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Understanding the Fragmentation Pattern of Marine Plastic Debris

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Supporting Information

ABSTRACT: The global estimation of microplastic afloat in the ocean is only approximately 1% of annual global plastic inputs. This reflects fundamental knowledge gaps in the transformation, fragmentation, and fates of microplastics in the ocean. In order to better understand microplastic fragmentation we proceeded to a thorough physicochemical characterization of samples collected from the North Atlantic subtropical gyre during the sea campaign Expedition seventh Continent in May 2014. The results were confronted with a mathematical approach. The introduction of mass distribution in opposition to the size distribution commonly proposed in this area clarify the fragmentation pattern. The mathematical analysis of the mass distribution points out a lack of debris with mass lighter than 1 mg. Characterization by means of microscopy, microtomography, and infrared microscopy gives a better understanding of the behavior of microplastic at sea. Flat pieces of debris (2 to 5 mm in length) typically have one face that is more photodegraded (due to exposure to the sun) and the other with more biofilm, suggesting that they float in a preferred orientation. Smaller debris, with a cubic shape (below 2 mm), seems to roll at sea. All faces are evenly photodegraded and they are less colonized. The breakpoint in the mathematical model and the experimental observation around 2 mm leads to the conclusion that there is a discontinuity in the rate of fragmentation: we hypothesized that the smaller microplastics, the cubic ones mostly, are fragmented much faster than the parallelepipeds.

INTRODUCTION

More than 300 million tons of plastic are used each year.1 On the basis of population density and the economic status of coastal countries, the mass of land based plastic waste entering the ocean was recently estimated at between 4.8 and 12.7 million metric tons per year.2 Most striking is the estimation for 2025: this amount will increase by an order of magnitude if waste management infrastructures are not improved.2 Plastic debris is abundant and widespread in the marine habitat; buoyant plastic particles have been reported in the water column worldwide,3–6 whereas nonfloating debris accumulates on the sea floor7–9 and in bed sediments from shorelines.10,11 Very recently, small plastic particles have even been found in deep sea habitats ranging in depth from 1100 to 5000 m.12

Marine plastic pollution has recently been recognized as a global environmental threat.13,14 The first reported potentially harmful effects are due to ingestion and entanglement.15,16 Plastic debris from several tens of centimeters to micrometers in size are available to a wide range of marine biota from sea turtles,17 seabirds,18,19 marine mammals,20 and fish.21 Furthermore, it was recently demonstrated that even smaller animals can be affected: copepods have the capacity to ingest microscopic polystyrene beads (1.7 to 30.6 μm), which consequently significantly decrease their algal feeding.22 Another problem associated with this pollution is that plastic debris is transported by ocean currents over very large distances and serves as a novel habitat for various organisms.23 Plastic debris transports algae, invertebrates, and fish to non-native regions.24 Recently, plastic-associated microbial communities were characterized by next-generation sequencing by Zettler et al.25 The impact of this attached community, which they named...
the "Plastisphere", on the oligotrophic open ocean has not been estimated so far. Plastic also transports potentially toxic compounds into the ocean.\textsuperscript{15} Some preliminary studies suggest that there is transfer of chemicals from ingested plastic debris to the tissues of sea birds.\textsuperscript{18,19} The transfer of chemicals from marine plastic debris to other organisms in the oligotrophic open ocean is yet to be evaluated. Because of the diversity of plastic debris sizes (from centimeter to micrometer scale) they are considered as being bioavailable to organisms throughout the food web; the transfer of chemicals should be considered at the base of the food chain where bioaccumulation can potentially begin to take place.\textsuperscript{27}

Large pieces of plastic debris (referred as macroplastics) break down into numerous small plastic fragments. Marine plastic debris collected in convergence zones is mainly in millimeter sized pieces,\textsuperscript{3,4,22,28,29} which are defined as microplastics (defined as plastic debris inferior to 5 mm). Although all plastic debris collected were about the same size (mostly between 0.5 mm and 5 mm), we know nothing about their sources or the pathways that they might have taken. They have followed very diverse journeys in terms of trajectory and time of residency in land, rivers, and finally the sea. All these factors mean that the composition and physicochemical characteristics of microplastics are very diverse.

As recently pointed out by Filella,\textsuperscript{30} a thorough characterization of the plastic debris is an important and necessary step if serious environmental questions are to be addressed. In this context, the present study gives a detailed physicochemical characterization of the plastic debris collected at the sea surface in the North Atlantic subtropical gyre. A detailed description of the plastic in terms of size, thickness and mass, together with a microscopic and infrared spectroscopy characterization is provided. A fragmentation pattern based on the physicochemical data gathered is proposed.

