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Microbial hitchhikers of marine plastic debris: human exposure risks at bathing waters and beach environments

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ABSTRACT

Keywords: marine plastic debris, microplastics, biofilms, microorganisms, pathogens, bathing water quality, human health

1. Introduction

Marine plastic debris is an environmental pollutant of growing concern, with detrimental effects on aquatic reptiles, birds and mammals already well documented (Gregory, 2009; Derraik, 2002). The durable, light weight and inexpensive nature of plastic has made it a ubiquitous choice for many industrial and consumer products (Osborn and Stojkovic, 2014). Its widespread use has facilitated its entry and accumulation within coastal waters and beach environments, with global changes in rainfall, wind speed, and more frequent flood and storm events predicted to further increase the amount of stranded and drifting plastics in the coastal zone (Young *et al*, 2011; Gulev and Grigorieva, 2004; Meier and Wahr, 2002; Goldenberg *et al*, 2001).

Globally, more than 200M tonnes of plastic is produced annually (Ivar do Sul and Costa, 2014), with approximately 4.8 – 12.7M tonnes of plastic waste entering the ocean in 2010 (Jambeck *et al*, 2015). Marine plastic debris includes large, macro particles such as carrier bags, bottles, packaging, and fishing gear (Eriksen et al., 2014), and more commonly microplastics and nanoplastics (Andrady, 2011). Sources of plastic debris entering the oceans are numerous, with the majority a result of direct disposal of litter from land and at sea (including the illicit dumping of fishing gear), the blowing of litter from landfill sites, public littering, and losses from ship transport and accidental spills (Barnes *et al*, 2009). In a study assessing plastic debris in Latin America and the wider Caribbean region, land-based plastic debris was found to be the most abundant along continental shores (Ivar do Sul and Costa, 2007), whilst fishing-related plastics are often more prevalent on remote islands

(Barnes *et al*, 2009). Recently, there has been an increasing focus on "microplastics", which is defined by the National Oceanic and Atmospheric Administration (NOAA) as plastic particles less than 5 mm in diameter (NOAA, 2009). Sources of microplastics in marine environments include "primary" microplastics present in cosmetic care products such as facial exfoliating scrubbers, clothes fibres washed into the sea through sewage effluent from washing machines, and industrial discharge of virgin plastic production pellets and those used as air-blasting media (Browne *et al*, 2011; Cole *et al*, 2011; Fendall and Sewall, 2009; Derraik, 2002). In addition, "secondary" microplastics frequently enter waterways through the breakdown of macro particles by a combination of physical, biological and chemical processes (Ryan *et al*, 2009; Thompson *et al*, 2004). Rivers, tides, wind, heavy rainfall, and storm and sewage discharge facilitate dispersal of microplastics within the marine environment (Reisser *et al*, 2013), with an estimated 5.25 trillion plastic particles weighing approximately 269,000 tonnes now floating in the sea (Eriksen *et al*, 2014).

The impacts of marine plastic debris go beyond simply posing a threat to marine wildlife (Figure 1). Marine plastics can lead to significant economic losses by interfering with the shipping and fishing industries – e.g. drifting fishing gear becoming entangled on propellers and rudders; large floating debris causing damage by collision; and ghostnets trapping commercial fish (Pichel *et al*, 2007; Sheavly and Register, 2007). Furthermore, floating plastic bags are often the cause of blocked and damaged water pumps, causing substantial vessel damage, costly repairs and danger to operating staff (Sheavly and Register, 2007; Aliani *et al*, 2003). Plastic debris also poses a significant threat to the tourism industry. For example, beaches polluted with medical and sanitary waste poses a public health risk, devalues the experience of beachgoers, and can often require costly beach-cleaning efforts (Moore, 2008). With quantities of beach-cast plastic expected to rise due to increased frequency of storm and severe weather events, coastal areas dependent on tourism are likely to face a number of socio-economic challenges (McIlgorm *et al*, 2011).

