The plastic we don't see

The impact of plastic on benthic marine organisms

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16-04-19

Abstract

Plastic marine litter is now found in every sea and ocean around the world. Research on the impact of plastics on wildlife has mainly focused on floating macroplastics and (pelagic) macrofauna, such as turtles, marine mammals and seabirds. The majority of produced plastics have however a higher density than seawater and would therefore sink to the seafloor when they reach the marine environment. Even very buoyant plastics can reach the seafloor through processes such as biofouling. Sediments have been suggested as a major sink for plastic marine litter which highlights the importance of studying the impact this will have on the benthic organisms that inhabit these areas. The goal of this essay is to assess the impact macro- and microplastic have on benthic marine organisms. By reviewing the available literature I try to answer this question by: 1) giving an overview of the plastics most commonly encountered in and on the sediments, 2) making a comprehensive evaluation of the known direct and indirect effects of plastic on benthic marine organisms and 3) critically evaluating current results by analyzing the commonly used methods for assessing the abundance and effects of plastics on the benthos. In general, plastics seem to occur more in coastal seas and areas of low circulation. Organisms inhabiting these areas therefore seem to be most at risk. However, studies on the effects of plastic uptake by benthic organisms display mixed results. Some organisms are seemingly not affected by the uptake of plastic, whereas others show heavily impaired growth, feeding activity or fecundity. Plastic exposure studies have often used unrealistic experimental conditions thereby not mimicking the natural environment but rather showing proof of principle. In conclusion, both macro and microplastics have the potential to greatly impact benthic organisms, but a standardization in methods and more realistic effect assessments are urgently needed for a better understanding of the magnitude of this issue in its current state.

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1. Introduction

Plastics comprise a large group of synthetic polymers (Ryan, 2015). Plastics are extremely durable, bio-inert, have low thermal conductivity and a high strength-to-weight ratio, but most of all they are cheap to manufacture (Andrady & Neal, 2009). They consequently form the ideal material for a large set of products used in everyday life. Their popularity has been reflected by the annual increase in plastic production over the last decades since the start of their mass production in the 1950s (Figure 1). Even though plastic offers many benefits to mankind, the material poses a great risk for the (marine) environment (Van Cauwenberghe et al., 2015). To date, it is estimated that more than 8300 million metric tons of plastic has been produced of which the largest part is accumulated in landfills or the natural environment (Geyer et al., 2017). Due to improper waste disposal a fraction of the produced plastic eventually ends up in the marine environment, where it now forms the biggest portion of marine litter (Derraik, 2002). It is estimated that 4.8 to 12.7 million metric tons of plastic enters the ocean each year (Jambeck et al., 2015). Plastic marine litter is globally omnipresent and contaminates a wide range of habitats, such as beaches (e.g. Merrel, 1980; Jayasiri et al., 2013), coral reefs (Lamb et al., 2018), the open ocean (Cózar et al., 2014) and even the deep sea (Van Cauwenberghe et al., 2013) ranging from the tropics (Costa & Barletta, 2015) to the poles (Barnes et al., 2010). Organisms are therefore exposed to marine debris worldwide and this has raised a lot of concern. Entanglement by and ingestion of plastic debris has been reported for tens of thousands individual organisms (Gall & Thompson, 2015). More than 557 species of animals, including all turtle species, half of all marine bird species and two-thirds of marine mammal species have been shown to suffer from plastic entanglement or ingestion (Kühn et al., 2015). Plastics can furthermore function as a raft and facilitate the spread of invasive species (Goldstein et al., 2014).

While the problems of big pieces of floating plastic have been apparent and widely researched for a long time, research on the effects of microplastics is relatively young but has gotten a lot more abundant in the last decade. Microplastics are usually defined as plastic particles with a size of <5 mm (e.g. Moore, 2008; Watts et al., 2014; Van Cauwenberghe et al., 2015) and are manufactured in this size range (primary microplastics) or are formed through the fragmentation of larger pieces of plastic (secondary microplastics) through physical erosion or UV radiation (Van Cauwenberghe et al., 2015). Primary plastics are for example present in human cosmetic products (Fendall & Sewell, 2009) which find their way to the marine environment through sewage effluent. A major source of secondary microplastics comes from washing synthetic clothing, a single piece of clothing can release almost 2000 microfibers per wash (Browne et al., 2011). As microplastics can have the same size and color as potential prey items for different types of organisms, they could be easily mistaken for food. Indeed many animals have been reported to ingest microplastic in both natural and laboratory settings (Gall & Thompson, 2015).

Another concern on plastics is their ecotoxicological properties (Anbumani & Kakkar 2018). Plastics can pose a chemical hazard as they have been shown to absorb organic contaminants from aquatic environments (Rochman et al., 2013). This in turn increases the exposure of wildlife to these harmful toxins (Rochman et al., 2013). Plastics can also contain a lot of chemical additives, such as thermal stabilizers, biocides, flame retardants etc. (Hahladakis et al., 2018), which can have biological consequences when they come in contact with animals (Teuten et al., 2009). An increasing amount of studies now focus on the effects of ingestion of contaminated microplastic by organisms (e.g. Besseling et al., 2012; Koelmans et al., 2013; Batel et al., 2018).

Studies on plastics affecting wildlife have stereotypically focused on (pelagic) macrofauna, such as turtles, marine mammals, fish and seabirds (Kühn et al., 2015). However, many types of plastics will sink and end up in the marine sediments. In fact, circa 54% of manufactured plastics have a higher density than seawater (Watts et al., 2015). Not only heavy plastics will reach the sediments, even very buoyant plastics can eventually sink to the seafloor. Biofouling causes an accumulation of biomass on plastics which can affect their buoyancy (Andrady, 2011; Zettler et al., 2013; Van Cauwenberghe et al., 2015). In an experiment performed by Lobelle and Cunliffe (2011), the development of microbial biofilms on submerged polyethylene bags was monitored. It was found that biofilms formed quickly on the plastic (within three weeks) changing physiochemical properties including a decrease in buoyancy. The global amount of plastic on the surface of the open-ocean is much lower than expected from estimates of waste input which could be partly explained by biomass accumulation of plastic fragments (Cózar et al., 2014). Furthermore, it has been opted that zooplankton can package ingested microplastics into their fecal pellets, thereby contributing to the vertical flux of microplastics to the seafloor in the form of marine snow (Van Cauwenberghe et al., 2013; Katija et al., 2017).