\section*{EXPERIMENTAL SECTION}

\textbf{Sample Collection.} Microplastic was collected from 6 net tows in the North Atlantic accumulation zone during daytime hours, from 18 to 22 May 2014, using the sailing vessel Guyavoile (details in Table S1 of the Supporting Information). One net tow was performed every day between 18 and 22 May. On 21 May, two tows were performed, the second being named tow bis. Net tows were conducted using Neuston nets with a standard mesh size of 300 μm. Plastics were collected in a 0.5 m × 0.3 m\textsuperscript{2} rectangular frame fitted with 2 m long, 300 μm mesh nets. The plastic debris was collected from the surface-layer in a depth of 0–30 cm. Tow durations were set to 60 min and were all undertaken while the vessel was traveling at a speed of 2 to 3 kn. The captain, who had 20 years’ sailing experience, estimated sea state of each sampling period. For all net tows, the Beaufort number was between 1 and 3, and the waves were always less than 1.5 m high.

\textbf{Sample Preservation.} After each tow, we transferred the collected contents into flasks and added Lugol’s iodine solution to preserve the plankton for identification and numbering; the final iodine concentration was 0.01 g/L. The volume of the solution was chosen so that there was no air entrapped in the flask, for better storage at room temperature. For the net tow of 21 May bis the plastic pieces were sorted with forceps on the boat and stored frozen in a glass vial.

\textbf{Microplastic Counting, Weighing, and Measuring.} In the laboratory, all samples preserved with Lugol solution were drained and put into seawater. Most of the plastic floated to the surface while biological tissues (mostly zooplankton) sank. Using a binocular microscope (using magnifying by 5 and 10), plastic debris was manually separated from natural matter with forceps. The remaining sample was inspected again on a glass plate. The plate was placed successively on top of white, black and red paper in order to sort out all the plastic debris. Sargassum was carefully inspected as plastic lines were often entangled in it. Once plastics were visually identified they had been separated from marine organisms and rinsed with distilled water and oven-dried at 65 °C for 2h. The contents of each net tow were treated separately. Plastic pieces were arranged in 20 cm diameter glass Petri dishes according to their size and color. Fishing lines were treated separately; they were manually measured with a ruler because they were often twisted. The Petri dish containing the pieces was scanned. The image was treated with the software Mesurim (Madre J.F. software). The pieces of plastic debris were individually identified and their length and width determined. Of the two dimensions established by Mesurim, we attributed the larger dimension to the length and the second to the width. All plastic debris were then weighed to the nearest 0.01 mg. Finally they were stored individually in glass vials at −18 °C for further characterization.

\textbf{Thickness Measurements.} Among the 478 pieces of plastic debris from the tow of 21 May, 422 were fragments and the 55 others were lines. The thickness of each of the 422 fragments was measured individually with an electronic micrometer (Electronic Outside Micrometer, 0.001 mm precision). The thicknesses were between 0.30 and 3.64 mm, with an average value of 1.08 mm and a standard error of 0.47 mm. There was over 1 order of magnitude of difference between the thinnest and the thickest fragments.

\textbf{Microscope Imaging.} Optical images of the plastic debris stored in the Lugol solution were recorded using an Olympus BX53 microscope with a PLN X10 lens; images were processed with Stream Basic software (Olympus Inc.).

\textbf{Data Analysis.} The mass distribution of the collected microplastics (except plastic lines) was studied through a statistical procedure based on Kolmogorov–Smirnov tests. Such tests quantify a distance between the distribution of two samples to discuss if they are drawn in the same way or not. The approach consisted in considering a theoretical agnostic fragmentation model. We compared the observed distribution of the masses of collected debris with the distribution obtained from this theoretical model. The goal is to identify the mass at which the tests began to reject the theoretical model in order to detect any change in the way pieces break down according to their masses. We applied our statistical analysis to 1093 microplastics, which had masses between 0.01 mg and 202.3 mg.

