Highlights

- We exposed 3 species of corals to microplastics with and without natural prey
- We assessed active (ingestion) and passive (surface adhesion) removal of plastic
- Adhesion is 40 times more efficient than ingestion as removal process
- Mucus production is a defense mechanism that inhibits plastic adhesion
- Natural feeding is not impaired by microplastic exposure

EXPERIMENTAL DESIGN

LIVE CORAL

CONTROL
(coral skeletons)

BLANK
(empty chamber)

MAIN FINDING

$1.1 \pm 0.3 \%$

ACTIVE REMOVAL
(INGESTION)

$40.6 \pm 5 \%$

PASSIVE REMOVAL
(ADHESION)

$58.3 \pm 5.2 \%$

LEFT IN
SUSPENSION

Adhesion to coral surface as a potential sink for marine microplastics

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Abstract

Only 1% of plastic entering the ocean is found floating on its surface, with high loads in ocean accumulation zones and semi-enclosed seas, except for the Red Sea, which supports one of the lowest floating plastic loads worldwide. Given the extension of reefs in the Red Sea, we hypothesize a major role of scleractinian corals as sinks, through suspension-feeding, and assessed microplastic removal rates by three Red Sea coral species. Experimental evidence showed removal rates ranging from 0.25×10^{-3} to 14.8×10^{-3} microplastic particles $\text{polyp}^{-1} \text{ hour}^{-1}$, among species. However, this was only $2.2 \pm 0.6\%$ of the total removal rate, with passive removal through adhesion to the coral surface being 40 times higher than active removal through suspension-feeding. These results point at adhesion of plastic to coral reef structures as a major sink for microplastics suspended in the water column after sinking, helping explain low concentrations in Red Sea surface waters.

Keywords: Plastic, Coral Reef, Scleractinia, Ingestion, Red Sea

Capsule:

Corals remove microplastics from the water mainly through a passive mechanism of surface adhesion, which is a removal process 40 times more efficient than retention after ingestion.

1. Introduction

Mismanaged plastic waste reaching the marine environment eventually breaks into microplastics (<5 mm, Moore, 2008) and enters global oceanic transport, where it is expected to persist over hundreds of years (Barnes *et al.*, 2009). However, floating plastics in the ocean are only 1% of the expected loads (Van Sebille *et al.*, 2015; Law, 2017) and, although plastic fragmentation should follow an exponential curve of increasing abundance with decreasing size (Timár *et al.*, 2010), microplastics below 1 mm are missing in surface waters (Cozar *et al.*, 2014; Eriksen *et al.*, 2014). These findings suggest presence of sinks (i.e. coastlines, sea ice, sediments and biota, Law, 2017) and particularly size-dependent loss processes, like ingestion by organisms with a prey size <1 mm (Cózar *et al.*, 2014). Hence, there is growing interest in examining the retention of small microplastics in the smallest, yet most abundant, fraction of marine biota (Andrady, 2011; Cole *et al.*, 2013; Hämer *et al.*, 2014; Desforges, Galbraith and Ross, 2015; Van Cauwenberghe, Claessens, *et al.*, 2015; Watts *et al.*, 2015; Gigault *et al.*, 2016).

Red Sea surface waters sustain one of the lowest concentrations of plastic and, similar to global findings, fragments below 0.5 mm are reported to be missing (Martí *et al.*, 2017). In pelagic environments, vertebrates like mesopelagic fishes may play a major role in removing floating microplastics (Boerger *et al.*, 2010; Davison and Asch, 2011; Baalkhuyur *et al.*, 2018). In coastal and shallower waters, microplastics sunken and resuspended from the surface to the water body are more easily available to benthos, particularly invertebrates, which are likely to be significant contributors. The Red Sea is a coastal sea and hosts an extended web of coral reefs, about 2000 km long. Therefore, reef builders as scleractinian corals are abundant and through suspension-feeding screen the large volumes of water that flow daily on reefs (Reidenbach *et al.*, 2006), which posits them as significant removers of microplastics from the water. Indeed, plastic

47 ingestion in scleractinian corals has been demonstrated (Hall *et al.*, 2015; Allen, Seymour and
48 Rittschof, 2017; Reichert *et al.*, 2018). However, often, in ingestion experiments no alternative
49 food choice is given to corals, potentially biasing plastic ingestion rates, and ingestion alone is
50 held accountable for loss of particles in the water (Hall *et al.*, 2015; Allen, Seymour and
51 Rittschof, 2017), despite rates of plastic removal from the water often being inconsistent with
52 plastic recovery inside the polyps. For instance, Hall *et al.* (2015) estimated ingestion rates on
53 microplastics from particle loss in the water to be comparable to those on plankton, while plastic
54 recovered inside the polyps was less than plankton generally ingested per polyp by scleractinian
55 corals (Hall *et al.*, 2015).

56 Coral reefs are complex three-dimensional structures (Bradbury and Reichelt, 1983), buffering
57 water turbulence (Moberg and Folke, 1999) and, potentially, intersecting microplastics
58 transported with the flow passing over the reefs. Adhesion of microplastic to reef surfaces is
59 therefore a possible mechanism of plastic removal. For instance, high loss rates of plastic beads
60 in a tropical seagrass meadow were largely due to adhesion to leaves, covered with sticky
61 coatings produced by epifauna (Agawin and Duarte, 2002), and high plastic adhesion rates were
62 recently estimated on Red Sea giant clam shells (Arossa *et al.*, 2019), also important reef-
63 builders. Likewise, the contact with the fractal structure of scleractinian corals and possibly the
64 microbial biofilm covering the carbonate skeletons (Dunne, 2002) may lead to loss rates of
65 microplastics advected over coral reefs. Hence, microplastic removal by reef-forming
66 scleractinian corals may involve active (i.e. ingestion) and passive (i.e. adhesion) mechanisms.
67 However, only the former has been quantitatively assessed, often indirectly and/or in the absence
68 of prey, whereas adhesion to corals has been observed but never quantified (Allen, Seymour and
69 Rittschof, 2017; Reichert *et al.*, 2018).

Here we estimate active and passive microplastic removal by three species of scleractinian corals common in the Red Sea. The chosen corals have different skeleton macro- and micro-structures (e.g. growth forms and corallite dimension), which allow to study plastic adhesion on different surfaces. Moreover, we included an alternative diet, a natural prey, to verify whether plastic consumption rates change in conditions where food sources are multiple and we used an array of plastic sizes to determine if plastic ingestion and adhesion are size-dependent.

2. Materials and Methods

2.1 Specimens sampling and acclimation

We conducted microplastics exposure experiments with three coral species common in the Red Sea: *Acropora hemprichii*, *Goniastrea retiformis* and *Pocillopora verrucosa*. *Acropora hemprichii* and *P. verrucosa* are branching species with small-size polyps (approx. 0.5 mm), while *G. retiformis* is a massive coral with 2-3 mm large polyps. Branches of ~10 cm of *A. hemprichii* and *P. verrucosa* and chunks of *G. retiformis* were collected with hammer and chisel from different colonies in the Central Eastern Red Sea (Al Fahal reef, 22.25° N 28.96° E) in shallow waters (<10 m) and transported right after to aquaria facilities in tubs with fresh seawater equipped with air pumps. There, branches of *A. hemprichii* and *P. verrucosa* were fixed upright on tiles using epoxy and all specimens, including *G. retiformis*, were transferred into open flow system aquaria at ambient conditions (raw seawater, 28° C and L:D cycle of 12 hours, from 6 am to 6 pm). One week after the collection, water flow was switched to 0.2 µm filtered seawater to better match the condition used later during the experiment and corals acclimation was allowed for an additional week. During the two weeks acclimation, corals were fed every three days with hatched *Artemia salina* (48 hours old) at 6 pm.