As shown above, many plastics could end up on the bottom of our oceans. This highlights the importance of studying the effects of plastics on benthic marine organisms that inhabit these areas. This essay aims to examine the impact of plastics on benthic organisms by: 1) providing an overview of the different types of plastic most commonly found in and on the marine sediments, (2) making a comprehensive evaluation of the known direct and indirect effects of plastic on benthic marine organisms and (3) discussing the commonly used methods for assessing the abundance and effects of plastics on the benthos.

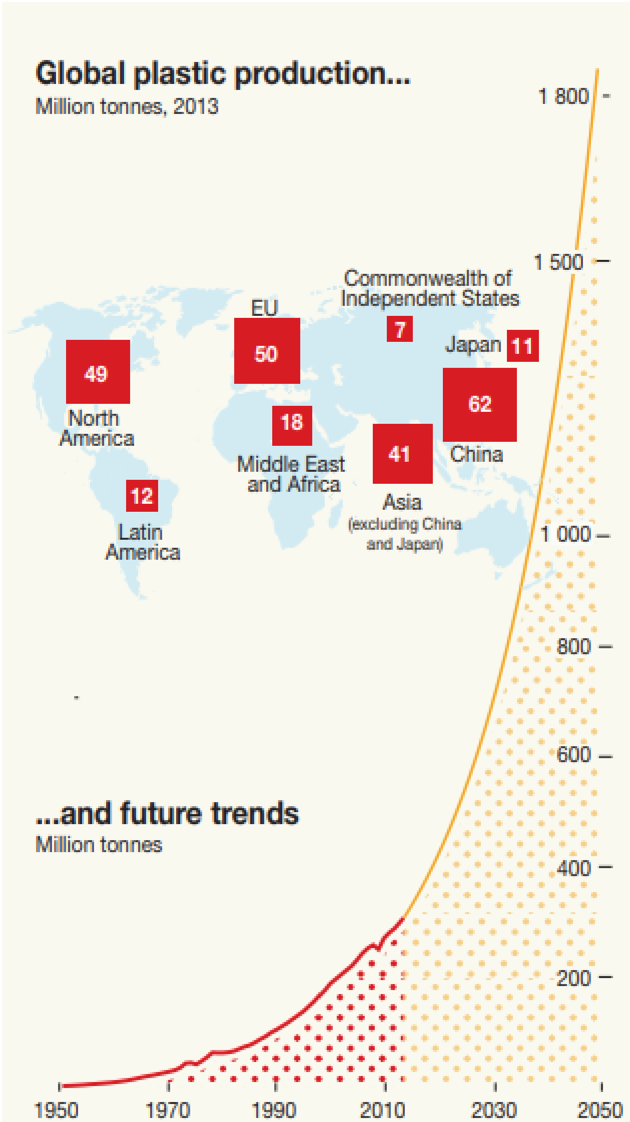

Figure 1: Global plastic production (Mt) and future trends. Taken from GRID-Arendal (2016).

2. Plastics in marine sediments

2.1. Types of plastic in the marine environment

At present, there are many classes of plastic in production (Geyer et al., 2017). Some plastics soften when heated and can therefore be easily remolded (thermoplastic polymers), whereas other plastics consist of permanent cross-linked polymers which cannot be remolded (thermosetting polymers) (Galloway, 2015). Based on these properties, plastics are grouped in seven categories concerning their recyclability: 1) Polyethylene terephthalate (PET), 2) High-density polyethylene (HDPE), 3) Polyvinylchloride (PVC), 4) Low-density polyethylene (LDPE), 5) Polypropylene (PP), 6) Polystyrene (PS) and 7) Remaining plastics. However, only a small percentage of all plastics gets recycled (table 1). Most plastics are not recycled and could therefore end up in nature. The chance that a specific type of plastic will enter the ocean is dependent on its abundance as well as on the type of product the plastic is used in. Geyer et al. (2017) performed a global analysis of all plastic ever to be mass-produced. They calculated that 7300 Mt of non-fiber plastics were produced between 1950 and 2015 which consisted mainly of PE (36%), PP (21%) and PVC (12%). The remaining portion consisted primarily of PET, Polyurethane (PUR) and PS (<10% each). Fibrous plastic mainly consisted of polyester (mostly PET) which accounted for 70% of the total production of 1000 Mt. More than half of all plastics ever produced are no longer in use and are accumulating in landfills or nature (Geyer et al., 2017). Around 80% of marine debris comes from land-based sources (Andrady, 2011). Based on just their shear abundance, the formerly mentioned types of plastic are more likely to find their way to the marine environment. The fate of plastic types are further influenced by their respective usage. PE (high and low-density), PET, PP, PS and PVC are often used in packaging materials and therefore have a high chance of ending up as litter (Andrady, 2011). This is because packaging includes disposable single-use items which are often found on beaches. 75-80 Mt of packaging is annually produced, but what fraction of this eventually reaches the marine environment has not been accurately estimated (Andrady, 2011). Non-packaging plastics, such as Cellulose Acetate (CA), are found in cigarette filters, which are often the most common litter item found on beaches (e.g. Santos et al., 2005; Martinez-Ribes et al., 2007; Munari et al., 2016). Nylons are mainly found in the oceans in the form of lost or discarded fishing gear, such as nets and traps (Andrady, 2011). Fishing gear actually accounts for almost one fifth of marine debris (Andrady, 2011). Table 1 presents an overview of the different types of plastic mostly found in the marine environment together with examples of typical products the plastic is used for.

2.2. Plastic properties

Not all plastic that enters the marine environment is a definite threat to benthic organisms, pieces of plastic must first make their way to the lower regions of the water column. Plastics that are less dense than seawater are buoyant and therefore float on the surface. Examples of these types of plastic are PP and PE which are used in bottle caps and shopping bags, respectively (Andrady, 2011). Most types of manufactured plastic have however a higher density than seawater and therefore float in the water column or sink to the bottom, such as CA, PVC and PET found in cigarettes, plastic films and soft drink bottles (table 1). However, as previously mentioned, through multiple types of modification by organisms (e.g. biofouling or fecal packaging) even floating plastics can reach the seabed (Zettler et al., 2013; Van Cauwenberghe et al., 2015). Plastic items that would otherwise float are furthermore observed on the seafloor through the embedment of sand (Latin et al., 2004). As a result of the sinking plastics, it has been found that more plastic is present in the zone just above the benthos than in the rest of the water column (Latin et al., 2004).