The procedure did not take time into account and focused only on how a debris broke down. Thus, it was sufficient to assume that a piece of plastic debris split into only two lighter parts at each step. The theoretical fragmentation process that we considered was based on uniform splits: for a given piece of plastic of mass \( m \), the two pieces obtained after one split had masses \( U^*m \) and \( (1-U^*)m \), where \( U \) is a uniform random variable between 0 and 1. This assumption means that the way a piece of debris breaks down does not depend on its geometry. To simulate such fragmentation from our data set, we considered the heaviest 10% of the debris and applied the theoretical process to their masses. The In that way, the masses of the 110 pieces of debris between 4.93 mg and 13.81 mg were
used to compute a simulated fragmentation that we compared with that observed for debris between some minimal mass \(m_{\text{min}}\) and 4.93 mg. The value of \(m_{\text{min}}\) varied from 0.01 mg to 3.43 mg to ensure that at least 10% of the remaining data was kept. For each value \(m_{\text{min}}\) a Kolmogorov–Smirnov test was done at the 5% level to compare the observed distribution of mass with the theoretical one. For that, the distance between the distributions is computed according to the value \(m_{\text{min}}\) and is referred to as the KS statistic in the sequel. The principle of the test is to accept the uniform fragmentation if the KS statistic is small enough with respect to some critical value discussed later.

**Infrared Spectroscopy.** Infrared spectra were recorded by attenuated total reflectance (ATR) using a diamond ATR crystal on a Thermo Nicolet Nexus apparatus. All images were taken with a nominal spectral resolution of 4 cm\(^{-1}\) and the experiment used 16 scans. The recorded data were corrected in order to obtain transmission-like spectra using the ATR Thermo correction (assuming the refractive index of the crystal on a ThermoNicolet Nexus apparatus. All images were obtained by infrared microscopy in transmission mode using OmnicSectra (ThermoScientific IN10. The nature of the plastic was determined using infrared analysis.

The smallest pieces of debris (<0.2 cm) were too small to be analyzed by ATR so they were placed on a glass plate and analyzed by infrared microscopy using a ThermoScientific OMNISectra (Thermoscientific software, database: Hummel Polymer library, HR Polymer and additives, HR Polymer, additives, and plasticizers). The oxidation profiles were obtained by infrared microscopy in transmission mode using a resolution of 8 cm\(^{-1}\) and 3S analysis (MCT detector was used) on semi thin sections. The sections (2 \(\mu m\)) were prepared on a Leica EM UC7 ultramicrotome and mounted on BaF2 or CaF2 discs (diameter 13 mm, thickness 2 mm).

**Scanning Electron Microscopy (SEM).** In order to remove the biofilm, plastic debris was washed for 48 h in 1 M sodium hydroxide solution. It was then abundantly rinsed with distilled water and dried overnight at room temperature. For comparison, polyethylene (PE) reference pellets were washed in 1 M sodium hydroxide solution. It was then abundantly rinsed with sodium hydroxide solution under the same conditions. Samples were mounted on microscope stubs and sputtered with platinum. Specimens were examined on an FEI Quanta 250 FEG scanning electron microscope at an accelerating voltage of 5–10 kV.

**X-ray Tomography.** Computed X-ray Tomography images were obtained using a GE/Phoenix Nanotom 180 at the French Research Federation FERMaT (FR3089) based in Toulouse, France. A cone shape X-ray beam with energy of 80 keV was transmitted and recorded 1440 images as the specimen underwent a 360° rotation (1.7 um/voxel). An image stack of the volume was constructed using Datos X (Phoenix X-ray system) and VG Studio Max (Volume Graphic GmbH, Germany).

## RESULTS AND DISCUSSION

### Length and Mass Distribution.

Plastic debris are generally categorized by size. It is explained by methodological reasons when they are sorted. But these size classes have induced reasoning errors. The fragmentation of an object is conservative only if the mass of the object is considered, not its size. For the rest of the discussion we have presented here both size and mass classification. The size classes established for plastic debris can vary widely from one study to another. In view of the multiplicity of reported methods, Hidalgo-Ruz suggested standardized procedures and classification in 2012.\(^{31}\) Two main size categories were proposed: debris smaller than 500 \(\mu m\) and microplastics (between 500 \(\mu m\) and 5 mm). Andrady suggested adding the term mesoplast for bigger pieces, between 5 and 200 mm.\(^{32}\) In this paper we will also distinguish pieces from lines as recently discussed by Reisser et al.\(^{38}\) As explained in detail by Filella\(^{39}\) the particle size determination is dependent on the measuring method, the weighing of a plastic debris is unequivocal. The weighing of all the microplastics individually was time-consuming. In order to have a point of comparison with previous studies we also discuss the size abundance distribution. The method used for the determination of the length is described in the Experimental Section.

The 1275 items of plastic debris had a total mass of 5.8 g. There were 175 lines, which represented 13% of the total debris collected in number and nearly 6% of the total mass. The line lengths were between 0.15 and 19.2 cm with an average size of 2.2 cm.