2.2 Experimental set-up

After acclimation, we selected 12 specimens per species and transferred each of them in a feeding chamber, a 2 L-capacity glass beaker closed with a lid, filled with 1.5 L fresh 0.2 μm filtered seawater and equipped with an air pump, enough for microplastics to recirculate in the chamber. The water flow derived from the air pump would also mimic the water turbulence naturally found at a reef crest. Feeding chambers were allocated in water baths to maintain a constant temperature of 28° C, the *in situ* temperature at Al Fahal reef at the time of collection, during the experiment. Corals were let to acclimate at experimental conditions for 1 hour prior to particles exposure in order to allow re-extension of polyps. We prepared 6 additional chambers without organisms (“blanks”) to account for environmental particles losses and 18 chambers with dummies (referred as “controls”) to account for particles losses due to interaction with the rugose surface of the coral. Dummies were composed of coral skeletons, 9 branching corals and 9 massive corals with corallites of dimensions similar to those of species used in the experiment, coated with varnish to mimic the smoother surface of a living coral branch compared to the bare skeleton.

Specimens were assigned to three treatments: exposure to microplastics (from now on referred as “plastic” treatment), *Artemia salina* (referred as “*Artemia*” treatment) and both (referred as “mix” treatment). Each treatment included 12 chambers with live coral specimens (4 from each of the coral species), 2 blanks and 6 controls (3 including a branching dummy and 3 with a massive dummy), with a total of 20 chambers for each treatment (Supplementary Fig. 1). For the plastic treatment, we used polyethylene (~ 1 g/cc) fluorescent microbeads (Cospheric LLC). We chose polyethylene being the most abundant polymer type in Red Sea waters (Martí *et al.*, 2017). For each chamber, we prepared a mix of microbeads from 5 size classes (53-63 μm , 125-150

116 μm , 215-250 μm , 300-355 μm and 425-500 μm), 4094 ± 55 beads per size class, a total of 20,468
117 ± 209 beads (approx. 0.3 g) per chamber (13645 ± 139 beads L^{-1} , approx. 0.2 g L^{-1}). We selected
118 this concentration based on Hall et al., 2015 paper, where corals were exposed to 0.2 g L^{-1} of
119 microplastics ranging from 10 μm to 2 mm (Hall *et al.*, 2015), which is also similar to the
120 concentration used in other microplastic exposure experiments involving corals (Allen, Seymour
121 and Rittschof, 2017; Reichert *et al.*, 2018; Tang *et al.*, 2018; Rotjan *et al.*, 2019).
122 The microbeads mix was left in raw seawater exposed to sunlight for 1 week prior to the
123 experiment to allow biofouling, which may be involved in signaling for ingestion and in order to
124 mimic microplastic in the marine environment. For the *Artemia* treatment, we cultured *A. salina*
125 cysts 2 days prior to the experiment start. Few hours before the experiment start, we allowed the
126 hatched *A. salina nauplii* to settle and we separated them from unhatched eggs and cysts in a
127 second container. To infer density of *A. salina* in the culture batch, we homogenized the batch
128 one hour before the experiment, we withdrew 3 aliquots of 1 ml subsample and counted *nauplii*
129 and eggs, which were still present despite the cleaning step, in the aliquots under a stereoscope
130 using a Sedgewick rafter counting chamber. We then added to each feeding chamber a volume of
131 previously homogenized *A. salina* culture solution that contained an estimated number of 20,796
132 ± 102 *nauplii* and eggs (13864 ± 68 *A. salina* L^{-1}). For the mix treatment, we added 9787 ± 84
133 microbeads (1957 ± 20 per size class) and 9878 ± 48 *A. salina* eggs and *nauplii* to each chamber,
134 in order to attain similar concentrations of particles, hence potential food source, in all
135 treatments. Incubation in microplastics and/or *A. salina* started at 7 pm, after sunset, when coral
136 polyps are actively feeding, and was carried out for 24 h in *P. verrucosa* chambers and blanks,
137 which were part of a first run of the experiment, and 28 h in *G. retiformis*, *A. hemprichii* and
138 controls chambers, which were part of a second run.

2.3 Measurements

2.3.1 Microplastic and *Artemia salina* loss rates through time

During incubation, we collected 2 ml aliquots of water at 0, 2, 4, 6, 12, 24 and 28 h after the start of the experiment, 3 replicates from each chamber. From chambers including *P. verrucosa* and blanks we did not collect aliquots at 4 h and 28 h, sampling times that were added only in the second run of the experiment. Aliquots were filtered using a 25 µm nylon mesh and plastic beads on the filter were counted under a Discovery V.20 Carl Zeiss stereoscope using UV light, in order to easily visualize fluorescent beads, and characterized by size using AxioVision 4.8.2 software. *Artemia salina* eggs and nauplii were instead counted under halogen light.

2.3.2 Particles adhered to the coral surface and left in the water suspension

After incubation, corals and dummies were collected from the feeding chambers and rinsed thoroughly of particles (beads and *A. salina*) attached to the surface. When rinsing branching corals, we carefully avoided the supporting tile. Specimens were then frozen for further inspection. The water from each feeding chamber was also collected and the beaker washed thoroughly to ensure all particles were saved for further counting. Samples obtained after rinsing each *P. verrucosa* specimen and the corresponding beaker were not discriminated. However, after the first run of the experiment, we noticed that many plastic beads were adhering to the coral surface. Hence, for the second run we separated the coral and the beaker washing in two samples in order to account for total plastic adhesion to coral surface and distinguish it from plastic left in the water suspension.

Coral specimens exposed to plastic beads (from both the plastic and the mix treatment) were then inspected under a stereoscope where we observed that many beads were still strongly adhered to the coral surface, requiring removal by tweezers. Beads removed with tweezers will be now on

referred as “strongly-attached beads”, while “totally-adhered beads” include also beads from the coral washing. Specimens were inspected under UV light, used to easily visualize fluorescent beads, in order to ensure that the entire surface was cleared. A similar inspection was carried out for dummies. Considering the high abundance of particles adhering to the coral surface and left in the water suspension, samples from washes were first concentrated to a 200-ml solution each and only particles from a 5-ml subsample of the previously homogenized solution were filtered through a 25 μm nylon mesh and counted under the stereoscope.

2.3.3 Microplastic ingestion and retention in the polyp

During inspection at the stereoscope, we picked microplastic found under the tissue at the opening of the polyp mouth, hence ingested and successively retained (not egested). Finally, to account for plastic ingested and retained in the gut cavity, the same coral specimens were soaked in 5.25% sodium hypo-chlorite for 2 h, enough to allow complete digestion of the coral tissue. The solution was then filtered using a 25 μm nylon mesh and beads on filter were counted and measured at the stereoscope under UV light. Coral skeletons were also inspected under the stereoscope using UV light and all beads found in corallites were picked, counted and measured.

2.4 Calculations and statistical analyses

The surface area of each coral and dummy was calculated by 3D modelling, obtained from 360° videos recorded during acclimation and built using Autodesk ReCap 5.0. To estimate the average number of polyps cm^{-2} in each species, we counted polyp density in a known surface on three corals per species.

Particle loss rates through time were calculated from the slope of the fitted natural log estimated concentrations of particles in the water, obtained from aliquots sampling during the first 12 h,

when an exponential decrease of particles was observed. The role of the treatment, of the species and of the control on the particle loss rate was derived from Analysis of Covariance (ANCOVA). We composed a mass balance of the fate of plastic removed from the water in the presence of corals. We calculated, from direct retrieval of plastic beads inside the polyp or adhered to the coral surface, the passive (adhered) and active (ingested) removal rates by the coral. Differences in ingested or adhered beads at the end of the experiment among species and between mix and plastic treatments, and interactions of the two factors, were tested using two-way ANOVAs. The same test was used to compare adhered plastic between live specimens and dummies in the two treatments. If the two way-ANOVA showed only one factor to be significant, we pooled data from the non-significant factor and we proceeded using non-parametric statistical tests: Mann-Whitney tests were used to verify differences between two populations (e.g. beads abundance between plastic and mix treatments) or Kruskal-Wallis for more than two populations (e.g. differences among the three corals species). Kruskal-Wallis tests were followed by a pairwise t-test (p-value adjusted with Benjamini and Hochberg correction). Distribution of plastic beads in size classes, and particularly preference towards smaller or bigger sized beads, was also tested with a linear fit model of natural log transformed data. Correlations between parameters were verified with Rho-Spearman tests of correlation. All values are reported as mean $\pm$ SE. Actual p-values are indicated according to Amrhein et al., 2019 (Amrhein, Greenland and McShane, 2019). All statistical analyses were performed in R Studio 1.1.383.