Plastic debris can also provide a novel mechanism for the spread of invasive and alien species, in addition to that facilitated by natural substances like rafts of vegetation, wood, or pumice (Bryan *et al*, 2012; Minchinton, 2006; Jokiel, 1990). A diverse range of organisms have already been found colonising macro-plastics, and in some cases have led to the introduction of non-native species into new habitats (Gregory, 2009; Barnes 2002*a*;*b*). Until very recently, however, little attention has been paid to the concept of plastic providing a novel means of spatial and temporal transport for microorganisms across marine and coastal environments (Caruso, 2015). The physical properties of plastic can provide a unique habitat capable of supporting diverse microbial communities (Zettler *et al*, 2013). The buoyant and persistent nature of plastic could contribute significantly to the survival and transport, across large distances, of microorganisms that associate with its surface. The biofilms that make up this so-called “plastisphere” could also be a significant reservoir for pathogenic microbes, faecal indicator organisms (FIOs) and harmful algal bloom (HAB) species. Plastic debris could be acting as a potential vector for the wide-scale dissemination of these organisms (Zettler *et al*, 2013; Masó *et al*, 2003).

A few recent studies have shown evidence for the formation of biofilms by bacteria and FIOs (such as *E. coli*) on plastic water distribution pipes (Yu *et al*, 2010; Lehtola *et al*, 2004), and the persistence of pathogens (such as *Vibrio* spp.) on plastic debris (McCormick *et al*, 2014; Quilliam et al., 2014; Zettler *et al*, 2013). However, the ability of microorganisms to persist on beach-stranded plastic debris, and increase dissemination of potentially pathogenic microbes in coastal zones needs addressing to allow regulators and beach managers to make more informed decisions about public safety at beach environments. In Europe, the quality of bathing water and safety of beaches is governed by the EU Bathing Water Directive (BWD; 2006/7/EC). The BWD sets standards for microbial water quality and guides monitoring programmes to regulate bathing waters via the use of FIOs for the assessment of faecal pollution. The BWD also requires the production of a Bathing Water Profile (BWP) for all designated EU bathing waters (Mansilha *et al*, 2009), which contains details on the nature of possible pollution sources that could have negative impacts on a bather's health (Schernewski *et al*, 2012). Designations such as the Blue Flag award are also largely driven by the BWD.

Epidemiological studies have reported the relationship between bathing water quality and the occurrence of adverse human health effects such as gastrointestinal (GI) symptoms, respiratory diseases, and eye, nose and throat infections (Wade *et al*, 2006, Zmirou *et al*, 2003; Prüss, 1998). Whilst most of these studies have focused on waters impacted by municipal-wastewater effluent, the impacts of other diffuse sources of pollution remain relatively unexplored (Soller *et al*, 2010). With the potential for plastic providing a possible site for pathogen and FIO attachment and the dissemination of these organisms in the marine environment, we require a better understanding of these processes in order to not compromise beach safety and management of risks to bathing water quality. Assessing beach and bathing environments for beach-cast plastic debris and analysing it for associated pathogens could provide a better insight into the quality of European bathing waters through the production of a more detailed BWP, as well as to qualify plastic debris as a potential indicator and carrier of pathogens of risk to human health. This could further help prevent economic losses associated with beach closures, and enable beaches to maintain their Blue Flag status (Schernewski *et al*, 2012; Wyer *et al*, 2010).

Beaches and coastal environments form some of the most ecologically and socio-economically important habitats worldwide (Harley *et al*, 2006), and ecosystem services in these areas are already facing significant pressure from anthropogenic activities (Quilliam *et al*, 2015; Schlacher *et al*, 2007*a*; 2006). Against a backdrop of climate change, and projected changes in ocean currents, upwellings, water temperature and wind speed, the persistent multi-pollutant effects of plastic debris in coastal environments increases the urgency to understand the risks of human exposure to plastic pollution and inform more sustainable beach management options. The aim of this review is to explore the potential for marine plastics to serve as a mechanism for the persistence and transmission of FIOs and pathogenic or harmful microorganisms, and the pathways of human exposure risk in coastal environments.

2. Biofilm formation and the "Plastisphere": a man-made ecological habitat

Biofilms are formed by the microbial secretion of extracellular polymeric substances (EPS) that act as a type of architectural scaffolding, or support 'glue', for microbial cells to 'stick' together, and further provide a protective environment that enables microbial cells to grow in hostile habitats and facilitate dispersal (Hall-Stoodley and Stoodley, 2005). Microorganisms can form biofilms on any artificial or natural surface, such as medical implants, rocks, copper and plastic pipes of water distribution systems (Costerton *et al*, 2005; Lehtola *et al*, 2004; Lee *et al*, 2003). Some studies have demonstrated that the surfaces of different types of plastics, for e.g. polyethylene (PE) and polyethylene terephthalate (PET), when submerged in seawater can become rapidly colonised by heterotrophic bacteria, and that these organisms are able to persist for longer periods than those in the surrounding seawater (Webb *et al* 2009; Lobelle and Cunliffe, 2011). Interestingly, these studies had found lower levels of EPS secretion and bacterial colonisation on these plastic surfaces compared to that on other substrates.