The lengths of the 1100 pieces ranged between 0.51 and 55 mm and their masses were between 0.01 and 1810 mg. The mass and length ranges of the collected pieces are given in Table S2. There was a total of 101 mesoplastics with a corresponding mass of 3.7 g (i.e., 63 wt %). One mesoplastic was markedly larger than the others; it was a very thick piece of black polyethylene 5.5 cm long. This piece weighed 1.81 g, corresponding to 31% of the total mass. Microplastics were largely superior in number with a total of 999 (78%), which represented 31% of the total mass. It is interesting to note that mesoplastics dominated in terms of mass (63%) whereas microplastics were the most abundant in number (78%). The relative contribution of microplastic versus the total plastic load is certainly system dependent (costal or deep sea area for example) but, unfortunately, these data are not clearly stated in published studies.

The size distribution of plastic pieces showed a peak in abundance around 1.5 and 2 mm (see Figure S1). This is in accordance with the distribution described by Cozar et al.,\(^3\) which was obtained from all five accumulation areas around the world. The gap observed below 1.5 mm has been discussed, and four main possible sinks have been proposed.\(^3\) Reisser et al. demonstrated that the vertical mixing process was size selective and affects the size distribution of the debris located at the sea surface.\(^{28}\) The proportion of plastics at depths greater than 0.5 m favored the smaller microplastics. The lack of smaller microplastics at the surface could thus be attributed partly to vertical mixing, an effect that is often left aside in discussions on fragmentation.\(^3\) However, vertical mixing increases with Beaufort numbers and for the present data set, all samples were collected at the sea surface when wave height was less than 1.5 m and therefore the vertical mixing could be neglected.
Figure 1 depicts the mass and length distribution in abundance of the plastic debris for the first time, contrarily to the size distribution. As expected with a fragmentation process, the mass distribution seems comparable to a power-law decay. The mass particle is a much relevant parameter than the size to discuss the fragmentation process. When a piece of plastic splits into two parts, the sum of their masses is still equal to the mass of the initial piece but this is not the case for the length. Such a conservative property is crucial for dealing with any fragmentation process. The mathematical model proposed here basically relies on this important remark to describe the plastic debris fragmentation. Figure 2 shows the results of the statistical procedure described in the Experimental Section. The uniform fragmentation model can be accepted for debris heavier than 1 mg but is definitely not acceptable for masses lighter than 0.8 mg. The main reason for rejecting the uniform fragmentation model below 1 mg is the total mass of lighter plastic debris. We observed a summed mass of 240.08 mg below 1 mg when the theoretical model predicted a total mass of around 4800 mg, i.e., approximately 20 times more than what was observed. Such a lack of mass means that there are other phenomena at work with debris lighter than 1 mg. Debris lighter than 1 mg correspond for 75% to debris smaller than 2 mm (Figure S1).

Microscopic Observations. Floating plastics readily develop extensive surface fouling. The steps have already been described: the debris is rapidly covered with a biofilm followed by an algal mat and then a colony of invertebrates. SEM images of microplastics show the rich microbial community developing on their surface. We will not discuss the kinetics of fouling and defouling of plastic debris in this study. The sinking of plastic debris due to biofouling and its “cyclic bobbing motion” was described by Andrady. In order to observe the plastic surface and remove the biofilm, the debris was washed with sodium hydroxide solution. Figure 3A shows the typical condition of a plastic fragment after being washed. Plastic debris presented multiple cracking running along the faces. Figure 3B shows the cross section of a plastic fragment; the cracks were generally a few tens of microns deep. For comparison with nonaged plastic, Figure 3C shows the transversal section of a new container made of PE. It presents...
no cracks. Tens of prefabrication pellets, plastic objects from everyday life, and packaging products in PE were analyzed. None showed cracking similar to that of the plastic debris collected from the North Atlantic gyre. It was checked that the washing step with the sodium hydroxide solution did not alter the surface of the plastic.

Forty-one plastic fragments were also studied by optical microscopy. They were mostly parallelepipeds but the smaller ones were cubic. We observed a general tendency: about 80% of the parallelepipeds (figure obtained from microscopic observations) did not present the same characteristics on both faces. One face showed much more cracking than the other; if the plastic debris were colored, the face with the cracks looked more faded. We also noticed that the face with less cracks hosted more organisms. In Figure S3 is given the abundance distribution of the cubic pieces collected in the manta net from the 21 of May (among 478 pieces). Most of the cubic pieces were smaller than 2 mm (75%). By microscopy, we observed that the cubic pieces presented the same characteristics on all faces. The cracks were less numerous, and there also seemed to be fewer organisms attached to the pieces (Figure 4).