3. Results

3.1 Microplastic and *Artemia salina* loss rates through time

Particles (microplastic beads and *Artemia salina* eggs and nauplii) declined exponentially in the presence of live corals over the first 12 h of the experiments, while reaching a plateau after 12 h (Supplementary Fig. 2). Loss rates are, therefore, reported over the first 12 h of exposure.

Microplastic and *A. salina* loss rates were independent and did not differ when using single particle sources or both particle sources combined (Table 1 and Supplementary Table 1.1 and 1.2).

Table 1 Linear models fitted on natural log transformed estimated concentrations of plastic and/or *Artemia salina* in the water, obtained from aliquots sampling of feeding chambers with live specimens treated with plastic and/or *A. salina* in the first 12 h.

Treatment	Particle type	Equation of linear model	Loss rate in 12 h	F-value	df	P-value
Plastic	Plastic beads	$\text{Ln}(y) = -0.71 \pm 0.1 \text{ Ln}(x) + 9.5$	83.8 %	51.93	1, 166	1.9e^{-11}
Mix	Plastic beads	$\text{Ln}(y) = -0.72 \pm 0.07 \text{ Ln}(x) + 8.7$	84.0 %	100.7	1, 166	2.2e^{-16}
<i>Artemia</i>	<i>Artemia salina</i>	$\text{Ln}(y) = -1.02 \pm 0.07 \text{ Ln}(x) + 9.8$	92.7 %	220.2	1, 166	2.2e^{-16}
Mix	<i>Artemia salina</i>	$\text{Ln}(y) = -0.9 \pm 0.08 \text{ Ln}(x) + 8.8$	89.9 %	138.2	1, 166	2.2e^{-16}

Microplastic loss rates were lower (about 20% after 1-hour exposure and 58% after 12 hours) for *A. hemprichii* compared to those for *G. retiformis* and *P. verrucosa* (about 50% and 45% after 1-hour exposure and 97% and 89% after 12 hours, respectively, Fig. 1a, Supplementary Table 1.3). Except for *A. hemprichii*, microplastic loss rates in the presence of living corals treated with plastic were remarkably similar to those in the presence of coral skeletons (dummies, Fig. 1a - c, Supplementary Table 1.4), which were nearly twice as high as those in

blanks (i.e. empty chambers, Fig. 1b, c). Of the three species, *G. retiformis* showed the highest *A. salina* loss rates (Fig. 1d, Supplementary Table 1.5). In contrast to microplastic particles, *A. salina* did not decline in empty chambers and showed only a weak tendency to decline in those including coral skeletons (Fig. 1d, e, Supplementary Table 1.6).

Fig. 1 Plastic and *Artemia* loss rates in chambers with live specimens, controls and blanks. Decrease of plastic bead concentration (in N beads L⁻¹) and of *Artemia salina* concentration (in N of *A. salina* L⁻¹) during 12 h in the (a – c) Plastic treatment and the (d – f) *Artemia* treatment for (a, d) live corals of the three species (*Acropora hemprichii*, *Pocillopora verrucosa* and *Goniastrea retiformis*), for (b, e) controls, including the blank, and (c, f) comparison of the slopes ($\pm$ SE) of the linear models. Lines represent the linear fit of natural log-transformed. Equations of linear fits are reported and p-values (*p*) are indicated. Linear models for the Mix treatment are reported in Supplementary Table 2.

3.2 Microplastic ingestion and retention in the polyp

Coral polyps ingested and retained microplastics (Fig. 2) at rates ranging from 4×10^{-5} to 6×10^{-2} microplastic particles polyp⁻¹ h⁻¹ (i.e. from about 1 to 144 microplastics every 100 polyps a day, Supplementary Table 3), independently of the presence of natural food (Supplementary Table 4, Fig. 3a). *Acropora hemprichii* and *P. verrucosa* preferably ingested and retained smaller microplastic particles, while no preference was observed for *G. retiformis* (Supplementary Fig. 3a), which also supported ingestion rates ca 50-fold higher than those of the other two species (Fig. 3a, Supplementary Table 3). The ingested microplastics were found both at the opening of the polyp mouth (Fig. 2) and mostly inside the gut cavity ($93.6\% \pm 1.7\%$ of totally ingested microplastic particles).

Fig. 2 Plastic ingestion in corals. Plastic beads ingested in polyps of *Acropora hemprichii* (a), *Goniastrea retiformis* (b) and *Pocillopora verrucosa* (c, detail in d) under halogen light (a, b, d) and UV light (c). The white arrow in panel b shows a bead that adhered to the surface of *G. retiformis* hence not ingested or not retained in the coral polyp. The color code is consistent among figures.

3.3 Microplastic adhesion to the coral surface

A large fraction of the microplastic particles added (mean $\pm$ SE after 28 h = $30.4\% \pm 2.9\%$, N = 27) were found attached to the skeletal surfaces of both living corals (*A. hemprichii* and *G. retiformis*, not quantified for *P. verrucosa*, Fig 3b, Supplementary Table 3) and dummies (skeletons only, Supplementary Table 5), at rates independent of the growth form and presence of biofilm on the skeleton, but dependent on the treatment (Supplementary Table 6). A fraction of the microplastic particles were strongly attached (trapped in the corallite septa or costae above the polyp tissue, Fig. 2b), requiring tweezers to be removed, particularly so for *G. retiformis* ($4 \pm 0.6\%$ of microplastics added and $10.5 \pm 1.9\%$ of totally adhered plastic, N = 8) compared to *P. verrucosa* ($0.08 \pm 0.04\%$ of added beads, N = 8) and *A. hemprichii* ($0.2 \pm 0.1\%$ of added beads, N = 8, and $1\% \pm 0.4\%$ of totally adhered plastic, N = 7), with more microplastic particles strongly attached in the plastic treatment (Supplementary Table 7, Fig. 3c). Moreover, the total microplastic particles adhered to *G. retiformis* surfaces tended to be larger than those adhered to *A. hemprichii* surfaces (Supplementary Fig. 3b, Supplementary Table 1.9), while no size-dependency was highlighted for strongly-attached beads in none of the three studied species (Supplementary Fig. 3c, Supplementary Table 1.10). The number of microplastic particles adhered to the skeletal surface of the coral increased with the number of microplastic particles ingested by the corals (Rho-Spearman test of correlation, $\rho = 0.68$ and 0.56 for total and strongly-adhered microplastic particles, respectively, Supplementary Fig. 4). However, ca 40 times more microplastic particles were found attached to coral structures at the end of the experiment than ingested, independently of the presence of *A. salina* (Table 2). *Artemia salina* eggs, but not *nauplii*, were also found adhered to the coral surface ($10\% \pm 1.2\%$ of the initial *A.*

275 *salina* concentration, $27.1\% \pm 2.8\%$ of the initial eggs concentration, $N = 27$), at rates similar for
 276 live corals and dummies.

277
 278 **Fig. 3 Plastic ingestion (a) and surface total- (b) and strong- (c) adhesion rates.** **a.** Ingestion rate
 279 (expressed as number of ingested beads polyp⁻¹ h⁻¹) in 24 specimens of three coral species (*Acropora*
 280 *hemprichii*, *Goniastrea retiformis* and *Pocillopora verrucosa*) from two treatments, mix (empty boxes)

and plastic (full boxes) treatments. **b.** Adhesion rate of totally adhered beads (expressed as number of beads $\text{cm}^{-2} \text{h}^{-1}$) in 15 specimens of 2 coral species (7 *A. hemprichii* and 8 *G. retiformis*) from two treatments, mix and plastic. **c.** Adhesion rate of strongly-attached beads (expressed as number of beads $\text{cm}^{-2} \text{h}^{-1}$) in 23 specimens of three coral species (7 *A. hemprichii*, 8 *G. retiformis* and 8 *P. verrucosa*) from two treatments, mix and plastic. Boxplots show the median (line across a box), quartiles (upper and lower bounds of each box) and extremes (upper and lower whiskers).

Table 2 Estimated proportion of beads that resided in the water, adhered to the coral surface and was ingested by *Acropora hemprichii* and *Goniastrea retiformis* (data not available for *Pocillopora verrucosa*) in plastic and mix treatment, relative to total plastic retrieved at the end of the experiment.

