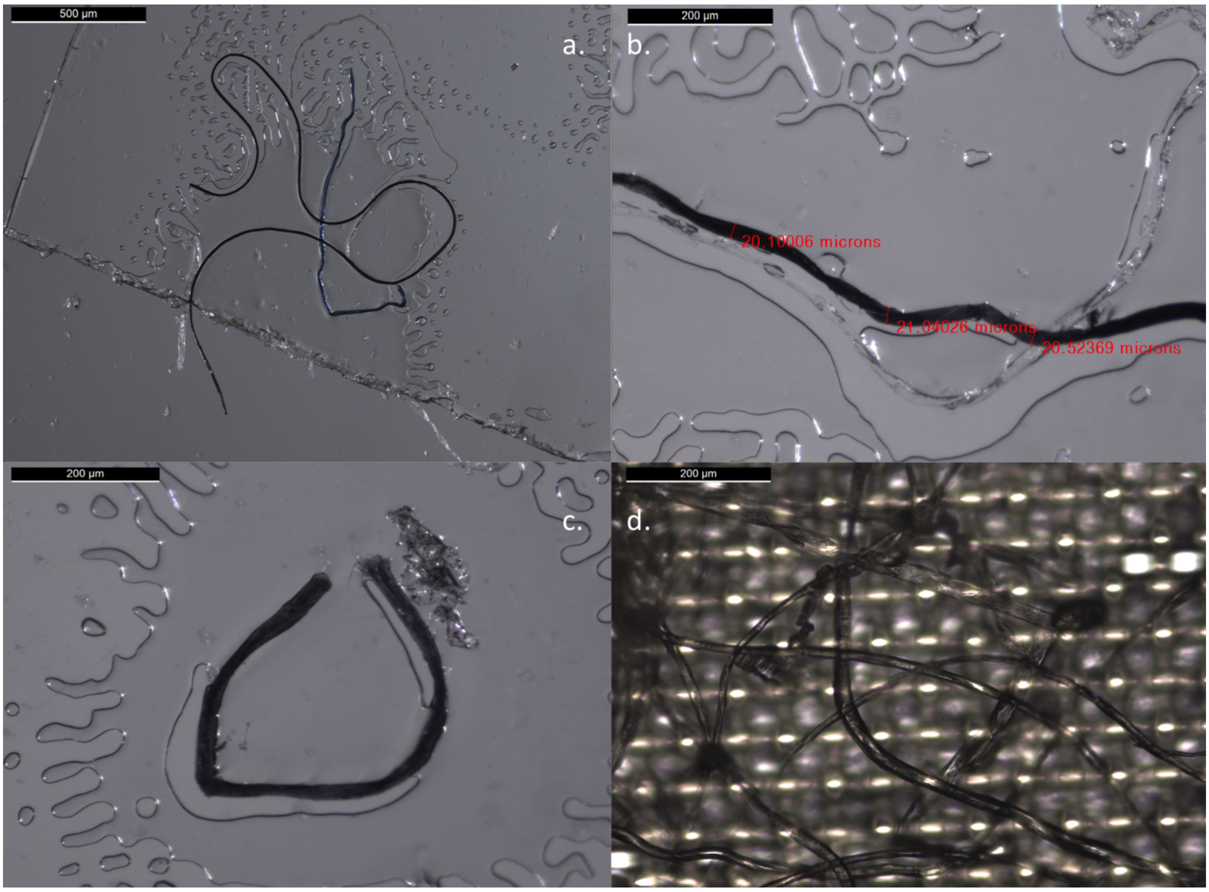
Response variable	Fixed effects	Intercept	d.f.	logLik	AIC	ΔAIC	Weight	Adj. weight
Expected fibres	**~ Length**	**-11.57**	**3**	**-149.26**	**304.5**	**0.00**	**0.37**	**1.00**
	~ 1	3.685	2	-160.62	325.2	20.72	0.00	

d.f.: degrees of freedom. logLik: log likelihood. AIC: Akaike's Information Criterion. Adj. weight: adjusted weight.