The importance of knowing what types of plastic come in contact with benthic organism lies in the differences in toxicity. Different plastics contain different (amounts of) additives (Lithner et al., 2011). These additives, such as flame retardants and pigments, can leach from the plastic into the environment (Rani et al., 2015) and can be carcinogenic and/or mutagenic in nature (Lithner et al., 2011). Based on their chemical composition, Lithner et al. (2011) made a hazard ranking model for the different types of plastic. The hazard scores for the previous mentioned plastics are given in table 1. A higher score reflects a higher health hazard. Of the most common plastic types PVC seems to be the most hazardous. This is due to the carcinogenic monomers and high amounts of additives found in PVC (Lithner et al., 2011). PVC has a higher density than seawater and could potentially be a big threat to benthic marine life.

Table 1: Commonly found plastics in the marine environment and their respective densities and typical usage. Based on Andrady (2011), Higalgo-Ruz et al. (2012) and Galloway (2015).

Plastic class	Abbreviation	Recycle code	Percentage recycled	Density (g/cm^3)	Hazard score	Typical products
(Seawater)				1.025		
Low-density Polyethylene	LDPE	4	6	0.91-0.93	11	Plastic bags, straws, bottles
High-density Polyethylene	HDPE	2	11	0.94	11	Milk and juice jugs
Polypropylene	PP	5	1	0.83 – 0.92	1	Bottle caps, rope
Polystyrene	PS	6	1	1.01 – 1.05	30-1628	Floats, food containers
Polyamide (nylon)	PA	7	0	1.01 – 1.15	50-63	Fishing nets
Polyethylene terephthalate	PET	1	20	1.35 – 1.37	4	Soft drink bottles
Polyvinylchloride	PVC	3	0	1.16 – 1.58	5001-10,551	Plastic film, tubes
Cellulose acetate	CA	7	0	1.24	N.A	Cigarette filters

2.3. Occurrence on the seafloor

To understand which organisms are most prone to plastic contamination it is important to know which areas accumulate the most plastic. Research has shown that the occurrence of plastic litter varies greatly among different sites and can also fluctuate over time within sites due to seasonal changes in hydrodynamics (Galgani et al., 1995). Macroplastics have been found on the bottom of every sea and ocean of the world with plastic abundancies ranging from zero to almost eight thousand items per km^2 (Galgani et al., 2015). In general, coastal seas contain higher amounts of litter than the open ocean due to residual ocean flow patterns and river inputs (e.g. Lee et al., 2006; Wei et al., 2012). Furthermore, marine litter tends to accumulate in low circulation areas due to the entrapment by sediments (Galgani et al., 1996; Schlining et al., 2013; Pham et al., 2014). Additionally, deep sea surveys have reported that submarine canyons can act as a accumulation zone and channel to transport litter to the deep sea (Galgani et al., 2000; Wei et al., 2012; Pham et al., 2014). Macro debris made of plastics with high densities should hypothetically be the most abundant type of macroplastic found on the seafloor. Indeed many studies have found derelict fishing gear, such as nets, traps and lines, to account for the majority of plastic debris found on the bottom (e.g. Vieira et al., 2015; Fischer et al., 2015; Pham et al., 2014).

Fishing nets and other gear are often made of high density plastics, such as nylon (table 1). Other non-buoyant plastic such as PET is also often found on the seabed in the form of plastic bottles (Galgani et al., 2000; Sanchez et al., 2013). Surprisingly, plastic bags, which are usually constituted of the buoyant LDPE, have been reported to be the most abundant form of macroplastic litter in coastal European seas (e.g. Galgani et al., 1995; Galgani et al., 2000; Pasquini et al., 2016). Their high occurrence on the seabed is probably the result of biofouling which can rapidly alter the buoyancy of plastic bags. The high variety in litter densities between studies on macrolitter abundance and occurrence on the seafloor can be seen from table 2.

Next to macroplastics, it is important to know the occurrence and abundance of microplastics in the sediment. Since many benthic organisms are small invertebrates that feed on microscopic particles, microplastics form a great threat as they are more likely to be ingested by these organisms than macroplastics. Research on the occurrence of microplastics in and on marine sediments has gained more interest in the last decade and has mostly been focused on beaches (Van Cauwenberghe et al., 2015). An overview of microplastic occurrences in the sediments across the world is provided in table 3. In general, microplastic concentrations seem to be higher in more densely populated regions (Browne et al., 2011). Fresh water inputs have also been found to be great sources of sedimentary microplastics (Vianello et al., 2013). Microplastics are mostly found in highly populated coastal areas, but their presence has even been showed in the deep sea (Van Cauwenberghe et al., 2013). Because deep sea sediments from the Mediterranean Sea, the Atlantic and Indian Ocean contained more microplastic per unit of volume than the overlying surface waters, Woodall et al. (2014) suggested that the deep sea is a major sink for microplastics. A lack of standardization in methods used in studies (more on this later) make it rather difficult to accurately establish trends concerning composition and occurrence of microplastics in the sediments (Hidalgo-Ruz et al., 2012). High-density non-floating types of plastic can be expected to contribute most to the composition of microplastics in the sediment. Interestingly, Vianello et al. (2013) found the low-density polymers polyethylene and polypropylene to be the most abundant type of microplastic in sediments of the Lagoon in Venice, Italy. Polystyrene spheres have also been found as a big contributor to microplastics in sediments (Claessens et al., 2011). It seems therefore also possible that light plastics could compose a big proportion of benthic microlitter. Since microplastics have a higher surface to volume ratio compared to macroplastic, they are more susceptible to biofouling and consequently to a decrease in buoyancy. Besides sinking microplastics, macroplastics present on the seafloor could fragment and therefore add to the microplastic concentrations in the sediment. The ageing of plastics at great depths is however unknown (Galgani et al., 2015) making it therefore difficult to say if macroplastic at the bottom of the sea will contribute greatly to the abundance of microplastics in the sediments.

Table 2: Overview of estimations of litter densities in different areas. Details on the different assays are given. Taken from Galgani et al. (2015).