Species	N	Plastic in water (mean % $\pm$ SE)	Totally adhered plastic (mean % $\pm$ SE)	Ingested plastic (mean % $\pm$ SE)
Plastic treatment				
<i>A. hemprichii</i>	8	71.2 $\pm$ 11.4	28.7 $\pm$ 11.4	0.09 $\pm$ 0.03
<i>G. retiformis</i>	8	49.7 $\pm$ 4.6	48.2 $\pm$ 4.6	2.1 $\pm$ 0.5
Mix treatment				
<i>A. hemprichii</i>	7	74.3 $\pm$ 11.8	25.6 $\pm$ 11.8	0.1 $\pm$ 0.05
<i>G. retiformis</i>	8	56.1 $\pm$ 4.8	42.1 $\pm$ 4.4	1.8 $\pm$ 0.8
Total	31	58.3 $\pm$ 5.2	40.6 $\pm$ 5.0	1.1 $\pm$ 0.3

4. Discussion

The experimental results presented show occurrence of both passive, through adhesion, and active ingestion, with passive processes being far more important (40 times higher microplastic loss rates) than retention in the polyp after ingestion as a removal process. This indicates

that adhesion to the coral surface is the major process responsible for the loss of plastic from the water measured along the experiment. The finding that adhesion rates were similar for live corals and coral skeletons alone confirms adhesion to be a passive process, dependent on skeletal morphology, rather than the presence of a live animal. This does not apply to natural food: blanks and controls showed only a weak decrease of *A. salina*, demonstrating that its removal in feeding chambers hosting a live coral was due to ingestion, although adhesion of *A. salina* eggs was also observed. Indeed, unhatched eggs resemble plastic beads and their movement and interaction with the coral surface is likely subjected to the same mechanisms. However, thanks to the cleaning step in preparing the *A. salina* solution for feeding during the experiment, eggs were infrequent and ingestion still prevailed as removal process of *A. salina*. Since adhesion was the dominant plastic removal process, our experiments could not show that microplastic ingestion and retention by the coral species tested depends on the presence of the natural prey. However it showed that, as already demonstrated in cold water corals by Chapron *et al.*, 2018, microplastics does not impair natural feeding, since no difference in *A. salina* loss rates, happening mainly for ingestion, were highlighted between treatments.

Despite its high contribution as a removal process, adhesion has been unconsidered in experiments involving corals (Hall *et al.*, 2015), or, if detected, either adhered plastic was rinsed off from samples and discarded (Allen, Seymour and Rittschof, 2017) or only its occurrence was reported (Reichert *et al.*, 2018). Adhesion was observed also in copepods (Cole *et al.*, 2013), oysters gametes (González-Fernández *et al.*, 2018), sea urchin and mussels embryos (Della Torre *et al.*, 2014; Balbi *et al.*, 2017), or inferred as a translocation mechanism in mussels (Kolandhasamy *et al.*, 2018), but, except for Red Sea giant clams (Arossa *et al.*, 2019), it has never been quantified nor considered as a sink for microplastics in the water.

320 Microplastic removal rates differed, greatly, among coral species. Microplastic loss rates were
321 lower for *A. hemprichii* compared to the other two species. We indeed observed a high
322 production of mucus by *A. hemprichii* in the presence of microplastics, particularly during the
323 first two hours of exposure. Plastic beads were initially trapped by the mucus, which following
324 breaking due to the air flow, released them back in the water suspension. Mucus production is a
325 defense mechanism in corals, here possibly triggered by presence of extraneous particles, and
326 might have prevented adhesion of microplastic particles on *A. hemprichii* surfaces. Besides,
327 mucus has already been proposed as a trap and carrier for particles to sediments in reefs
328 ecosystems (Wild *et al.*, 2004) and may have the same role for microplastics, that conveyed by
329 mucus would more easily deposit on and get sequestered in sediments, already considered by
330 many one of the major microplastic sinks (Van Cauwenberghe, Devriese, *et al.*, 2015). Mucus
331 production was not observed in *G. retiformis* and *P. verrucosa*, where we indeed measured
332 higher plastic loss rates, indicating that adhesion was not inhibited. All corals exposed to plastic
333 beads were viable and fed actively for the entire duration of the experiment, including *A.*
334 *hemprichii* fragments regardless of the mucus production in the first hours of exposure. These
335 signs and the absence of further stress responses show that the exposed corals were healthy and
336 could have possibly used their natural cleaning mechanisms to clear their surface from
337 microplastics. However, cleaning mechanisms are energetically expensive (Riegl and Branch,
338 1995) and the difficulty we experienced in removing strongly-attached beads from the coral
339 surface suggests that these mechanisms are not enough to ensure microplastic detachment.
340 Specifically, more beads were strongly adhered to *G. retiformis* compared to the other two
341 species. Because there is no correlation between adhered microplastic particles and coral surface,
342 this difference cannot be ascribed to the larger dimension of *G. retiformis* chunks compared to *A.*

hemprichii and *P. verrucosa* fragments, and must be related, instead, to the surface properties of the three species. *Goniastrea retiformis* presents a highly rugose surface and larger polyps and septa, compared to *A. hemprichii* and *P. verrucosa* (Fig. 2), which presumably act as a comb efficiently and permanently trapping microplastic particles. Conversely, initial adhesion of plastic to the coral surface, similarly to what already inferred for copepod molts (Cole *et al.*, 2013), is most probably due to weak forces (e.g. van der Waals and electrical double layer forces, Alimi *et al.*, 2018), reason why most of the plastic could be rinsed easily from the specimens. Attachment efficiency in this case would be determined by multiple conditions (e.g. water chemistry and turbulence, plastic item size and surface roughness, Alimi *et al.*, 2018), which could justify why *A. hemprichii* smoother surfaces tended to attract smaller-size microplastic particles compared to *G. retiformis*. The adhesion efficiency is possibly also determined by the concentration of the plastic in the water as suggested by the lower beads adhesion on corals treated with both *A. salina* and plastic, exposed to a plastic concentration of 0.1 g L⁻¹, compared to corals treated with exclusively plastic (exposed to 0.2 g L⁻¹). Previous findings of giant clams exposed to different plastic concentrations also support this hypothesis (Arossa *et al.*, 2019). The concentration selected for our experiment might be considered high compared to environmental concentrations of plastic recorded in the surface waters of the Red Sea, potentially biasing adhesion rates that are dependent on plastic concentrations. In emphasizing that the aim of this work is mainly to compare active and passive removal processes and not to give absolute values of plastic adhesion rates on corals, we also highlight that the much lower plastic load in natural waters is not a valid parameter to select environmentally relevant concentrations for experiments. Indeed, the flow of water over coral reefs can be quite high (velocities reported for a Red Sea reef 5 to 20 cm s⁻¹, Reidenbach *et al.*, 2006) and exposure is the product of the concentration and

the water flowing over the corals during a certain length of time. Provided a conservative velocity of 10 cm s^{-1} , the water flowing on a day (exposure time in our experiment) over a 1 m section of 1-m deep Red Sea coral reef would be $8,640 \text{ m}^3$, 5.7 million times the volume of our experimental chambers. The loads of plastic we exposed the corals to in our experiment are indeed comparable to the loads present in $8,640 \text{ m}^3$ of water in the Red Sea, particularly in hotspot areas where plastic concentrations reach $3.84 \text{ items m}^{-3}$ (Martin, Agusti and Duarte, 2019). Moreover, the concentration in the water body is supposedly higher than the one in surface waters, that carry only 1% of the plastic discarded in the marine environment (Law, 2017). Hence, the likelihood of plastic particles entering in contact with the corals in our experiment, are of the order of that expected in a reef.

Goniastrea retiformis also supported the largest microplastic retention rates after ingestion, a behavior confirmed by this species showing the highest feeding rates on *A. salina* too. The size of microplastic particles ingested and retained in the three species most likely relates to the dimension of the polyp. Indeed, the two branching species, characterized by smaller polyps (Fig. 2), preferentially ingested and retained smaller-sized microplastic particles, while *G. retiformis* indistinctly ingested and retained all microplastic sizes available. The similar pattern in the size of adhered and retained microplastic particles together with the correlation between microplastic particles adhered and ingested suggest a possible link between adhesion and ingestion in corals.