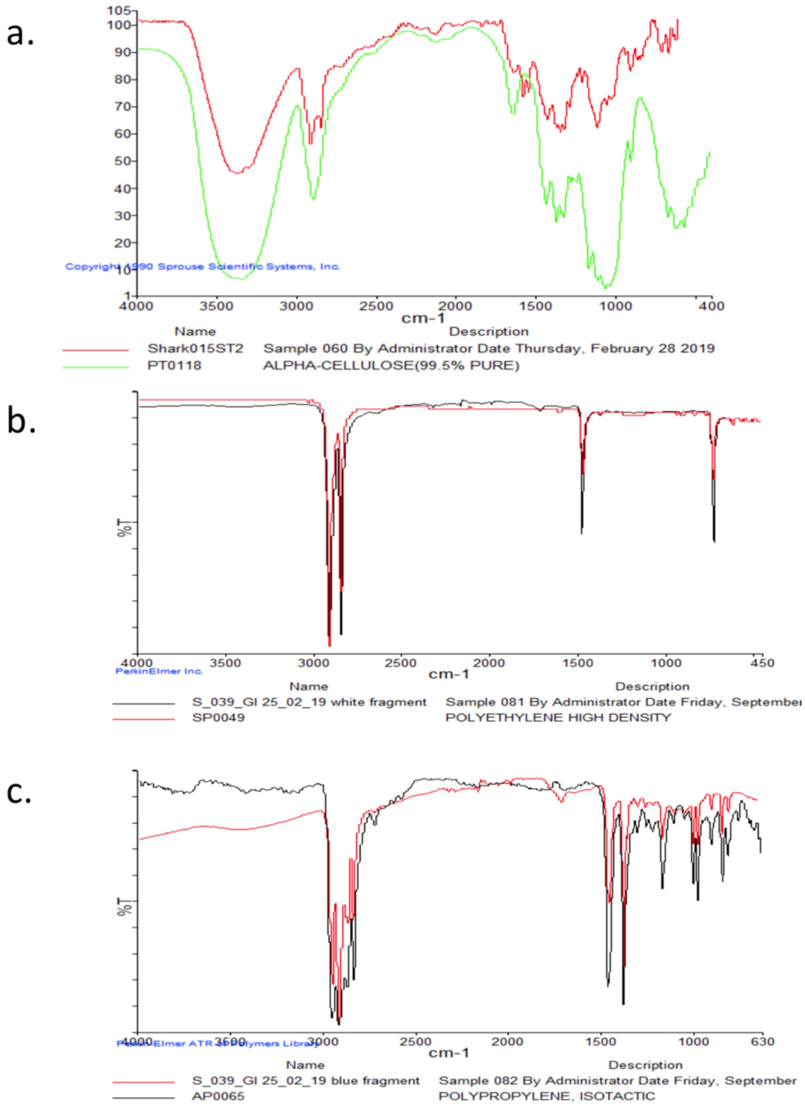


	Crustacean	Fish	Cephalopod
Small-spotted catshark	✓ 5/12	✗ -	✓ 1/12
Starry smooth-hound	✓ 10/12	✗ -	✗ -
Spiny dogfish	✗ -	✓ 4/12	✗ -
Bull huss	✓ 1/10	✓ 3/10	✗ -