Location	Habitat	Date	Sampling	Depth (m)	Density (min-max)	Plastic (%)
Southern China	Benthic	2009–2010	4 trawl (mesh not available)/1 dive	0–10	693 (147–5,000) items km^{-2}	47
France-Mediterranean	Slope	2009	17 canyons, 101 ROV dives	80–700	3.01 km^{-1} survey (0–12)	12 (0–100)
Thyrenian Sea	Fishing ground	2009	6 × 1.5 ha samples, trawl, 10 mm mesh	40–80	5,960 ± 3,023 km^{-2}	76
Spain-Mediterranean	Fishing ground	2009		40–80	4,424 ± 3,743 km^{-2}	37
Mediterranean Sea	Bathyal/abyssal	2007–2010	292 tows, otter/ Agassiz trawl, 12 mm mesh	900–3,000	0.02–3,264.6 kg km^{-2} (incl. clinker)	n.d.
Malta	Shelf	2005	Trawl (44 hauls, 20 mm mesh)	50–700	102	47
Turkey/Levantin Basin	Bottom/bathyal	2012	32 hauls (trawl, 24 mm mesh)	200–800	290 litter (3,264.6 kg km^{-2})	81.1
Azores, Portugal	Condor seamount	2010–2011	45 dives	185–256	1,439 items km^{-2}	No plastic/89 % fishing gear
Goringe Bank, NE Atlantic	Gettysburg and Ormonde seamounts	2011	4 ROV dives (124 h video, 4,832 photographs), total distance of 80.6 km	60–3,015	1–4 items·km^{-1}	9.9/56 fishing gear
US west coast	Shelf	2007–2008	1,347 sites (total, trawling, 38 mm mesh)	55–183	30 items km^{-2}	23
	Slope	2007–2008		183–550	59 items km^{-2}	n.d.
	Slope/bathyal	2007–2008		550–1,280	129 items km^{-2}	n.d.
Mediterranean Sea, France	Shelf/canyon	1994–2009 (16 years study)	90 sites (trawls, 0.045 km^2/tow, 20 mm mesh)	0–800	76–146 km^{-2} (0–2,540)	29.5–74

Table 2. Continued.

Location	Habitat	Date	Sampling	Depth (m)	Density (min-max)	Plastic (%)
Japan, offshore Iwate	Trench	Jamstek database	3 dives on 4,861 available,	299–400, 1,086–1,147, 1,682–1,753	15.9 items h^{-1}	42.8
Kuril-Kamchatka area (NW Pacific)	Trench/bathyal plain	2012	20 box cores (0.25 m^2) (Agassiz trawl, camera epi-benthic sledge)	4,869–5,766	60 → 2,000 micro-plastics m^{-2}	(Trawl samples: mostly fishing gear)
Fram Strait, Arctic	Slope	2002–2011 (5 surveys)	One OFOS camera tow $year^{-1}$, 5 transects (1,427–2,747 m^2)	2,500	3,635 (2002)–7,710 (2011) items km^{-2}	59
Northern Antarctic Peninsula and Scotia Arc	Slopes/bathyal	2006	32 Agassiz trawls	200–1,500	2 pieces only	1 plastic
Monterey Canyon, California	From margin to abyssal	1989–2011	ROVs, 2,429 km^2 in total	25–3,971	632 items km^{-2}	33
ABC islands, Dutch Caribbean	Sandy bottoms to rocky slopes	2000	24 video transects, submersibles	80–900	2,700 items km^{-2} (0–4590)	29

Table 3. Worldwide occurrence and abundance of microplastics in the sediments. Taken from Van Cauwenberghe et al. (2015).

Continent	Location	Location specification	Particle size	Measured abundance
Africa	Canary Islands	Beach	1 mm–5 mm	<1 – >100 g/L
America	Hawaii	Beach	1 mm–15 mm	541–18,559 items/260 L
	US	Florida subtidal	250 μm–4 mm	116–215 items/L
		Maine subtidal		105 items/L
	Brazil	Beach	2 mm–5 mm	60 items/m^2
	Brazil	Beach	0.5 mm–1 mm	200 items/0.01 m^2
			1 mm–20 mm	100 items/0.01 m^2
	Hawaii	Beach	250 μm–10 mm	0.12%–3.3% plastic by weight
	Brazil	Tidal plain	1 mm–10 cm	6.36–15.89 items/m^2
	Chile	Beach	1 mm–4.75 mm	<1–805 items/m^2
	Québec	River sediment	400 μm–2.16 mm	52–13,832 beads/m^2
	Nova Scotia	Beach	0.8 μm–5 mm	20–80 fibres/10 g
Asia	Singapore	Beach	1.6 μm–5 mm	0–4 items/250 g dry
	India	Ship-breaking yard	1.6 μm–5 mm	81.4 mg/kg
	South Korea	High tide line	2 mm–10 mm	913 items/m^2
	India	Beach	1 mm–5 mm	10–180 items/m^2
	South Korea	Beach dry season	1 mm–5 mm	8205 items/m^2
		Beach rainy season		27,606 items/m^2
	Singapore	Mangrove	1.6 μm–5 mm	36.8 items/kg dry
	NW Pacific	Deep sea trench	300 μm–5 mm	60–2020 items/m^2
	South Korea	Beach	50 μm–5 mm	56–285,673 items/m^2
Europe	UK	Beach	1.6 μm–5 mm	0.4 fibres/50 mL
		Estuary		2.4 fibres/50 mL
		Subtidal		5.6 fibres/50 mL
	Sweden	Subtidal	2 μm–5 mm	2–332 items/100 mL
	UK	Beach	1.6 μm–1 mm	<1–8 items/50 mL
	UK	North Sea beach	38 μm–1 mm	0.2–0.8 fibres/50 mL
		English Ch. beach		0.4–1 fibres/50 mL
	Belgium	Harbour	38 μm–1 mm	166.7 items/kg dry
		Continental Shelf		97.2 items/kg dry
		Beach		92.8 items/kg dry
	Portugal	Beach	1.2 μm–5 mm	133.3 items/m^2
	Germany	Urban beach	1 mm–15 mm	5000–7000 items/m^3
		Rural beach		150–700 items/m^3
	Germany	Tidal flat	1.2 μm–5 mm	0–621 items/10 g
	Italy	Sub-alpine lake	9 μm–5 mm	1108 items/m^2
	Greece	Beach	1 mm–2 mm	57–602 items/m^2
			2 mm–4 mm	10–575 items/m^2
	Belgium	High tide line	38 μm–1 mm	9.2 items/kg dry
		Low tide line		17.7 items/kg dry
	Italy	Subtidal	0.7 μm–1 mm	672–2175 items/kg dry
	Germany	Beach	<1 mm	1.3–2.3 items/kg dry
	Slovenia	Beach	0.25–5 mm	177.8 items/kg dry
		Infralittoral		170.4 items/kg dry
Worldwide		Deep sea	5 μm–1 mm	0.5 items/cm^2