**Microtomography.** The degradation of plastic debris is thought to occur mainly via photochemical processes, and the scientific community wonders to what extent the bacterial degradation contributes at sea. The 3D reconstruction of X-ray tomographic images of a microplastic is given in Figure 5. It shows sharpness at the surface. Cracks are also clearly identified closer to the surface; some are a few hundreds of microns deep (Figure 5a and b). The cracks seem to converge toward the center of the sample without reaching it. In the region of interest (ROI) selected and described in Figure 5 (right-hand side) the total porous volume is 3%, determined after calculation using manual separation of phases and the only threshold algorithm. No isolated cavity or porosity is observed in the plastic debris. If bacterial degradation has occurred, then the bacterial development does not seem to have penetrated into the plastic.

**Infrared Spectroscopy.** In the present study, a total of 58 pieces were analyzed by infrared spectroscopy; 92% were made of PE and the rest of polypropylene (PP). The predominance of polyolefins is in agreement with previous studies. Reisser et al. reported that 98.5% of the floating plastic debris collected in Australian coastal waters was made of PE and PP. Rios analyzed samples from coastal sites in the North Pacific Ocean and showed that the most abundant polymers were PP (80–90%) and PE (10%). The proportion of PE and PP is not reported for samples collected in gyres. It is interesting to compare this ratio with the production figures. Thermoplastic polyolefins account for most of the plastic produced worldwide. In 2012 the European plastic demand by resin type was 29.5%
for PE and 18.8% for PP (Plastic Europe figures), which corresponds to a PE/PP ratio of 1.5 and is much lower than the ratio PE/PP = 9 found here. A discussion of the mechanisms leading to such a difference should integrate multiple factors like the difference in the duration of the use of the objects between PE and PP, their probability of being discarded in the environment and their rate of photodegradation and fragmentation. All the lines collected and analyzed by infrared spectroscopy were shown to be made of polyethylene. These lines probably came mostly from fishing nets and lines. Nets and lines can be made of PE, nylon, or acrylic fiber (polyacrylonitrile), yet the only buoyant polymer of the three found in the tow was PE.

PE aging induces changes in its morphology and chemical structure. UV radiation induces free radical reactions in the polymeric chain; photo-oxidation is the major mechanism of PE degradation. The carbonyl groups are often used to characterize the advancement of PE weathering as the band appears broader with increased aging. All ketone, carboxylic acid, and ester functional groups formed upon UV irradiation contribute to the carbonyl signal in the 1650–1850 cm⁻¹ interval. The carbonyl index, also often referred to as the aging index, is often used to characterize the advancement of the oxidation of polymers. The carbonyl indexes of the plastic debris were between 0.13 and 0.74, indicating advanced stages of oxidation.

Usually, the oxidation products are confined to the first layer of the polymer. This is explained by the strong light absorption capacity of the sample, i.e., light does not penetrate deep inside it. Classically, photo-oxidation is limited to the top 100 μm. The depth of oxidation corresponds to the mobility of the radicals that are produced at the surface of the sample upon UV radiation. In order to study the profile of oxidation of the microplastics, we analyzed 2 μm thick slices. The carbonyl index was plotted as a function of the distance from one face of the debris (Figure S2). Five parallelepipeds were analyzed. For each piece, five profiles were recorded on distinct sections (2 or 3 sections for each slice). The carbonyl index was higher at the edges of the plastic and decreased rapidly toward the inside the sample. The carbonyl band was detected within the first 50 to 100 μm. Among the 11 profiles obtained, 10 showed a carbonyl index twice as high on one face of the sample as on the other. Only one profile presented the same oxidation state on both sides. The oxidation profile of the parallelepipeds demonstrates that one face was preferentially oxidized. We were not successful in obtaining sections from the cubes because they were too fragile.

**Fragmentation.** The physicochemical characterizations led us to propose two distinct behaviors for microplastics at sea. The bigger parallelepipeds float flat at the surface of the water (when the sea is calm), with one face preferentially exposed to the sun. The shear or tensile stresses together with a loss of mechanical properties due to photodegradation induce fragmentation (Scheme 1). The fragmentation probably occurs via breakdown along the cracks on their surfaces (Figure 3A) to give smaller parallelepipeds. The smaller cubic pieces are certainly formed when the length and width of the parallelepipeds approach the thickness of the piece. The cubic pieces tend to roll at the surface of the water. The motion of the cube shaped pieces seemed to prevent the development of a biofilm (by optical microscopy we observe less colonization of the cubical pieces). Erosion of the edges seems more likely. These differences in the behavior of buoyant plastic debris are
The authors declare no competing financial interest.

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