In summary, the experimental evidence presented showed that corals remove microplastics from the water suspension both actively (ingestion) and, particularly, passively (adhesion), with the later processes supporting 40-fold greater removal rates than ingestion. In some coral species, adhesion can be prevented thanks to efficient defense mechanisms, like mucus production, but

after it has occurred, the chances to contrast it are reduced. Indeed, a large fraction of the microplastic particles adhered to coral surfaces was strongly attached, suggesting that they would remain attached even in turbulent environments (e.g. reef crests), becoming a component of the epilithic algal matrix recovery reef structures (Wilson *et al.*, 2003; Arossa *et al.*, 2019) or accumulating in the animal tissue, as already proposed for mussels (Kolandhasamy *et al.*, 2018). Phthalates, microplastic-associated contaminants, were recently detected in scleractinian corals collected from the field (Saliu *et al.*, 2019) and, while ingestion was proposed as the mechanism responsible of the phthalate transfer in the coral tissue, our findings suggest that transfer through adhesion is more likely to happen. In another study, microplastic was detected in wild corals (Rotjan *et al.*, 2019); however, since coral fragments were rinsed but not checked under the stereoscope before decalcification, it should not be excluded that the detected plastic particles, instead of or in addition to being ingested, were strongly attached to the surface. Because coral reefs form extensive and complex 3-D structures in coastal waters (McCormick, 1994), effectively increasing the surface area of coral per unit projected area of seabed (in coral reefs by 5 to 15 m² of coral surface per m² of projected reef area, Dahl, 1973), they are likely, as hypothesized here, to act as major sinks for microplastic particles. Indeed, the different adhesion rates showed in the three species possibly determined by their skeleton complexity, and the significantly lower removal rate in blanks where surfaces are smooth, suggest that the fractal structure of the coral surface plays an important role in retaining microplastics, that might adhere consistently less in environments with a simpler architectural complexity (e.g. sandy and rocky substrates or dead and eroded coral reefs, Bozec, Alvarez-Filip and Mumby, 2015). However, the role of coral reef structures as a possible sink for plastic particles through the adhesion mechanism demonstrated here remains hypothetical, as it cannot be simply extrapolated from the

short-term and small-scale experimental results presented here. Evidence for an important functional role of structural complexity of coral reefs (Graham and Nash, 2013) need to be expanded to include this as a driver for a role of coral reefs as filters and sinks for microplastics. Structural complex reefs are also characterized by supporting higher fish biomass (Graham and Nash, 2013). Hence, Red Sea reef fish may also be exposed to particle trapped by the reef, as supported by the finding of microplastics in the gastrointestinal tract of Red Sea coral reef fish (Baalkhuyur *et al.*, 2018), consistent with the important role of the epilithic algal matrix covering reef structures, where the bulk of microplastic particles are likely attached, as a food source for reef fish (Wilson *et al.*, 2003). Additionally, the prolonged adhesion of plastics to the coral surface poses risks to the same coral, like the demonstrated increase in disease susceptibility (Lamb *et al.*, 2018).

The extended coral reefs of the Red Sea, and possibly those of other coral regions, have, therefore, the potential to account for the very low plastic loads reported in these waters (Martí *et al.*, 2017), where microplastics become a component of the reef and maybe retained and/or be transferred to the reef food web. Further experimental assessments (e.g. on the coral community rather than on single specimens) and field evidences are needed to support the role of surface adhesion as an effective sink of microplastics in coral reefs.

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6. Author contributions

C.M.D. conceived the work; C.M., E.C. and G.M. designed and executed the experiment; C.M., E.C. and G.M. acquired and analyzed data; C.M., E.C., G.M. and C.M.D interpreted data; C.M. drafted the manuscript; all authors contributed to improving the manuscript and approved the submission.

Competing interests

The authors declare that there is no competing interest.

References

- Agawin, N. S. R. and Duarte, C. M. (2002) 'Evidence of direct particle trapping by a tropical seagrass meadow', *Estuaries*. Springer, 25(6), pp. 1205–1209.
- Alimi, O. S. *et al.* (2018) 'Microplastics and Nanoplastics in Aquatic Environments: Aggregation, Deposition, and Enhanced Contaminant Transport', *Environmental Science and Technology*. doi: 10.1021/acs.est.7b05559.
- Allen, A. S., Seymour, A. C. and Rittschof, D. (2017) 'Chemoreception drives plastic consumption in a hard coral', *Marine Pollution Bulletin*, 124(1), pp. 198–205. doi: 10.1016/j.marpolbul.2017.07.030.
- Amrhein, V., Greenland, S. and McShane, B. (2019) 'Scientists rise up against statistical significance', *Nature*. doi: 10.1038/d41586-019-00857-9.
- Andrady, A. L. (2011) 'Microplastics in the marine environment', *Marine Pollution Bulletin*, pp.

458 1596–1605. doi: 10.1016/j.marpolbul.2011.05.030.

459 Arossa, S. *et al.* (2019) ‘Microplastic removal by red sea giant clam (*Tridacna maxima*)’,

460 *Environmental Pollution*. Elsevier.

461 Baalkhuyur, F. M. *et al.* (2018) ‘Microplastic in the gastrointestinal tract of fishes along the

462 Saudi Arabian Red Sea coast’, *Marine Pollution Bulletin*, 131, pp. 407–415. doi:

463 10.1016/j.marpolbul.2018.04.040.

464 Balbi, T. *et al.* (2017) ‘Impact of cationic polystyrene nanoparticles (PS-NH₂) on early embryo

465 development of *Mytilus galloprovincialis*: Effects on shell formation’, *Chemosphere*. doi:

466 10.1016/j.chemosphere.2017.07.120.

467 Barnes, D. K. A. *et al.* (2009) ‘Accumulation and fragmentation of plastic debris in global

468 environments’, *Philosophical Transactions of the Royal Society B: Biological Sciences*,

469 364(1526), pp. 1985–1998. doi: 10.1098/rstb.2008.0205.

470 Boerger, C. M. *et al.* (2010) ‘Plastic ingestion by planktivorous fishes in the North Pacific

471 Central Gyre’, *Marine Pollution Bulletin*, 60(12), pp. 2275–2278. doi:

472 10.1016/j.marpolbul.2010.08.007.

473 Bozec, Y. M., Alvarez-Filip, L. and Mumby, P. J. (2015) ‘The dynamics of architectural

474 complexity on coral reefs under climate change’, *Global Change Biology*. doi:

475 10.1111/gcb.12698.

476 Bradbury, R. R. H. and Reichelt, R. R. E. (1983) ‘Fractal dimension of a coral reef at ecological

477 scales’, *Marine Ecology Progress Series*-pages: 10: 169-171.

478 Van Cauwenberghe, L., Claessens, M., *et al.* (2015) ‘Microplastics are taken up by mussels

479 (*Mytilus edulis*) and lugworms (*Arenicola marina*) living in natural habitats’, *Environmental*

480 *Pollution*, 199, pp. 10–17. doi: 10.1016/j.envpol.2015.01.008.

481 Van Cauwenberghe, L., Devriese, L., *et al.* (2015) 'Microplastics in sediments: A review of
 482 techniques, occurrence and effects', *Marine Environmental Research*, 111, pp. 5–17. doi:
 483 10.1016/j.marenvres.2015.06.007.

484 Chapron, L. *et al.* (2018) 'Macro- and microplastics affect cold-water corals growth, feeding and
 485 behaviour', *Scientific Reports*. doi: 10.1038/s41598-018-33683-6.

486 Cole, M. *et al.* (2013) 'Microplastic ingestion by zooplankton', *Environmental Science and*
 487 *Technology*, 47(12), pp. 6646–6655. doi: 10.1021/es400663f.

488 Cozar, A. *et al.* (2014) 'Plastic debris in the open ocean', *Proceedings of the National Academy*
 489 *of Sciences*, 111(28), pp. 10239–10244. doi: 10.1073/pnas.1314705111.

490 Cózar, A. *et al.* (2014) 'Plastic debris in the open ocean.', *Proceedings of the National Academy*
 491 *of Sciences of the United States of America*. doi: 10.1073/pnas.1314705111.