3. Direct effects of plastic on benthic organisms

It is now clear that plastic is present on the seabed and as long as the input of plastics to the marine environment continues, more and more plastics will sink to the bottom. Due to the omnipresence of all types and sizes of plastics in and on the sediment, the question arises how this will affect the benthos. The harmful effects of plastics on biota of higher trophic levels has been widely shown with marine birds, turtles and mammals receiving the most attention (Kühn et al., 2015). The research on the effects of plastic on benthic organisms is however gaining attention (Kühn et al., 2015). As the scope of this review is to assess the impact of plastic on benthic organisms, only studies concerning species such as bivalves, polychaetes, crustaceans, echinoderms and demersal fish are reviewed.

3.1. Habitat modification

Just the presence of plastic on the sediment can already alter the composition of sediment associated organisms. Soft-sediment habitats are especially susceptible as the presence of plastic alters the characteristics of this biotope by providing hard substrata (Katsanevakis et al., 2007). Uneputty and Evans (1997) studied the effects of macroplastics on the assemblages of organisms on the tidal flats of Ambon Bay (Indonesia) . By comparing litter with litter-free areas of the littoral zone, the authors found that in the sediment below litter items diatoms were at lower densities. This was presumably caused by the blockage of light by the plastic items. Interestingly, the abundance of meiofauna (e.g. polychaetes, nematodes, copepods) was much higher in the sediment collected beneath litter items than in sediment collected from litter-free areas. Authors pose that this might be caused by an increased amount of bacteria forming under litter items. In a research performed in coves of the Saronikos Gulf (Aegean Sea), Katsanevakis et al. (2007) studied the impact of plastic bottles on the abundance and community structure of soft-bottom epibenthic megafauna. It was found that the litter provided refuge sites for mobile species as well as a hard-substratum for sessile species that would otherwise not colonize the soft-bottom habitat. As a result the total abundance and number of species was higher in litter impacted areas. This result could be interpreted as a beneficial effect of plastic on soft-bottom habitats, the presence of litter may however facilitate the displacement of indigenous species by invasive species which is against the policy of conserving habitats (Katsanevakis et al., 2007).

3.2. Uptake and effects

Besides habitat modification, an important issue of marine litter arises from its uptake by organisms. Many benthic organisms are suspension or deposit feeders or detritivores (Wright et al., 2013). Since microplastics exhibit similar or even smaller sizes than sediment grains and many lower trophic-level organisms capture almost anything of the appropriate size (Moore, 2008), the ingestion of plastics by benthic organisms seems inevitable. Indeed, many organisms either collected in the field or investigated in the lab have shown to uptake plastics. Karlsson et al. (2017) screened nine invertebrate species collected from the North Sea for microplastics. They found microplastics in eight out of the nine species collected, highlighting the wide variety of organisms contaminated with microplastics. Another interesting finding was that there was a difference in plastic load between organisms with different feeding strategies. Suspension feeders such as the blue mussel (*Mytilus edulis*) and brittle star (*Ophiura* sp.) contained the highest concentrations of microplastics. This finding is consistent with the research of

Setälä et al. (2016) who showed that bivalves ingested more 10 µm polystyrene microbeads than benthic deposit feeders and free swimming crustaceans in a mesocosm experiment. Blue mussels have even been shown to contain a thousand times more microplastics per volume than the surrounding water and sediment (Karlsson et al., 2017). Mussels are often selected as model species because they are sedentary filter feeding animals that inhabit a wide geographic range (Van Cauwenberghe et al., 2015). Plastic research in mussels is furthermore important for assessing the risk of mussel consumption by humans. Other human food sources such as crustaceans have also been shown to be contaminated with plastic. Murray and Cowie (2011) found 83% of the scavenger *Nephrops norvegicus* sampled in the Clyde Sea contained plastic in their gut. The most found plastics were filaments, sometimes observed as tangled balls inside the animals. They state that the high prevalence of plastic may have implications for Norway's most valuable fishery.

The ingestion of plastic by different benthic organisms has also been widely demonstrated in laboratory settings by exposing organisms to food, water or sediment spiked with plastic microparticles (e.g. Thompson et al., 2004; Browne et al., 2008; Watts et al., 2014; Sussarellu et al., 2016). Such proof of principle experiments are important to assess which animals ingest plastic and are in most potential danger. For example, some organisms such as sea cucumbers have been shown to selectively feed on microplastics (Graham and Thompson, 2009). Besides ingestion, adherence has also been described as a way for animals to uptake plastic particles (Kolandhasamy et al., 2018). It is apparent that many different species of benthic organisms are able to uptake plastics. However the question then arises if this has any detrimental effects on those organisms. After all, if plastics are excreted again shortly after ingestion the expected impact of these plastics would be small. Studies focusing on the impact of plastic ingestion display mixed results between and within different feeding guilds. For example, Browne et al. (2008) showed than microplastic particles translocated from the gut to the circulatory system of the filter-feeding blue mussel and were retained for over 48 days. However, no biological effects such as a change in filter-feeding activity or oxidative status of hemolymphs was detected. This is in line with the results of Van Cauwenberghe et al. (2015) who found no significant changes in cellular energy allocation between blue mussels exposed to high concentrations of PS particles (110 particles/mL, 14 days) compared to control treatments. On the contrary, a different type of bivalve, the Pacific oysters (*Crassostrea gigas*), exposed to polystyrene microbeads (2 and 6 µm in diameter; 0.023 mg/L) for two months displayed significant changes in microalgae consumption and absorption efficiency (Sussarellu et al., 2016). Furthermore, oyster reproduction was heavily impaired with a 23% reduction in sperm velocity. Scavenging crustaceans have also been investigated. Decapod *N. norvegicus* fed with squid spiked with polypropylene fibers for eight months showed a decrease in growth and a reduction in blood protein stored lipids compared to fed controls (Welden and Cowie, 2016). In a similar experiment performed by Watts et al. (2015), the crab *Carcinus maenas* also showed a reduction in growth and energy balance. Deposit-feeding worms have also been the subject of several plastic effect assessments (Van Cauwenberghe et al., 2015). For example, the lugworm *Arenicola marina*, a keystone species that bioturbates and irrigates the sediments of intertidal flats in Northern Europe (Wright et al., 2013), has been exposed to spiked sediment in multiple studies (Van Cauwenberghe et al., 2015). In an experiment where *A. marina* were exposed to high concentrations of PVC (5% by weight, mean diameter of 130 µm) for a month, Wright et al. (2013) found a reduction in overall feeding activity of 25%. In an exposure study spanning 14 days with sediment spiked with 110 PS particles/g sediment (10, 30, 90 µm) there was however no significant effect found on energy metabolism. As *A. marina* is an important prey species for fish and birds (Wright et al., 2013), there is potential for the biomagnification of plastics in higher trophic levels. Trophic transfer of microplastics has been demonstrated before (Setälä et al., 2014). The great variety in found effects for different biota can be seen in the overview of different exposure studies presented in table 4.