Successional changes in bacterial colonisation of plastics with varying degrees of hydrophobicity have been demonstrated in seawater, with early-stage colonisation often marked by higher species richness (Jones *et al* 2006; Jackson *et al*, 2001). *Alphaproteobacteria* was found to be the most dominant group of colonising bacteria, with *Gammaproteobacteria* mainly occurring during the early colonisation stages (Lee *et al*, 2008; Jones *et al*, 2006). However, a range of environmental factors such as grazing pressure from macrozooplankton could also significantly influence bacterial colonisation of plastic surfaces (Webb et al, 2009). *Gammaproteobacteria* are an ecologically diverse group of gram-negative bacteria that contain a number of pathogens harmful to human health, and which include *Salmonella* spp. and *Vibrio cholera*. Since pathogenic strains of *Vibrio* are recognised to readily colonize a diverse range of marine substrates, their potential to colonise plastic debris requires urgent investigating, particularly in light of prescient knowledge that plastic debris can be easily dispersed in the marine environment (Zettler et al., 2013).

Reports on biofilms on plastic waste in the environment are limited (summarised in Table 1). Biofilm formation on plastic debris was first reported in 1972 in the Sargasso Sea, where bacterial communities were found colonising floating microplastic particles (Carpenter *et al*, 1972; Carpenter and Smith, 1972). Zettler *et al* (2013) conducted the first study of its kind, which characterised the composition of microbial communities colonising micro and macro pieces of PE and polypropylene (PP) collected from geographically distinct open ocean areas of the North Atlantic Subtropical Gyre. The plastisphere community consists of a morphologically diverse range of microbes that comprise a dense mix of eukaryotic and prokaryotic cells, such as diatoms, coccolitophores, dinoflagellates, fungi and bacteria (Reisser *et al* 2014; Zettler et al, 2013). The size, type and surface roughness of marine plastic debris can also influence the diversity and abundance of the microbial taxa that will colonise it (Carson *et al,* 2013).

Microbial assemblages associated with marine plastic are distinctly different from those of the surrounding seawater (Harrison *et al*, 2014; McCormick *et al*, 2014; Oberbeckmann *et al*, 2014). PET drinking water bottles attached to buoys in the North Sea, UK, showed clear differences in the composition of the plastisphere community compared to microbial communities of seawater and those attached to plankton and debris (Oberbeckmann *et al*, 2014). The study also illustrated temporal differences in microbial community composition colonising the plastic bottles, revealing a higher abundance of photosynthetic brown algae and cyanobacteria during the summer months compared to a dominance of heterotrophic bacteria and photosynthetic diatoms during the winter (Oberbeckmann *et al*, 2014).

In a study by Harrison *et al*. (2014) employing a laboratory-based microcosm setup containing sterile artificial seawater and inoculated with low-density polyethylene (LDPE) microplastics collected from marine sediments of the Humber Estuary, UK, over time the microplastics were observed to become colonised by morphologically distinct prokaryotic cells, predominantly bacteria. Molecular analysis revealed significant differences between the bacterial communities found attached to the LDPE microplastics and those within the sediment (Harrison *et al*, 2014). This finding corroborates that of McCormick *et al* (2014) who demonstrated significant differences in microbial communities found on microplastics in an urban Chicago River compared to those of the surrounding water column and suspended organic matter. Harrison *et al* (2014) also highlighted significant time-dependent variation in the structural community of the LDPE bacterial community. Initial observations showed the existence of sediment type-specific communities present on microplastics, with shifts towards "LDPE-specific" bacterial communities occurring at days 7 and 14 of the experiment, indicating a possible adaptation and change in community structure of these bacteria to microplastic waste (Harrison *et al*, 2014). The tendency of microplastics to attract a bacterial community that differs from that of the surrounding environment is further supported by a study conducted in a freshwater system, where bacterial communities on plastic litter from the Chicago River and Chicago's Lake Michigan beaches differed significantly from those colonising organic substances such as leaves and cardboard (Hoellein *et al*, 2014). The prevailing evidence appears to indicate that the plastisphere microbial communities are distinctly different from those found colonising other substrates or within the same environment but not associated with the plastic debris, indicating the possibility of specific adaptation to this novel man-made habitat. Plastics could therefore provide a new ecological niche or biotope, which, owing to its longevity in the environment, could help facilitate the persistence and transport of microorganisms across oceans and into new geographic areas (De Tender *et al*, 2015).