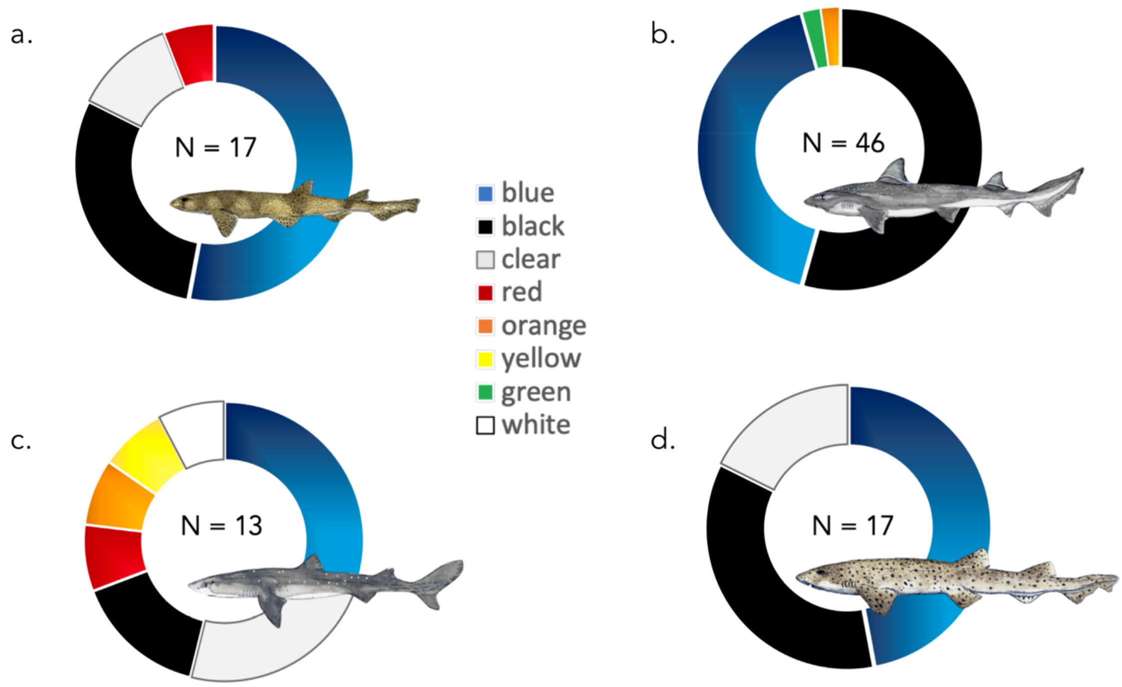
Supp Figure S1: Dietary tick chart. Different dietary items found/not found in each species during visual inspection of stomach contents. Frequency occurrence annotated on figure. “-“ = Not found. Some contents were too digested to visually determine their origins and therefore are not included in the counts here. Elasmobranch drawings by Lucie Jones.



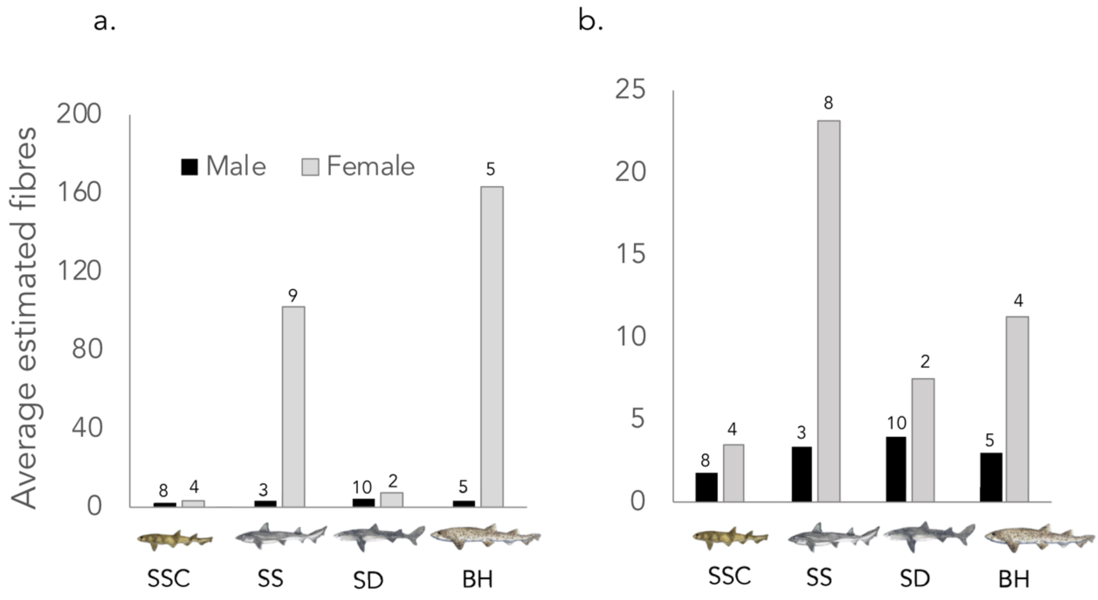
Supp Figure S2: Microscope imagery of fibres found in shark samples, as well as laboratory treated known fibre types. a.) Cellulosic fibre - 500um scale bar. b.) Cellulosic fibre - 200um scale bar, with added measurements displaying uniform diameter indicative of anthropogenic fibres. c.) Cellulosic fibre, 200um scale bar, displaying damaged fibre end. d.) Laboratory treated cotton fibres, 200um scale bar, showing dimensional and morphological similarities to fibres found within shark samples.