The presence of plastic on the seafloor can have multiple effects on benthic organisms. Macroplastics can offer refuge sites or hard substrates which can alter the species abundance and composition. It can furthermore smother underlying organisms e.g. by the blockage of light. The (potential) uptake of (micro)plastics by animals has been demonstrated in many benthic species but exposure studies display different results. Where some organisms exhibit reduced growth, impaired fecundity, translocation of ingested particles or altered feeding activity, other organisms are found to be not affected at all. As can been seen from table 4, there is a lack of consistency in the assays mostly related to exposure material and time. Most studies on the effects of microplastic have used only one type of plastic or one size class. In a natural setting, plastics of many different types and sizes would be present. Furthermore, while most studies have used microbeads in experiments, microfibers are actually the most commonly found type of microplastics in the marine environment (Wright et al., 2013) and should therefore deserve more attention. Additionally, concentrations of plastic particles are generally much higher in lab experiments than observed in the wild (Van Cauwenberghe et al., 2015). The exposure time is also often relatively short whereas plastic pollution is a long time problem. These studies have shown a wide range of effects of plastic but the experimental conditions used do not often resemble natural conditions. This questions the applicability of results from laboratory studies to natural settings. The uptake of plastic seems to have detrimental effects on many benthic organisms, however more research is required for a more realistic and complete assessment of the problem.

Table 4: Direct effects of (micro)plastic exposure to benthic organisms investigated under laboratory conditions. Details on the used method are given. Adapted from Van Cauwenberghe et al. (2015).

Biota		**Feeding strategy**	**Exposure route**	**Particle and plastic type**	**Concentration and exposure time**	**Assay**	**Effect**	**Reference**
Bivalve	*Mytilus edulis*	Filter-feeder	Spiked seawater	Polystyrene microbeads 3.0 and 9.6 µm	15000 particles / 350 mL seawater 3 hours	Gut and hemolymph analysis	Microplastics translocate to the circulatory system	Browne et al. 2008
	Mytilus edulis		Spiked seawater	Polystyrene microbeads 10, 30 and 90 µm	110 particles / mL seawater 14 days	Cellular Energy Allocation analysis	No significant effects found	Van Cauwenberghe et al. 2015
	Mytilus edulis		Spiked seawater	Polyester microfibers >100 µm	2000 particles / L seawater 48h	Tissue analysis	Adherence is a novel way for animals to uptake plastics	Kolandhasamy et al. 2018
	Crassostrea gigas	Filter-feeder	Spiked seawater	Polystyrene microbeads 2 and 6 µm	0.023 mg / L seawater 2 months	Ecophysiological analysis of cellular responses; fecundity; and offspring development	Microplastic cause feeding modifications and reproductive disruption	Sussarellu et al. 2016
Polychaete	*Arenicola marina*	Deposit-feeder	Spiked sediment	Unplasticised polyvinylchloride 130 µm mean diameter	1 and 5 % by weight 48 hours and 4 weeks	Energy budget Feeding activity	Energy budget and feeding activity were impaired	Wright et al. 2013

Table 4: Continued.

Biota		**Feeding strategy**	**Exposure route**	**Particle and plastic type**	**Concentration and exposure time**	**Assay**	**Effect**	**Reference**
Polychaete	*Arenicola marina*	Deposit-feeder	Spiked sediment	Polylactic acid, polyethylene and polyvinylchloride 102.2 - 235.7 µm mean diameter	0.02%, 0.2%, 2% by wet weight 31 days	Faecal analysis	Microplastics altered metabolism and burrowing activity	Green et al. 2016
Crustacea	*Carcinus maenas*	Predator, scavenger, detritivore	Spiked seawater Spiked mussels Pre-exposed mussels	Polystyrene microspheres 8-10 µm	940000, 40000 particles / L 150000, 4000 particles / g of mussel 16 hours, 24hours, 21days	Tissue analysis Faecal analysis	Microplastics were retained for up to 21 days	Watts et al. 2014
	Carcinus maenas		Spiked food	Polypropylene microfibers 500 µm	0.3, 0.6, 1 % by weight 1 month	Energy budget	Reduction in consumption and growth rate	Watts et al. 2015
	Nephrops norvegicus	Predator, scavenger	Spiked food	Polypropylene microfibers 3-5 mm	5 fibers / 1,5 g food 8 months	Energy budget Feeding rate	Reduced body mass, blood protein and stored lipids.	Welden and Cowie 2016
Echinoderm	*Holothuria sp.*	Deposit/ suspension feeder	Spiked sediment	Nylon and PVC fragments 0.25 – 15 mm	2, 10, 65 g / 600 mL silica 4 hours	Ingestion analysis	Microplastic are selectively ingested	Graham and Thompson 2009

4. Indirect effects of plastics on benthic organisms

Besides the direct effects of plastic on organisms , there has been a lot of concern on the indirect effects. Indirect effects of plastics (mainly microplastics) are here defined as effects induced by plastics that act as a vector for chemicals (Van Cauwenberghe et al., 2015). Owing to their large molecular size, plastics are considered biochemically inert and not dangerous to the endocrine system (Teuten et al., 2009). However, due to their specific characteristics, plastics can carry smaller molecular sized chemicals which do have the potential to harm organisms. These chemicals can be hydrophobic in nature and are therefore adsorbed to plastics from surrounded seawater or sediment (Teuten et al., 2009). Furthermore, monomers and additives created or used in manufacturing of the plastic create additional toxic effects (see 'Plastic properties').