There is growing commercial interest in plastic biodegradation, with current research focussing on identifying the types of microorganisms capable of degrading plastics (Loredo-Treviño *et al*, 2012). Numerous studies have shown many different species of marine bacteria with the capacity to degrade hydrocarbons. Species of hydrocarbon- degrading bacteria, belonging to over 20 genera and distributed across some of the major bacterial Classes (*Alpha-*, *Beta-* and *Gammaproteobacteria*; *Actinomycetes*; *Flexibacter-Cytophaga- Bacteroides*), have been isolated and described (Floodgate, 1995; Head and Swannell, 1999; Head et al., 2006; Yakimov et al., 2007). These organisms are strongly enriched for during an oil spill at sea and they play an important role in the biodegradation of the oil (Gutierrez *et al*, 2014; Gertler *et al*, 2012). To our knowledge, the marine environment is the only place where we find bacteria with the ability to utilize hydrocarbons almost exclusively as a sole source of carbon and energy. Considering that plastic is composed of hydrocarbons, these types of bacteria could have important implications with respect to their role in degrading plastic debris. There are reports of changes in the surface topography of PET samples colonised by microorganisms, and microbial cells have been identified within pits and grooves, suggesting microbial degradation of the plastic surface (Reisser *et al* 2014; Webb *et al* 2009). However, the mechanics of biodegradation of marine plastic debris, and the underlying processes that influence this behaviour, are areas that need much further investigation to fully exploit the implications this can have on the environment.

3. Plastic dispersal: dissemination of pathogenic and harmful microbes

There are many examples on the introduction of invasive species into new habitats through colonisation of natural substances, such as wood, dead plants and pumice (Bryan *et al*, 2012; Minchinton, 2006; Van Duzer, 2004), and the ability of intertidal species to travel great distances offshore on floating rafts of seaweed (Ingólfsson, 2000). An increase in anthropogenic waste, in particular plastic litter, also provides another mechanism for facilitating the dispersal of non-native species in marine environments (Gregory, 2009; Jokiel, 1990). The buoyancy and durability of plastic makes it an ideal alternate substratum for a variety of colonisers, with plastics often shown to have higher diversity of species compared to other floating substrates, though this is likely to be dependent on the location and experimental sampling time (Bravo *et al*, 2011). The non-biodegradable nature of plastic increases its longevity in the marine environment, and which in turn significantly increases its potential for wide-scale dispersal of alien and invasive species (Barnes 2002a,b; Winston *et al*, 1997; Jokiel, 1990; Gregory, 1978). Increased survival and long-distance transport of native benthic invertebrates has been observed following their attachment to marine plastic debris (Barnes and Milner, 2005), with one study reporting the introduction of pathogens into a coral reef ecosystem through drifting plastic litter (Goldstein *et al*, 2014). Colonisation of a single piece of plastic by at least ten different species of marine animals has also been reported at remote locations such as the Southern Ocean, an area that has a relatively low input of anthropogenic litter (Barnes and Fraser, 2003). The size of the encrusting colonies indicated that this piece of plastic had been afloat for at least a year, indicating the potential for plastic-colonising organisms to survive and adapt at sea for many months, and potentially years (Barnes and Fraser, 2003). This provides important evidence that microbial “hitchhikers” on marine plastic debris could be widely disseminated, with the increasing amounts of global marine plastic providing opportunities for the transport of species into new habitats (De Tender *et al*, 2015).

Relatively little is known about the growth and dispersal dynamics of pathogenic and harmful microorganisms colonising the plastisphere, and the increased risk of human exposure from this poorly understood vector. Plastic-associated microbes from the Chicago River were dominated by taxa of potential pathogens and plastic decomposers, although these were less diverse than those of the surrounding water column and associated suspended organic matter (McCormick *et al*, 2014). LDPE-associated bacterial colonies were reported to be dominated by *Arcobacter* (Harrison *et al* (2014), and pathogenic species of *Vibrio* were found to dominate PP samples where they covered nearly 24% of the plastic surface (Zettler et al., 2013). Several *Vibrio* species, such as *V. cholerae* that causes cholera and *V. fluvialis* that can cause bloody diarrhoea and gastroenteritis, are known human pathogens, so their potential to colonise onto plastic marine litter represents an underexplored pathway for their dispersal. Essentially, plastic debris could represent a poorly-understood vehicle for the transport of these disease-causing organisms, particularly due to the ability of plastics to persist for significantly longer periods of time compared to other natural substances such as wood and feathers (Caruso, 2015; Zettler *et al*, 2013).