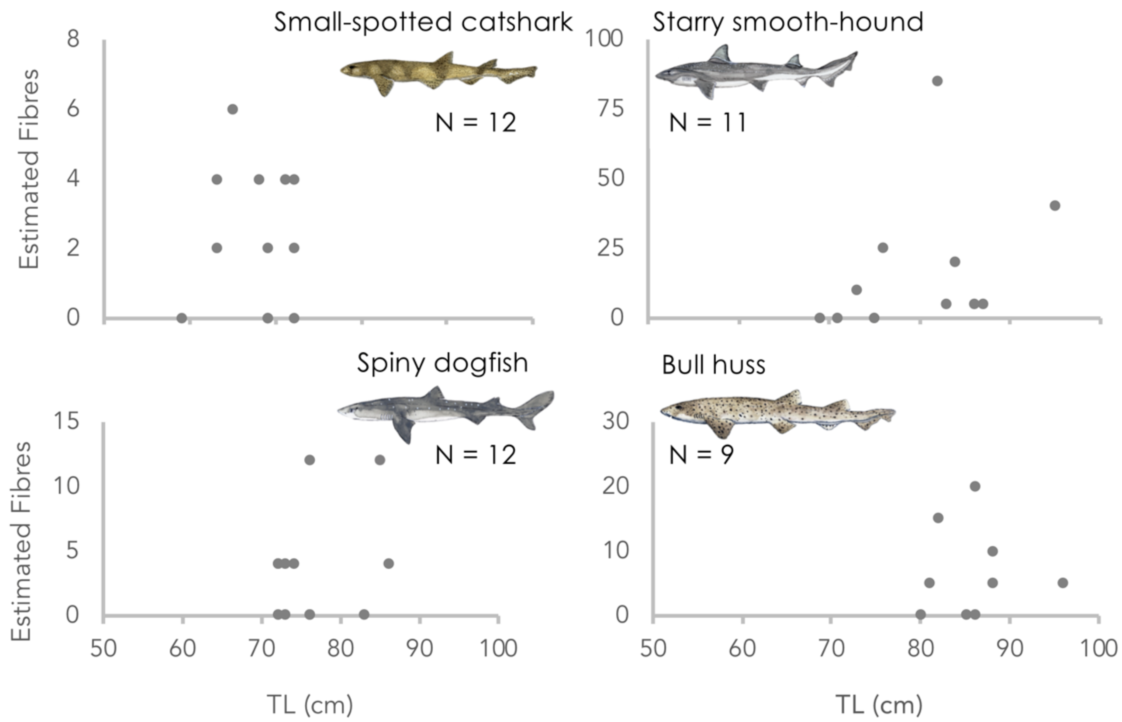
Supp Figure S3: FT-IR spectra. a.) Spectra for cellulosic fibres presumed to be cotton/regenerated cellulose. b. Spectra for polyethylene fragment found in shark sample. c.) Spectra for polypropylene fragment found in shark sample.