The presence of many different persistent organic pollutants (POPs) have been detected on different types of microplastics and from a variety of geographic areas (e.g. Rochman et al., 2013; Mato et al., 2001; Frias et al., 2010; Mendoza et al., 2015). Various conditions influence the adsorption and desorption of chemicals to microplastics, such as size and color of the plastic but also environmental factors such as salinity, temperature and pH of seawater (Ziccardi et al., 2016). The polymer type is also a big factor in determining the magnitude of contamination e.g. polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs) are found in higher concentrations on HDPE, LDPE and PP in comparison to PET and PVC under the same sorption conditions (Van Cauwenberghe et al., 2015). But while the evidence on contaminated plastics in the marine environment are plentiful, only few studies have examined whether these plastics actually transfers these chemicals to organisms (Browne et al., 2013). Some of these studies have focused on benthic organisms. Devriese et al. (2017) fed *N. norvegicus* with gelatin spiked with PCB-contaminated polyethylene or polystyrene microbeads (6 and 500-600 µm) for up to three weeks. They concluded that there was no significant bioaccumulation of PCBs in the exposed decapods caused by the presence of PCBs on ingested microplastics. Browne et al. (2013) exposed the lugworm *A. marina* to microplastics presorbed with nonylphenol, phenanthrene, Triclosan and PBDE-47. It was shown that concentrations of 5% contaminated microplastics (PVC, 230 µm) in clean sediment was sufficient to disrupt health-related ecophysiological functions due to the transfer of pollutants and additives to the worms' tissue. However, while the sorption of nonylphenol and phenantrene was much higher for PVC than for sand, the polychaetes exposed to contaminated sand accumulated >250% more of these chemicals than polychaetes exposed to clean sand with 5% contaminated PVC. On the contrary, Besseling et al. (2012) showed that the addition of polystyrene microbeads (400−1300 µm, 0.074% based on dry weight) to naturally PCB-contaminated sediment increased bioaccumulation of PCBs by a factor of 1.1-3.6 in comparison to contaminated sand alone. Noteworthy is that this effect decreased with increasing microplastic load. The bioaccumulation of pyrene has been demonstrated in mussels (Avio et al., 2015) by exposing them to polyethylene and polystyrene microplastics (<100 µm) with realistic contamination loads. Authors do however state that the observed adverse effects of contaminated microplastics did not differ from "clean" microplastics and mussels seem to activate defense mechanisms more towards the physical rather than the chemical characteristics of microplastics.

The hypothesis that microplastics act as a vector for bioaccumulation of chemical compounds in organisms has gained popularity and has been a big contributor to the perceived risk of microplastics (Koelmans et al., 2016). It has almost obtained a paradigm status now (Koelmans et al., 2016), but critical reviews suggest that the contribution of plastics on bioaccumulation of chemical compounds is negligible (Ziccardi et al., 2016; Koelmans et al., 2016). Computer models investigating plastics as carriers for POPs to benthic animals actually predicted that plastics will mostly reduce bioaccumulation due to the adsorption of chemicals by ingested "clean" plastics followed by excretion of these plastics (Koelmans et al., 2013). The potential danger of hydrophobic organic compounds (HOCs) associated with microplastics is probably much lower than HOCs transported through natural organic carbon (Ziccardi et al., 2016). Furthermore, studies on chemical transfer by microplastics to benthic organisms have mainly focused on POPs absorbed in the environment instead of harmful additives used in production. Future research should focus more on the hazards of plastic additives (Hermabessiere et al. 2017) as well as the transfer of more realistic concentrations of chemicals from plastics to organisms for a better comprehension of the impact plastic has as a vector for chemicals to benthic marine organisms.

5. Sampling and extraction methods

The previous chapters have reviewed the available literature on the impact of plastic on the benthos. However caution must be taken when interpreting results. To make good estimations of the impact of plastic debris on benthic ecosystems worldwide, it is important to compare different sites to look for trends and predict which areas are the most affected. There is however a lack in consistency in both sampling and extraction techniques for quantifying benthic macro- and microplastic. Studies on plastic in marine sediments are typically focused on continental shelves of coastal areas (Van Cauwenberghe et al., 2013). Inaccessibility with accompanied sampling difficulties and costs normally do not allow for sediment investigation in deeper waters (Galgani et al., 2015). Trawling of the seabed is suggested to be the best way to investigate marine bottom macrolitter as acoustic methods fail to discriminate between different litter items and can overlook small objects (Galgani et al., 2015). As most nets create a sample bias, pole trawling is considered the most consistent method for assessing marine debris for larger areas (Galgani and Andral, 1998). Pole trawling can however not be performed on hard substrates and does not provide information on specific locations of specific plastic items (Watters et al., 2010). This in combination with the destructive nature of pole trawling, causes the use of submersibles with cameras to sometimes be the preferred option (Galgani and Andral, 1998; Miyake et al., 2011; Fabri et al., 2014). Making use of camera footage to estimate plastic litter quantities is prone to errors as some plastics may be overlooked as weathering and biofouling may have altered their physical appearance. Moreover, many trawling studies use nets with different mesh sizes or have made different numbers of trawls and at different speeds (table 2). Where some studies use mesh sizes of around 10 mm (e.g. Sánchez et al., 2013; Ramirez-Llodra et al., 2013) others have used nets with mesh sizes twice (e.g. Mifsud et al., 2013) or even four times as big (Keller et al., 2010) and sometimes mesh size is not even mentioned (Zhou et al., 2011). As smaller mesh nets will retain more litter items than large mesh nets, a comparison of litter items per area or volume unit between different sites where studies were performed using different mesh sizes is not without caution. Different opening sizes, amount of trawls and speeds will all have an influence on the amount of plastic gathered.

The rapid increase in microplastic research has also resulted in an inconsistency in methods used to quantify microplastics present in both sediments and water columns (Hidalgo-Ruz et al., 2012; Van Cauwenberghe et al., 2015). Resulting from this is that comparisons between microplastic concentrations from different studies is either not possible or require assumption based calculations (Van Cauwenberghe et al., 2015). Most unconformities can be attributed to one of three categories: differences in size limits, effectiveness of the used extraction techniques or differences in sampling method (Van Cauwenberghe et al., 2015).