Drifting plastic debris can also be colonised by HAB species, such as *Ostreopsis* sp. and *Coolia* sp., in addition to resting cysts of unknown dinoflagellates, and temporary cysts and vegetative cells of *Alexandrium taylori* (Masó *et al*, 2003). Experiments using *A. taylori* cultured in plastic flasks showed the tendency of temporary cysts to attach to plastic surfaces (Masó *et al*, 2003), providing important insight toward our understanding on the global increase in HABs due to their dispersion via anthropogenic means. There is presently very little information on the role of plastic litter in the dispersion of HAB species, particularly in comparison to other natural debris (Carson *et al*, 2013), and therefore further studies will be needed to better understand this.

4. Implications for bathing water quality: human health and beach management

Faecal indicator organisms (FIOs), such as *E. coli* and intestinal enterococci, are widely used to monitor the quality of bathing waters and beach environments. These microorganisms mainly inhabit the mammalian gut, but can be delivered to the wider aquatic environment from numerous diffuse and point sources including sewage discharge, agricultural storm run-off, and sewer overflows (Oliver *et al*, 2015; Kay *et al*, 2008; Oliver *et al*, 2005). The rate of FIO delivery to receiving waters will therefore vary according to land-use and seasonal climatic conditions, e.g. patterns of localised storm events. The survival of FIOs in sand and water at beach environments is well documented (Halliday *et al*, 2015; Heaney *et al*, 2014), with Bonilla *et al* (2007) demonstrating significantly higher levels of bacteria in dry (2- to 23-fold) and wet (30- to 460-fold) sand compared to seawater. The harbouring of FIOs and human pathogens by certain species of freshwater macroalgae and beach-cast wrack (seaweed) have also been reported (Quilliam *et al*, 2014; Imamura *et al*, 2011; Ishii *et al*, 2006).

Beaches and bathing waters attract millions of tourists, swimmers, volunteers, and beach-goers each year and are a significant point of contact between humans and potential sources of pollution. Swimming is one of the most popular recreational activities at beaches (Wade *et al*, 2006), and epidemiological evidence shows a relationship between poor water quality and the occurrence of GI illnesses (Wade *et al*, 2010; Colford *et al*, 2007; Wade *et al*, 2006; Kay *et al*, 2004). Recreational water sports that are associated with varying degrees of potential water ingestion/contact, such as fishing, boating, wading and kayaking, are another emerging risk factor contributing towards possible GI illness (Dorevitch *et al*, 2011). However, beachgoers usually spend more time on the beach and strandline than in the water, with young children engaged in playing on the sand at the water's edge, whereas adults and elderly people are often found sunbathing (Heaney *et al*, 2012). Beach sands are known to harbour both FIOs and human pathogens in localised 'hotspots', often in concentrations much higher than those found in bathing waters (Sabino *et al*, 2014; Bonilla *et al*, 2007). Studies have demonstrated the occurrence of GI symptoms and diarrhoea in people exposed to sand via digging, building sandcastles and burying their bodies in sand at beaches with potential FIO contamination from nearby sewage treatment plants, with children found to be at higher susceptibility for contracting such illnesses (Heaney *et al*, 2012; Heaney *et al*, 2009).