492 Dahl, A. L. (1973) 'Surface area in ecological analysis: Quantification of benthic coral-reef
 493 algae', *Marine Biology*. doi: 10.1007/BF00389331.

494 Davison, P. and Asch, R. G. (2011) 'Plastic ingestion by mesopelagic fishes in the North Pacific
 495 Subtropical Gyre', *Marine Ecology Progress Series*, 432, pp. 173–180. doi: 10.3354/meps09142.

496 Desforges, J. P. W., Galbraith, M. and Ross, P. S. (2015) 'Ingestion of Microplastics by
 497 Zooplankton in the Northeast Pacific Ocean', *Archives of environmental contamination and*
 498 *toxicology*, 69(3), pp. 320–330. doi: 10.1007/s00244-015-0172-5.

499 Dunne, W. M. (2002) 'Bacterial adhesion: seen any good biofilms lately?', *Clinical*
 500 *microbiology reviews*. Am Soc Microbiol, 15(2), pp. 155–166.

501 Eriksen, M. *et al.* (2014) 'Plastic Pollution in the World's Oceans: More than 5 Trillion Plastic
 502 Pieces Weighing over 250,000 Tons Afloat at Sea', *PLoS ONE*, 9(12). doi:
 503 10.1371/journal.pone.0111913.

504 Gigault, J. *et al.* (2016) ‘Marine plastic litter: The unanalyzed nano-fraction’, *Environmental*
505 *Science: Nano*, 3(2), pp. 346–350. doi: 10.1039/c6en00008h.

506 González-Fernández, C. *et al.* (2018) ‘Cellular responses of Pacific oyster (*Crassostrea gigas*)

507 gametes exposed in vitro to polystyrene nanoparticles’, *Chemosphere*. doi:
508 10.1016/j.chemosphere.2018.06.039.

509 Graham, N. A. J. and Nash, K. L. (2013) ‘The importance of structural complexity in coral reef

510 ecosystems’, *Coral Reefs*. Springer, 32(2), pp. 315–326.

511 Hall, N. M. *et al.* (2015) ‘Microplastic ingestion by scleractinian corals’, *Marine Biology*,
512 162(3), pp. 725–732. doi: 10.1007/s00227-015-2619-7.

513 Hämer, J. *et al.* (2014) ‘Fate of microplastics in the marine isopod *Idotea emarginata*’,
514 *Environmental Science and Technology*, 48(22), pp. 13451–13458. doi: 10.1021/es501385y.

515 Kolandhasamy, P. *et al.* (2018) ‘Adherence of microplastics to soft tissue of mussels: A novel

516 way to uptake microplastics beyond ingestion’, *Science of the Total Environment*. doi:
517 10.1016/j.scitotenv.2017.08.053.

518 Lamb, J. B. *et al.* (2018) ‘Plastic waste associated with disease on coral reefs’, *Science*. doi:
519 10.1126/science.aar3320.

520 Law, K. L. (2017) ‘Plastics in the Marine Environment’, *Annual Review of Marine Science*, 9(1),
521 pp. 205–229. doi: 10.1146/annurev-marine-010816-060409.

522 Martí, E. *et al.* (2017) ‘Low abundance of plastic fragments in the surface waters of the Red

523 Sea’, *Frontiers in Marine Science*, 4(NOV). doi: 10.3389/fmars.2017.00333.

524 Martin, C., Agusti, S. and Duarte, C. M. (2019) ‘Seasonality of marine plastic abundance in

525 central Red Sea pelagic waters’, *Science of The Total Environment*. Elsevier.

526 McCormick, M. I. (1994) ‘Comparison of field methods for measuring surface topography and

527 their associations with a tropical reef fish assemblage', *Marine ecology progress series*.
 528 *Oldendorf*, 112(1), pp. 87–96.
 529 Moberg, F. and Folke, C. (1999) 'Ecological goods and services of coral reef ecosystems',
 530 *Ecological economics*. Elsevier, 29(2), pp. 215–233.
 531 Moore, C. J. (2008) 'Synthetic polymers in the marine environment: A rapidly increasing, long-
 532 term threat', *Environmental Research*, 108(2), pp. 131–139. doi: 10.1016/j.envres.2008.07.025.
 533 Reichert, J. *et al.* (2018) 'Responses of reef building corals to microplastic exposure',
 534 *Environmental Pollution*, 237, pp. 955–960. doi: 10.1016/j.envpol.2017.11.006.
 535 Reidenbach, M. A. *et al.* (2006) 'Boundary layer turbulence and flow structure over a fringing
 536 coral reef', *Limnology and Oceanography*. doi: 10.4319/lo.2006.51.5.1956.
 537 Riegl, B. and Branch, G. M. (1995) 'Effects of sediment on the energy budgets of four
 538 scleractinian (Bourne 1900) and five alcyonacean (Lamouroux 1816) corals', *Journal of*
 539 *Experimental Marine Biology and Ecology*. doi: 10.1016/0022-0981(94)00164-9.
 540 Rotjan, R. D. *et al.* (2019) ' Patterns, dynamics and consequences of microplastic ingestion by
 541 the temperate coral, *Astrangia poculata* ', *Proceedings of the Royal Society B: Biological*
 542 *Sciences*. doi: 10.1098/rspb.2019.0726.
 543 Saliu, F. *et al.* (2019) 'Microplastics as a threat to coral reef environments: Detection of
 544 phthalate esters in neuston and scleractinian corals from the Faafu Atoll, Maldives', *Marine*
 545 *Pollution Bulletin*. doi: 10.1016/j.marpolbul.2019.03.043.
 546 Van Sebille, E. *et al.* (2015) 'A global inventory of small floating plastic debris', *Environmental*
 547 *Research Letters*, 10(12). doi: 10.1088/1748-9326/10/12/124006.
 548 Tang, J. *et al.* (2018) 'Acute microplastic exposure raises stress response and suppresses
 549 detoxification and immune capacities in the scleractinian coral *Pocillopora damicornis*',

550 *Environmental Pollution*. doi: 10.1016/j.envpol.2018.08.045.

551 Timár, G. *et al.* (2010) ‘New universality class for the fragmentation of plastic materials’,

552 *Physical Review Letters*, 104(9). doi: 10.1103/PhysRevLett.104.095502.

553 Della Torre, C. *et al.* (2014) ‘Accumulation and embryotoxicity of polystyrene nanoparticles at

554 early stage of development of sea urchin embryos *Paracentrotus lividus*’, *Environmental Science*

555 *and Technology*. doi: 10.1021/es502569w.

556 Watts, A. J. R. *et al.* (2015) ‘Ingestion of Plastic Microfibers by the Crab *Carcinus maenas* and

557 Its Effect on Food Consumption and Energy Balance’, *Environmental Science and Technology*,

558 49(24), pp. 14597–14604. doi: 10.1021/acs.est.5b04026.

559 Wild, C. *et al.* (2004) ‘Coral mucus functions as an energy carrier and particle trap in the reef

560 ecosystem’, *Nature*, 428(6978), pp. 66–70. doi: 10.1038/nature02344.

561 Wilson, S. K. *et al.* (2003) ‘Detritus in the epilithic algal matrix and its use by coral reef fishes’,

562 *Oceanography and Marine Biology*. doi: doi:10.1201/9780203180570.ch6.

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Competing interests

The authors declare that there is no competing interest.

Supplementary material

for

“Adhesion to coral surface as a potential sink for marine microplastics”

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Supplementary Materials

Fig. S1 to S4

Table S1 to S7

Figures

Fig. S1 Scheme of the experimental design. Samples were subjected to 3 treatments: the “Plastic” treatment (addition of plastic beads), the “*Artemia*” treatment (addition of *Artemia salina* eggs and nauplii) and the “Mix” treatment (addition of both beads and *A. salina*). Each treatment comprises 20 beakers: 12 beakers host 12 corals of three species (1 coral for each beaker, 4 corals for each species), 6 beakers (called “controls”) contain 6 coral skeletons, named here dummies, of two growth forms (1 dummy per beaker, 3 dummies per growth form) and 2 beakers were left empty (“blanks”).