There is no fixed definition of microplastics in terms of size. Where some authors use an upper size limit of 5mm (Moore, 2008; Watts et al., 2014), others have used an upper size limit of 1mm (e.g. Browne et al., 2011; Claessens et al., 2014). Extraction and identification of microplastics becomes increasingly more difficult with decreasing size (Van Cauwenberghe et al., 2015) which is why some studies implemented a lower size limit, such as 1 or 2 mm (e.g. Jayasiri et al., 2013; Heo et al., 2013). As different size ranges for microplastics are used throughout the literature, a lot of information on occurrence and distribution of microplastics is lost (Van Cauwenberghe et al., 2015). To overcome this problem suggestions have been made to further categorize microplastics based on size (Hidalgo-Ruz et al., 2012; Galgani et al., 2013). Not including small microplastics will lead to an underestimation of microplastic concentrations in the sediment.

The units of microplastic abundance often differ between studies due to differences in sampling techniques (Van Cauwenberghe et al., 2015). Hidalgo-Ruz et al. (2012) reviewed the different methods used for identification and quantification of microplastics. They show that microplastic researchers sometimes sample quadrant, others sample linear extensions and some sample different depth strata. A large variety of tools is also mentioned from spoons and hands to box cores and Ekman and van Veen grabs. Resulting from these different sampling techniques, many studies report different units for the abundance of microplastics. Widely used reporting units are items per m^2, items per m^3, g per kg and mL per L (table 3). Conversion between the former two requires an understanding of the sampling depth and conversion between the latter two requires detailed information on sediment characteristics which is almost never given (Van Cauwenberghe et al., 2015).

After benthic sediment samples are taken, the next step is to extract the microplastics from the sand or mud. Density separation is the most common method to separate microplastics from benthic sediment samples (e.g. Thompson et al., 2004; Ng and Obbard, 2004; Vianello et al., 2013; Claessens et al., 2013). Most studies have used the method as described by Thompson et al. (2004) or a modification thereof. Using this method the sediment is drained in a concentrated NaCl solution on which microparticles with a lower density will float. The problem with this method is that the density of this solution is 1.2 g/cm^3 and as can be seen from table 1 many types of plastic exceed this density. The use of NaCl will therefore highly underestimate the abundance of microplastics in the sediments (Van Cauwenberghe et al., 2015). To overcome this issue and sort out a bigger portion of present microplastics some studies have used a zinc chloride (ZnCL) or sodium iodide (NaI) solution with densities of 1.5 and 1.6 g/cm^3, respectively (Liebezeit and Dubaish, 2012; Van Cauwenberghe et al., 2013). The use of density separation with a relatively low density solution has led to an underestimation of dense microplastics making trends on abundance and composition of benthic microplastics incomplete.

Another discrepancy is visible in the identification process of microplastics. Many microplastic assessments make use of visual inspection (Hidalgo-Ruz et al., 2012) results can therefore be biased as some types of plastic are more easily recognized such as microfibers (Van Cauwenberghe et al., 2015). Future studies should make more use of infrared spectroscopy as this is the best method for identification of microplastic (Hidalgo-Ruz et al., 2012).

For a better assessment of the impact of microplastics on benthic marine organisms there is an urgent need to standardize protocols for the assessment of both macro and microplastics in and on sediments. This will allow not only for a better comparison between areas, but it will also help in predicting trends and gaining knowledge on potential ways of conservation.

6. Conclusion and discussion

The ocean sediments have been hypothesized to be an ultimate sink for plastics (Woodall et al, 2014). High density plastics will be the first to descend in the water column but even low density plastics can reach the benthic environment through biofouling and other types of modification. Studies on the occurrence of plastic debris in and on the sediments have rapidly increased in the last decade. But while these studies have greatly improved our understanding of this issue, a wide variety of methods are used in collecting and analyzing samples (Van Cauwenberghe et al., 2015; Galgani et al., 2015). Macroplastic assessments have often used (pole) trawling with different net openings, mesh sizes, amount of trawls and trawling speeds. Investigations on sedimentary microplastic often differ in their extraction method to separate the plastic particles from collected sediments. These methods are usually based on separation by differences in density, but the incorporated extraction fluids vary greatly. These different solutions have different densities resulting in unequal extraction efficiencies. Lack of standardization in assessment protocols makes it difficult to compare different studies and establish trends. There are some indications that certain marine areas accumulate more plastics on the bottom than others due to low circulation or close proximity to urban areas. Future research should focus more on which specific benthic communities are most at risk to plastic exposure by sampling wide geographic areas in a standardized matter .

The presence of macroplastics on the sediment has been shown to alter certain benthic communities. Especially soft sediment communities seem to be most at risk. Plastics can smother organisms by the blockage of sunlight, but they can also offer substrates for sessile species or provide refuge sites. This sometimes even leads to an increase in species richness. The ingestion of plastic by benthic organisms poses a potential threat. The contamination of benthic organisms by plastics has been shown for many phyla. Filter feeding organisms such as mussels seem more susceptible to plastic uptake than deposit feeders. The question whether exposed benthic organisms are actually heavily affected by the exposure to plastic remains however uncertain. Suggested direct effects of plastic uptake include impairments of feeding activity, growth, energy expenditure and fecundity. Some studies find however no significant effects at all of ingested plastics. Furthermore, most effect assessment studies have used unrealistic exposure conditions that do not resemble the natural environment in its current state. The applicability of the outcomes of these laboratory studies must therefore be taken with caution. Future effect assessments should compare the effects of realistic concentrations of various compositions of plastics both in types and sizes. This will increase our understanding of the true impact plastic has on benthic organisms today.

The potential for plastic to act as a carrier for chemical pollutants that transfer to organisms upon uptake has become a popular theory that is often given in descriptions of the risks of plastics in the marine environment. Studies backing this claim are however sparse and the few studies that have looked at the transfer of chemicals by microplastics in benthic organisms did not find convincing evidence that plastics have negative indirect effects on organism health. It seems likely that the contribution of plastics to bioaccumulation of POPs in benthic organisms is negligible compared to other sources occurring naturally and in much higher numbers. Future research should instead focus more on the transfer of chemical compounds used in the production of plastic.

Overall, both macro and microplastics are potentially impacting benthic marine organisms. However, the magnitude of this impact in its current state needs further examination due to a lack of standardization in methods and realistic experiments on plastic exposure.

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