With plastics now widely present in sediments and beach sands, and representing a potential unknown reservoir of pathogens and FIOs, a series of emerging research questions relating to plastics as a vector for wider public health risks need critical investigation. Furthermore, increasing amounts of floating plastic debris in bathing waters could also contribute to negative health impacts on bathers and recreational water users, owing to the yet unquantified potential of plastic litter to harbour and transmit diseases. The abundance of stranded and drifting plastic debris (both macro and micro particles) along beaches and coastal areas is expected to increase with projected increases in sea level, wind speed, wave height, and altered rainfall conditions (Browne *et al*, 2015; Young *et al*, 2011; Gulev and Grigorieva, 2004; Meier and Wahr, 2002). This is also likely lead to even greater human exposure to washed-up plastic debris. The majority of studies of marine plastic debris have mainly focused on its occurrence in coastal waters and open ocean areas such as gyres. Limited research, however, has been performed to investigate stranded beach plastics at designated bathing waters or other public beaches (Table 2). Of these limited studies, the majority had investigated abundance and distribution of plastic debris, with a variety of citizen science-based studies further complementing these assessments (Hoellein *et al*, 2015; Eastman *et al*, 2014; Smith *et al*, 2014; Hidalgo-Ruz and Thiel, 2013). Links between the colonisation of stranded plastic litter with human pathogens and FIOs, and the impact this could have on beachgoers and their health, have not yet been established, even despite the likelihood of public exposure to beach-cast plastic waste being much higher compared to litter in the open ocean. Strandlines are also marked by large quantities of beach-cast wrack and plastics, both of which could contain potential human pathogens (Quilliam *et al.*, 2014). Faecal loading from animals, such as gulls, waterfowl and dogs, significantly contributes towards elevated FIO abundance on beaches and in recreational waters (Wither *et al*, 2013; Edge and Hill, 2007; Wither *et al*, 2005; Lévesque *et al*, 2000). This could further facilitate the colonisation of beach-cast plastic litter with pathogens, and which could be prone to further dispersed elsewhere by wind, an incoming tide, or other means.

The ingestion of colonised plastic debris (particularly microplastics) by fish and marine birds that mistake it as food represents a potential pathway for disease-carrying plastic particles to enter the food chain and be dispersed to other environments. Recent evidence has demonstrated that deposit-feeders, such as mussels and shrimps, can ingest microplastics (Li *et al*, 2015; Cauwenberghe *et al*, 2015; Setälä *et al*, 2014; Browne *et al*, 2008), highlighting the potential for the transfer of microplastics from one trophic level to another. Therefore, as microplastics and stranded plastic debris are so prevalent on beaches, surface waters, marine sediments and in the water column, it is important that we develop a better understanding on the fate of plastics colonised by FIOs and pathogens, and their potential to become incorporated into the food chain and to persist in the gut of animals. Clearly, this could have far-reaching consequences for human health, commercial fisheries and the environment (Lattin *et al*, 2004; Thompson *et al*, 2004; Moore *et al*, 2001).

Microplastics from cosmetic care products and fibres in clothing are not effectively removed by wastewater treatment plants (WWTPs) and accumulate in the environment (McCormick *et al*, 2014), with 250% more microplastics found in coastal WWTP disposal sites compared to reference sites in the United Kingdom (Browne *et al*, 2011). Microplastics entering aquatic systems from WWTPs have been in close contact with human faeces, hence facilitating their potential to be colonised by FIOs and a range of human faecal pathogens (Oberbeckmann *et al*, 2015). The potential for sewage-exposed microplastics to harbour pathogens has only recently been explored, and there are reports of microplastics with high levels of colonisation by members of the *Campylobacteraceae* downstream of a WWTP (McCormick *et al*, 2014). This reinforces the need for further work to understand the mechanisms by which microorganisms, especially pathogens, in sewage hitchhike onto microplastic particles and find their way onto beaches and surrounding bathing environments. Targeted research in these areas could have significant societal impact, perhaps most notably by advancing beach management protocols and providing improved evidence to informing EU BWPs for increased public protection.

5. Conclusion

The negative impacts of marine plastic debris are widespread, but not yet fully understood. Marine and freshwater plastic debris is constantly being modified by the chemical and physical environment; therefore biofilm communities colonising plastics need to be dynamic with an ability to adapt to their changing environment. The potential for complex interactions between plastic waste and microorganisms of human health significance are currently poorly understood, yet the implications from this in disease transmission are significant. Promoting increased knowledge of both the role and importance of plastic surfaces in facilitating the survival and transfer of pathogens, particularly with respect to plastisphere-pathogen associations, currently represents an emerging research agenda in the wider field of health-related water microbiology. Quantifying the spatial and temporal shifts in human exposure pathways to human pathogens that might occur from macro to micro plastic debris, and the changing magnitude of risks to human health, will be challenging. Recognising human health impacts is clearly important, but the nature of risk associated with this novel transport mechanism capable of transferring associated microorganisms across large geographic ranges also introduces new regulatory challenges associated with the environmental and socio-economic protection of bathing waters and waters of significant recreational interest.