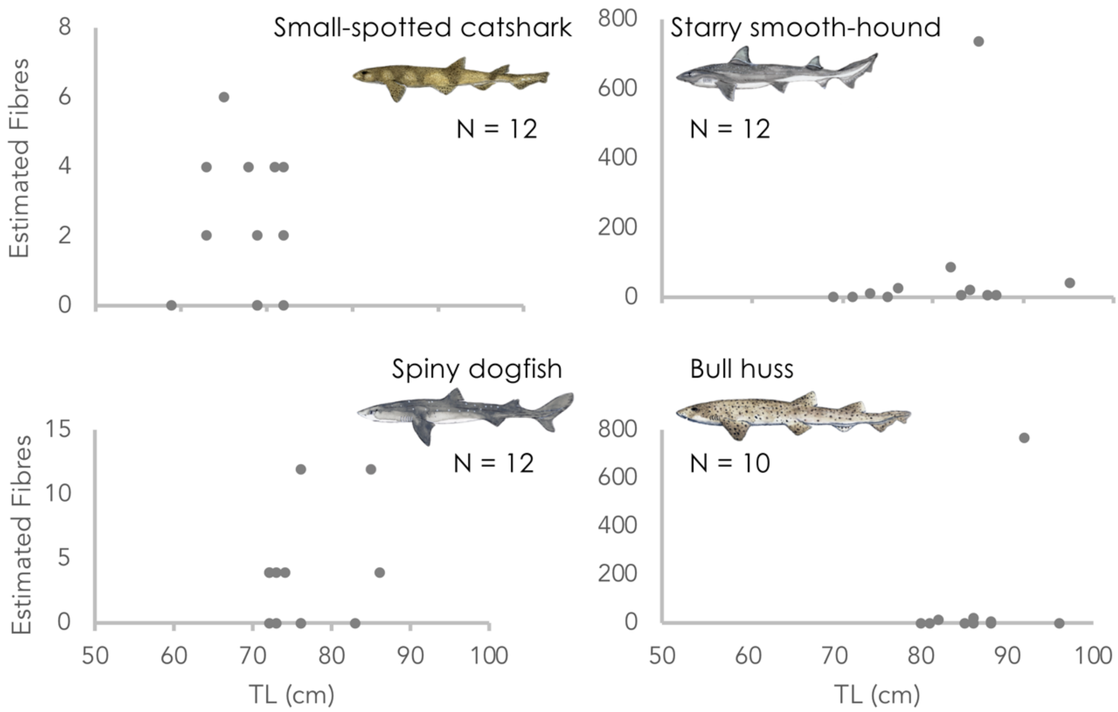
Supp Figure S4: Fibre colour composition with extreme values removed. Pie charts representing colours of ingested fibres, found across both the stomachs and intestines of four species of north-east atlantic demersal sharks: a. small-spotted catshark (*Scyliorhinus canicula*), b. starry smooth-hound (*Mustelus asterias*), c. spiny dogfish (*Squalus acanthias*) and d. bull huss (*Scyliorhinus stellaris*). Total N of coloured fibres identified annotated within figure. Elasmobranch drawings by Lucie Jones.



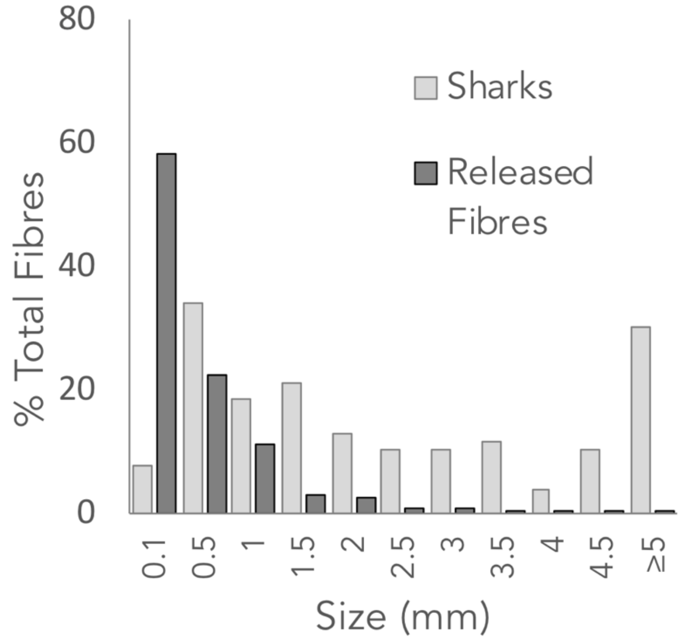
Supp Figure S5: Average estimated fibres breakdown between males and females. a. Two extreme values included (one female starry smooth-hound & one female bull huss). b. Two extreme values removed. SSC: small-spotted catshark, SS: starry smooth-hound, SD: spiny dogfish, BH: bull huss. N of Males/Females sampled annotated above bar. Elasmobranch drawings by Lucie Jones.