Fig. S2 Particles loss rates in 28 h. Decrease of plastic beads (a and b) and *Artemia salina* eggs and nauplii (c and d) concentration during 28 h in plastic (a), *Artemia* (c) and mix (b, d) treatments for feeding chambers with live specimens (data from all species are pooled). Lines represent the logarithmic fit of data.

Fig. S3 Distribution in size classes of ingested, totally- and strongly- adhered beads. **a.** Distribution of ingested beads in 5 size classes for 24 corals of three coral species (8 corals per species). **b.** Distribution of totally-adhered beads in 5 size classes for 15 corals of two coral species (7 *Acropora hemprichii* and 8 *Goniastrea retiformis*). **c.** Distribution of strongly-attached beads in 5 size classes for 23 corals of three coral species (7 *Acropora hemprichii*, 8 *Pocillopora verrucosa* and 8 *Goniastrea retiformis*). Size classes are here reported as the mean value of the class boundaries. Data from plastic and mix treatments are pooled considered that no significant difference between the two treatments arises (Supplementary Table 1.7, 1.9 and 1.10). Lines represent the linear fit of natural log-transformed data. P-values (p) of linear models are reported.

Fig. S4 Correlation between number of ingested beads and total number of adhered beads (a) or number of strongly-attached beads in each of 15 corals (a, 7 *A. hemprichii* and 8 *G. retiformis*) and 24 corals (b, 8 *A. hemprichii*, 8 *G. retiformis* and 8 *P. verrucosa*), respectively.

Tables

Table S1 Results of ANCOVA tests. For each ANCOVA test, we report the terms of comparison in the first column, followed by R², F-statistic, degrees of freedom (df), p-value of the whole model and p-value of each comparison. Numbers in the first column cluster groups of same model type applied on different species.

Comparison (ANCOVA test)	R ²	F- value	df	P-value
1. Plastic loss rates, first 12 h: Plastic treatment - Mix treatment	0.4	63.45	3, 332	< 2.2e ⁻¹⁶ 0.95
2. <i>Artemia salina</i> loss rates, first 12 h: Plastic treatment - Mix treatment	0.6	142.9	3, 332	< 2.2e ⁻¹⁶ 0.2
3. Plastic loss rates, plastic treatment, first 12 h: <i>A. hemprichii</i> – <i>G. retiformis</i> <i>A. hemprichii</i> – <i>P. verrucosa</i> <i>G. retiformis</i> <i>P. verrucosa</i>	0.6	55.2	5, 162	< 2.2e ⁻¹⁶ 0.0004 0.004 0.5
4. Plastic loss rates, plastic treatment, first 12 h: <i>A. hemprichii</i> - Blank <i>A. hemprichii</i> - Branching dummy Blank – Branching dummy Plastic loss rates, plastic treatment, first 12 h: <i>G. retiformis</i> - Blank <i>G. retiformis</i> - Massive dummy Blank – Massive dummy Plastic loss rates, plastic treatment, first 12 h: <i>P. verrucosa</i> - Blank <i>P. verrucosa</i> - Branching dummy Blank – Branching dummy	0.4 0.7 0.6	17.83 60.19 28.25	5, 123 5, 123 5, 111	< 2.8e ⁻¹³ 0.3 0.007 0.2 < 2.2e ⁻¹⁶ 0.03 1.0 0.04 < 2.2e ⁻¹⁶ 0.1 1.0 0.2
5. <i>A. salina</i> loss rates, <i>Artemia</i> treatment, first 12 h: <i>A. hemprichii</i> - <i>G. retiformis</i> <i>A. hemprichii</i> - <i>P. verrucosa</i> <i>G. retiformis</i> - <i>P. verrucosa</i>	0.7	95.91	5, 162	< 2.2e ⁻¹⁶ 0.002 0.4 0.0001
6. <i>A. salina</i> loss rates, <i>Artemia</i> treatment, first 12 h: <i>A. hemprichii</i> - Blank <i>A. hemprichii</i> - Branching dummy Blank – Branching dummy	0.7	45.96	5, 123	< 2.2e ⁻¹⁶ 3.9e ⁻⁷ 1.1e ⁻⁹ 0.7

	<i>A. salina</i> loss rates, <i>Artemia</i> treatment, first 12 h:	0.9	176.9	5, 123	$< 2.2e^{-16}$
	<i>G. retiformis</i> - Blank				$< 2.2e^{-16}$
	<i>G. retiformis</i> - Massive dummy				$< 2.2e^{-16}$
	Blank – Massive dummy				0.6
	<i>A. salina</i> loss rates, <i>Artemia</i> treatment, first 12 h:	0.6	26.73	5, 111	$< 2.2e^{-5}$
	<i>P. verrucosa</i> - Blank				$1.5e^{-5}$
	<i>P. verrucosa</i> - Branching dummy				$1.3e^{-7}$
	Blank – Branching dummy				0.7
7.	Ingested plastic, distribution in sizes, <i>A. hemprichii</i> :	0.6	18.68	3, 36	$1.8e^{-7}$
	Plastic treatment - Mix treatment				0.5
	Ingested plastic, distribution in sizes, <i>G. retiformis</i> :	0.3	5.122	3, 36	0.004
	Plastic treatment - Mix treatment				0.9
	Ingested plastic, distribution in sizes, <i>P. verrucosa</i> :	0.6	15.25	3, 36	$1.5e^{-6}$
	Plastic treatment - Mix treatment				0.2
8.	Totally adhered plastic, distribution in sizes, <i>A. hemprichii</i> :	0.3	3.613	3, 31	0.02
	Plastic treatment - Mix treatment				0.1
	Totally adhered plastic, distribution in sizes, <i>G. retiformis</i> :	0.3	4.352	3, 36	0.01
	Plastic treatment - Mix treatment				0.5
9.	Totally adhered plastic, distribution in sizes:	0.3	11.77	3, 71	$2.4e^{-6}$
	<i>A. hemprichii</i> - <i>G. retiformis</i>				0.04
10	Strongly-attached plastic, distribution in sizes:	0.6	18.81	11, 103	$< 2.2e^{-16}$
	Plastic treatment - Mix treatment				0.7
	<i>A. hemprichii</i> - <i>G. retiformis</i>				0.08
	<i>A. hemprichii</i> - <i>P. verrucosa</i>				0.8
	<i>G. retiformis</i> - <i>P. verrucosa</i>				0.1

Table S2 Linear models fitted on natural log transformed estimated concentrations of plastic and/or *Artemia salina* in the water, obtained from aliquots sampling of feeding chambers treated with both plastic and *A. salina* (“Mix” treatment) in the first 12 h.

Particle Type	Sample	Species/ Dummy	Equation of linear model	P-value
Plastic beads	Live specimen	<i>Acropora hemprichii</i>	$\text{Ln}(y) = -0.4 \pm 0.1 \text{Ln}(x) + 8.3$	$5.6e^{-4}$
Plastic beads	Live specimen	<i>Pocillopora verrucosa</i>	$\text{Ln}(y) = -1.0 \pm 0.1 \text{Ln}(x) + 9.6$	$1.4e^{-9}$
Plastic beads	Live specimen	<i>Goniastrea retiformis</i>	$\text{Ln}(y) = -0.7 \pm 0.1 \text{Ln}(x) + 8.3$	$1.5e^{-10}$
Plastic beads	Control	Branching dummy	$\text{Ln}(y) = -0.4 \pm 0.08 \text{Ln}(x) + 8.8$	$9.5e^{-6}$
Plastic beads	Control	Massive dummy	$\text{Ln}(y) = -0.5 \pm 0.1 \text{Ln}(x) + 8.1$	0.001
Plastic beads	Blank	--	$\text{Ln}(y) = -0.4 \pm 0.1 \text{Ln}(x) + 8.9$	0.036
<i>Artemia salina</i>	Live specimen	<i>Acropora hemprichii</i>	$\text{Ln}(y) = -0.9 \pm 0.1 \text{Ln}(x) + 8.8$	$3.1e^{-11}$
<i>Artemia salina</i>	Live specimen	<i>Pocillopora verrucosa</i>	$\text{Ln}(y) = -0.9 \pm 0.1 \text{Ln}(x) + 9.6$	$2.9e^{-8}$
<i>Artemia salina</i>	Live specimen	<i>Goniastrea retiformis</i>	$\text{Ln}(y) = -0.9 \pm 0.06 \text{Ln}(x) + 8.2$	$<2.2e^{-16}$
<i>Artemia salina</i>	Control	Branching dummy	$\text{Ln}(y) = -0.03 \pm 0.06 \text{Ln}(x) + 8.3$	0.645
<i>Artemia salina</i>	Control	Massive dummy	$\text{Ln}(y) = -0.26 \pm 0.09 \text{Ln}(x) + 8.6$	0.005
<i>Artemia salina</i>	Blank	--	$\text{Ln}(y) = -0.05 \pm 0.1 \text{Ln}(x) + 9.3$	0.627