Understanding the ecology of the plastisphere community will further inform regulators and environment mangers of the risks from particular types and sizes of plastics, and the effects of environmental stressors such as temperature and exposure to higher UV radiation on the survival of plastic colonising pathogens and harmful microorganisms. Advances in plastisphere ecology will also contribute towards our knowledge of biodegradation of plastics and its adsorbed pollutants, and could provide useful information for future remediation strategies.

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Table 1: Studies investigating environmental plastic debris and biofilm formation

Area Sampled	Size of Plastic	Microbial Issue Investigated	Reference
Open ocean	Micro (0.25-0.5cm)	Colonisation of plastic particles by diatoms & hydroids	Carpenter and Smith, 1972
Coastal waters	Micro (0.1-2mm)	Bacterial colonisation of polystyrene particles	Carpenter *et al*, 1972
Open ocean	Macro & Micro	Characterization of microbial plastisphere community	Zettler *et al*, 2013
Coastal waters	Macro (30x30cm)	Bacterial colonisation of polyvinylchloride by *Rhodobacterales*	Dang *et al*, 2008
Open ocean	Macro & Micro (<5mm)	Abundance, diversity & variation of microbial community	Carson *et al*, 2013
Open ocean	Macro & possibly micro (<2cm)	Characterization of microorganisms colonising plastic debris; relationship between size of plastic & number of observed taxa	Goldstein *et al*, 2014
Open ocean	Macro (15x10cm)	Variation of biofilm community on HDPE, LDPE, & PP coupons with season and polymer type	Artham *et al*, 2009
Coastal waters	Macro	Potential of floating plastics to disperse toxic algal species	Masó *et al*, 2003
Coastal waters	Macro	Biofilm formation on polystyrene particles by bacteria & diatoms	Briand *et al*, 2012
Laboratory experiment using seawater	Macro (PET bottle pieces)	Biofilm formation & attachment of marine bacteria to PET surfaces	Webb *et al*, 2009
Coastal and ocean waters	Macro & micro	Characterization of microorganisms colonising plastic debris	Reisser *et al*, 2014
Open ocean	Macro (PET bottles)	Seasonal and spatial differences in biofilm diversity	Oberbeckmann *et al*, 2014
Seafloor	Macro (> 25mm) & micro (< 5mm)	Comparison of the plastisphere community to bacterial community of beach microplastics, sediment & surrounding seawater	De Tender *et al*, 2015

Coastal waters	Macro (PE plastic food bags)	Early stages of microbial biofilm formation on marine plastics	Lobelle and Cunliffe, 2011
Beach sediments	Micro (<5mm)	Bacterial colonisation of low-density polyethylene (LDPE) microplastics from three different sediment types	Harrison *et al*, 2014
Urban river	Micro	Assessment of microplastic abundance in urban river & composition of bacterial biofilms on plastics	McCormick *et al*, 2014

Table 2: Studies conducted on plastic debris from public bathing water beaches (excluding citizen science volunteer data studies).

Area sampled	Size of plastic	Issue investigated	Reference
Beach sediments	Pellets (0.1-0.5cm)	Potential of PP plastic pellets to transport toxic chemicals	Mato *et al*, 2001
Beach sediments	Macro & micro (1-15 mm)	Abundance of small plastic debris on Hawaiian beaches	McDermid and McMullen, 2004
Beach, estuarine and subtidal sediments	Micro	Abundance and extent of microplastic pollution	Thompson *et al*, 2004
Coastal beach sediments and seawater	Micro (>1.6µm)	Presence and abundance of microplastics	Ng and Obbard, 2006
Beach shorelines	Macro (> 1mm) & micro (< 1mm)	Influence of wind on spatial patterns of plastic debris	Browne *et al,* 2010
Beach	Virgin pellets, small (< 20mm) & micro (<20mm)	Size & distribution of plastic fragments on Brazilian beach	Costa *et al*, 2010
Beach shoreline sediments	Micro (<1 mm)	Spatial distribution of microplastics along six different continents	Browne *et al*, 2011
Beach sediments	Micro (<5 mm)	Bacterial colonization of low-density polyethylene (LDPE) microplastics from 3 different sediment types	Harrison *et al*, 2014
Beach shoreline and coastal waters (70-100m)	Macro	Distribution of anthropogenic litter in freshwater system & microbial interactions	Hoellein *et al*, 2014
Beach	Macro	Predicting short-term quantities of plastic debris washing ashore on beaches using a particle tracking model (PTM) & webcam monitoring	Kako *et al*, 2014
Beach	Macro	Colonisation of plastic litter by *E. coli* and *Vibrio* spp.	Quilliam *et al*, 2014