Supp Figure S6: Estimated fibres as a function of total length (TL cm) for four shark species. N = annotated. Two extreme values removed (one starry smooth-hound, TL: 85cm, estimated fibres: 735, one bull huss, TL: 92cm, estimated fibres: 770). Elasmobranch drawings by Lucie Jones.



Supp Figure S7: Estimated fibres as a function of total length (TL cm) for four shark species. N = annotated. Extreme values included. Elasmobranch drawings by Lucie Jones.



Supp Figure S8: Fibre length distribution with extreme values removed from shark data. Fibre lengths as a proportion of total fibres for fibres found in shark species (light grey) and fibres released in laboratory conditions after washing of various cotton and polyethylene terephthalate textiles. Palacios Marin AV, (2019) Release of microfibres from comparative common textile structures during laundering (Unpublished Masters dissertation). University of Leeds, UK.

Supplementary text – Chapter Two.

Collection, necropsy and gut content analysis of shark samples

Sharks (n = 46) were obtained from fishermen based down in Cornwall, U.K. All samples were collected and dissected under permission by the University of Exeter ethics committee. Samples of the netting used by the fishermen were also collected and stored for analysis. Four species of NE Atlantic demersal sharks were obtained: small-spotted catshark (Scyliorhinus canicula), spiny dogfish (Squalus acanthias), starry smooth-hound (Mustelus asterias) and bull huss (Scyliorhinus stellaris). Sharks were transported to the University of Exeter, Penryn campus and stored in -80'c freezers until dissection.

Necropsy took place in the post mortem room under sterile conditions. Morphometric shark measurements were taken including: Total length (TL), Precaudal length (PCL), Fork length (FL), First Dorsal height (FDH), Mass (g), Stomach mass (g) and the presence or absence of claspers (M/F). Each species was separated into juvenile or adult individuals based on their size (TL cm) and their genital development.

Upon dissection, the stomach and intestinal tract were removed from the shark species and 10ml of content residue from each was stored in 50ml falcon tubes. This approximated between 20-50% of the total contents of stomach and intestines. Additional notes were taken on the contents of the stomachs to assess what the individuals had been feeding on.

Digestion of samples

Creation of KOH-

20% Potassium hydroxide (KOH-) solution was created using KOH- clusters at a ratio of 200g/1L of filtered water. Filtered water was created using a Nalgene rapid flow filter from filtered water taps in the laboratory. 20% KOH- was added to samples of stomach and intestinal tract at 1:4 ratio using a 40ml glass pipette, washed with Milli-Q water between uses. Treated samples were later oven heated for 48 hours at 60’C to aid in the digestion process.

Filtering of Samples

Filtered water was initially run through a Millipore filtration kit (MFK) to remove any contaminants present on the equipment, this was repeated between each sample. Treated samples were shaken and subsequently run through the MFK onto 30um filter paper cut into 6cm diameter circles. Biological material retained on the inside of the filtration kit was flushed through the filtration kit with Milli-Q water. Upon filtration, the 30um filters were quickly removed using stainless steel tweezers and placed into petri dishes, which were subsequently sealed with masking tape and stored for later analysis.

Microscopy analysis

Filtered shark samples were examined under a digital stereo light microscope (Leica M165C) at 8x magnification and scanned for contaminants. Samples were scanned across horizontally until all of the sample had been viewed. Microplastic contaminants were recorded and categorised as either: fibres, beads or fragments and further subcategorised into 5 colour categories: red, blue, black, yellow or other. Length of contaminants were measured, alongside the smallest diameter of any suspected fragments and beads and photographed by a digital camera (Leica DFC295; Leica Suite Application Version 3.6.X).