Table S3 Plastic ingested and adhered to surface of *Acropora hemprichii*, *Goniastrea retiformis* and *Pocillopora verrucosa* (24 corals of 3 species) exposed to two treatments (plastic and mix) for 28 h (*A. hemprichii* and *G. retiformis*) or 24 h (*P. verrucosa*). Estimated number of polyps, surface of each sample, rates of ingested plastic beads per polyp per hour and of totally adhered beads per cm² per hour are also reported. Some data on adhered beads are not available (*NA*) because samples were combined with plastic recovered from the feeding chamber at the end of the experiment.

Species	Polyps cm ⁻² mean ±SE	Coral surface (cm ²)	N ingested beads specimen ⁻¹	N ingested beads polyp ⁻¹ h ⁻¹	N adhered beads specimen ⁻¹	N adhered beads cm ⁻² h ⁻¹
Plastic treatment						
<i>A. hemprichii</i>	17.7 ± 4.7	35	11	6.4 x10 ⁻⁴	844	0.9
		58	12	4.3 x10 ⁻⁴	5643	3.5
		31	3	1.8 x10 ⁻⁴	1683	1.9
		112	11	2.1 x10 ⁻⁴	4515	1.4
Mean ± SE		59 ± 18.6	9 ± 2	3.7 (± 1) x10⁻⁴	3171 ± 1138	1.9 ± 0.6
<i>G. retiformis</i>	7.8 ± 0.7	37	447	0.06	7853	7.6
		72	360	0.02	6951	3.4
		76	201	0.01	4888	2.3
		190	153	0.004	6684	1.3
Mean ± SE		93.8 ± 33.2	290 ± 68	0.02 ± 0.01	6594 ± 621	3.6 ± 1.4
<i>P. verrucosa</i>	66.3 ± 7.2	66	6	0.4 x10 ⁻⁴	<i>NA</i>	<i>NA</i>
		42	18	2.5 x10 ⁻⁴	<i>NA</i>	<i>NA</i>
		37	36	6.3 x10 ⁻⁴	<i>NA</i>	<i>NA</i>
		43	14	2 x10 ⁻⁴	<i>NA</i>	<i>NA</i>
Mean ± SE		47 ± 6.5	19 ± 6	2.8 (± 1.2) x10⁻⁴	--	--
Mix treatment						
<i>A. hemprichii</i>	17.7 ± 4.7	63	13	4.3 x10 ⁻⁴	3169	1.8
		48	20	8.8 x10 ⁻⁴	<i>NA</i>	<i>NA</i>
		31	5	3.2 x10 ⁻⁴	721	0.8
		72	2	0.7 x10 ⁻⁴	1203	0.6

Mean ± SE		53.5 ± 9	10 ± 4	4.2 (± 1) x10⁻⁴	1698 ± 749	1.1 ± 0.4
<i>G. retiformis</i>	7.8 ± 0.7	130	3	1.1 x10 ⁻⁴	5000	1.4
		82	54	0.003	4320	1.9
		88	238	0.012	3680	1.5
		154	318	0.009	5320	1.2
Mean ± SE		113.5 ± 17.2	153 ± 75	0.006 ± 0.003	4580 ± 365	1.5 ± 0.1
<i>P. verrucosa</i>	66.3 ± 7.2	46	21	2.9 x10 ⁻⁴	NA	NA
		45	5	0.8 x10 ⁻⁴	NA	NA
		64	38	3.8 x10 ⁻⁴	NA	NA
		43	7	0.8 x10 ⁻⁴	NA	NA
Mean ± SE		49.5 ± 4.9	18 ± 8	2.1 (± 0.7) x10⁻⁴	--	--

Table S4 Effect of treatment (plastic or mix treatment), species (*A. hemprichii*, *G. retiformis*, *P. verrucosa*) and interaction of the two factors on plastic ingestion tested with Two-Way ANOVA. Degree of freedom (df), Sum of Squares (Sum Sq), Mean Squares (Mean Sq), F-value and P-value are reported. Kruskal-Wallis test using “Species” as the only variable was also significant (K = 10.745, df = 2, p-value = 0.005) and pairwise comparisons confirmed *G. retiformis* as the species ingesting more plastic (*G. retiformis* vs *A. hemprichii*/*P. verrucosa*, p-value = 0.016).

	df	Sum Sq	Mean Sq	F-value	P-value
Treatment	1	0.0002019	0.0002019	2.199	0.15539
Species	2	0.0011452	0.0005726	6.236	0.00876
Treatment*Species	2	0.0004018	0.0002009	2.188	0.14110
Residuals	18	0.0016528	0.0000918		

Table S5 Plastic adhered to surface on 6 branching and 6 massive dummies exposed to two treatments (plastic and mix treatments) for 28 h. Dummies surface and, as a result, number of adhered beads per cm² h⁻¹ are also reported.

Dummy growth form	Dummy surface (cm ²)	N adhered beads dummy ⁻¹	N adhered beads cm ⁻² h ⁻¹
Plastic treatment			
Massive	160	6214	1.4
	89	6903	2.8
	104	9220	3.2
Mean ± SE	117.7 ± 21.6	7446 ± 909	2.4 ± 0.5
Branching	116	3854	1.2
	51	4855	3.4
	48	2652	2.0
Mean ± SE	71 ± 22.1	3787 ± 637	2.2 ± 0.6
Mix treatment			
Massive	76	2902	1.4
	200	4112	0.7
	83	2806	1.2
Mean ± SE	119.7 ± 40.2	3273 ± 420	1.1 ± 0.2
Branching	84	4264	1.8
	40	1663	1.5
	43	5848	4.9
Mean ± SE	55.7 ± 14.2	3925 ± 1220	2.7 ± 1

Table S6 Effect of treatment (plastic and mix treatment), presence of biofilm on the surface (live coral and dummy), growth form (branching and massive) and interaction of the three factors on number of total beads adhered (following natural log-transformation) tested using a three-way ANOVA. Degree of freedom (df), Sum of Squares (Sum Sq), Mean Squares (Mean Sq), F-value and P-value are reported. Mann-Whitney *U* test using “Treatment” as the only variable was also significant: $W = 48$, $p\text{-value} = 0.037$.

	df	Sum Sq	Mean Sq	F-value	P-value
Treatment	1	1.449	1.4487	5.071	0.0364
Biofilm	1	0.067	0.0675	0.236	0.6325
Growth Form	1	0.1	0.1004	0.352	0.5602
Treatment*Biofilm	1	0.156	0.1559	0.546	0.4690
Treatment*Growth Form	1	0.447	0.4471	1.565	0.2261
Biofilm*Growth Form	1	1.065	1.0647	3.727	0.0686
Treatment*Biofilm*Growth Form	1	0.274	0.274	0.959	0.3397
Residuals	19	5.428	0.2856		

Table S7 Effect of treatment (plastic and mix treatment) and species (*A. hemprichii*, *G. retiformis* and *P. verrucosa*) on number of strongly-attached beads tested with a two-Way ANOVA. Degree of freedom (df), Sum of Squares (Sum Sq), Mean Squares (Mean Sq), F-value and P-value are reported.

	df	Sum Sq	Mean Sq	F-value	P-value
Treatment	1	0.05144	0.05144	7.962	0.0118
Species	2	0.2773	0.13865	21.461	2.2×10^{-5}
Treatment*Species	2	0.05985	0.02993	4.632	0.0248
Residuals	17	0.10983	0.00646		