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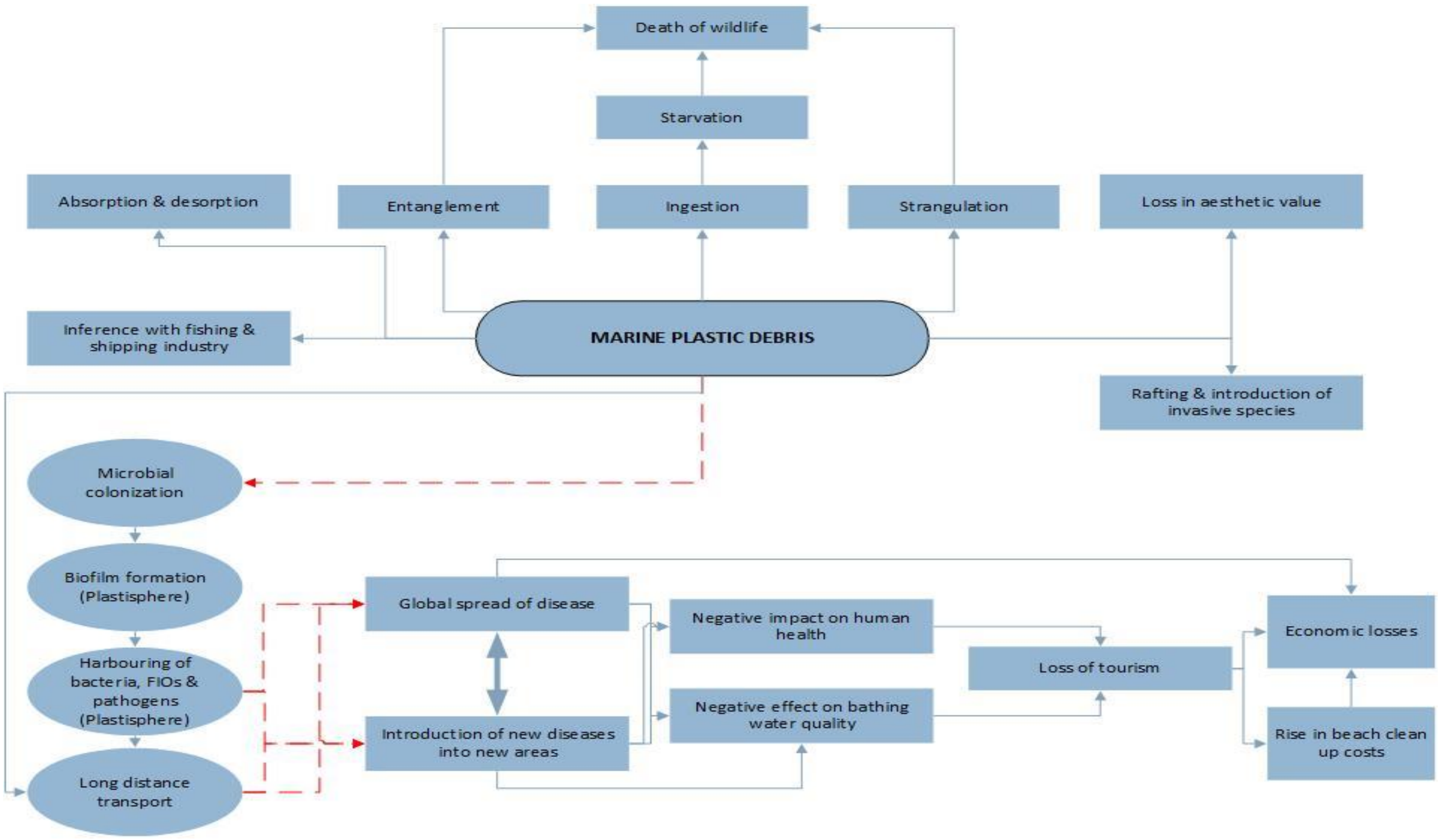

Figure 1: Impacts and interactions of marine plastic debris. Blue arrows indicate known effects; red dotted arrows indicate the yet unexplored effects/interactions as mediated by marine plastic debris.