Contamination prevention

Personal protective equipment was used at all times. As some microplastics/fibres may be on clothing, attached to laboratory equipment or airborne, we undertook several steps to control for and prevent contamination of shark samples. All equipment and apparatus were rinsed thoroughly throughout with Milli-Q water as well as between uses. Surfaces were wiped down with 70% ethanol prior to work commencing. Airborne contamination blanks (N = 25, one per bout of laboratory work) consisting of filter paper dampened with filtered water placed in a petri dish) were run throughout all stages of the process and were sealed with masking tape and stored for microscopic analysis upon completion of dissections, oven-heating, filtrations and microscopic analyses. Analysis of these filters showed minimal evidence of contamination with the presence of some fibres (n = 6 cases of single fibres), that visually appeared different to those found in the shark samples. As an extra precaution, for any samples processed during the same bout, if they contained any fibres of the same colour these were discounted.

Procedural blanks (N = 24) were treated in the same way as the shark gut content samples and were run parallel to the digestion, oven-heating and filtration processes. These were poured through the 30um mesh filters (as per the methods) and were stored for microscopic analysis to check for contamination. No evidence of any microplastic contamination was found.

Polymer Identification

A subsample of contaminants (n = 57) were investigated using Fourier Transform Infrared spectroscopy (FT-IR) to determine their polymer make-up.

Individual candidate materials (fibres and fragments) were positioned on the surface of a silver filter (47 mm diameter silver-coated membrane filter, pore size 5 μm, Sterlitech) held in a glass petri dish and their positions marked by scratching the filter surface both to facilitate orientation under the microscope and to ensure that only those fibres and fragments originating from the samples were subsequently analysed (i.e. to avoid any possible interference from airborne microplastics). Both the silver filters and petri dishes had been inspected before use using a dissecting stereomicroscope under both low and high magnification in order to verify that they were completely free from fibres and fragments. Candidate materials were examined using a PerkinElmer Spotlight 400 FT-IR Imaging System (MCT detector, KBr window) operating in reflectance mode across a wavenumber range from 4000 to 750 cm-1 and with a resolution of 4 cm-1.

The infrared spectra were acquired, processed and analysed using PerkinElmer Spectrum software (version 10.5.4.738), with polymers being identified by automated matching combined with expert judgment against commercially available spectral libraries (including polymers, additives, solvents, etc.) and an additional custom spectral library prepared in our laboratory using a range of polymer standards and potential contaminating materials (e.g. tissues, gloves, laboratory coats). Any fibres or fragments appearing on the filters other than those previously marked were excluded.The comparisons were made using PerkinElmer Spectrum software (version 10.5.4.738), incorporating a total of 8 different commercially available spectral libraries relating to polymers, polymer additives and adhesives as provided by PerkinElmer (adhes.dlb, Atrpolym.dlb, ATRSPE~1.DLB, fibres.dlb, IntPoly.spl, poly1.dlb, polyadd1.dlb & POLYMER.DLB) as well as an additional library compiled at the Greenpeace Research Laboratories in order to exclude common laboratory contaminants (fibres from tissues, blue roll, laboratory coats, glove fragments, etc.). The Spectrum software allows for the simultaneous comparison of spectra obtained for a sample against all nine libraries, and reports the 10 most likely matches across all of those libraries, in each case, matches which were then subsequently checked by the analyst in order to verify the quality of the match and the reliability of the identification.

On samples where there were multiple contaminants, a minimum of 5 contaminants were selected for analysis with FT-IR. Scores greater than 65% were considered reliable spectral matches. Some spectral matches of cellulose fibres between 65-70% were sent for visual analysis at Leeds university to confirm their identity by light microscopy / image analysis and were eventually accepted.

Statistical Analysis

A negative binomial generalised linear model (GLM) was used to investigate the influence of species, sex, and individual length on the expected number of ingested fibres, using the MASS package (Venables and Ripley 2002) in R v3.5.1.(R Core Team 2018). All combinations of terms were examined and ranked by Akaike's Information Criteria (AIC) using subset selection of the maximal model using the MuMIn package v1.42.1. (Barton 2015). Top ranked models were defined as models ΔAIC ≤ 2 units of the best supported model, after excluding further models where a simpler model attained stronger weighting (Richards et al 2011).

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