 <i>Corallium rubrum</i>: new insights on the "plastic benefits" paradox Laura Carugati¹, Lorenzo Bramanti², Bruna Giordano², Lucia Pittura³, Rita Cannas¹, Maria Cristina Follesa¹, Antonio Pusceddu¹ & Alessandro Cau¹⊠ ¹Dipartimento di Scienze della vita e dell'ambiente – Università degli Studi di Cagliari - Via Tommaso Fiorelli 1, 09126 Cagliari, Italy ²Sorbonne Universités, UPMC Univ Paris 06, CNRS, Laboratoire d'Ecogéochimie des
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39 Abstract

40 Seafloor macrolitter is ubiquitous in world's oceans; still, huge knowledge gaps exist 41 on its interactions with benthic biota. We report here the colonization of plastic substrates by 42 the Mediterranean red coral Corallium rubrum (L. 1758), occurring both in controlled 43 conditions and in the wild at ca. 85 meters depth in the Western Mediterranean Sea. Juveniles 44 settled on seafloor macro-litter, with either arborescent or encrusting morphology, ranged from 0.6 to 3.5 mm in basal diameter and 0.2 - 7.1 years of age, also including a fraction 45 (20%) of potentially sexually mature individuals. In controlled conditions, larvae settled and 46 47 survived on plastic substrates for >60 days. Our insights show that marine plastic debris can 48 provide favourable substrate for C. rubrum settlement either in controlled conditions or in the 49 wild, suggesting their possible use in restoration activities. However, we pinpoint here that 50 this potential benefit could result in adverse effects on population dynamics.

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52 Keywords: Corallium rubrum; Marine plastic debris; Litter-fauna interactions;

53 Mediterranean Sea; restoration;

54 Plastic is the main component of seafloor litter (Canals et al., 2021; Pham et al., 2014; Worm et al., 55 2017) and its presence has been documented across all geographic and bathymetric boundaries 56 (Bergmann and Klages, 2012; Cau et al., 2018a; Chiba et al., 2018). Its impact on marine ecosystems 57 has become a matter of great concern for scientists, conservationists and policy makers. One of the 58 reasons for such attention is linked to the wide spectrum of direct interactions occuring between 59 seafloor litter and marine biota (e.g., Romera-Castillo et al., 2018). Among these interactions, the 60 most emblematic examples refer to the accidental ingestion and retention /fragmentation of plastic by 61 organisms associated with the seabed (Cau et al., 2020, 2019; Courtene-Jones et al., 2017) and the 62 entanglement of Derelict Fishing Gears (DFGs) with structuring fauna, including corals (Angiolillo et 63 al., 2015; Angiolillo and Fortibuoni, 2020; Consoli et al., 2019).

64 Beside the abovementioned inteactions, as per any kind of 'new' surface that enters the 65 marine environment, plastic can be rapidly colonized by organisms that accumulate over time (Wright 66 et al., 2020a, 2020b). The organisms that colonize the hydrophobic surface of plastic can span from 67 microbes and fungi to macro-invertebrates such as bivalves, barnacles, gastropods, polychaetes, 68 bryozoans, hydrozoan colonies and anthozoan corals (e.g., Battaglia et al., 2019; Santín et al., 2020). 69 In the case of floating plastic, the colonizing fauna can be transported for very long distances 70 (Hoeksema et al., 2012) and thus, plastic surfaces can become potential vectors for their dispersion 71 (Barnes, 2002; Barnes and Milner, 2005). For instance, recent studies highlighted the possible role of 72 plastic surfaces in facilitating the dispersion of invasive alien species, which could often be more successful than endemic species in the competition for space and resources (Barnes, 2002; Barnes et 73 74 al., 2009; Li et al., 2016). While the role of floating plastic items has been repeatedly documented, the 75 role of seafloor litter as an artificial substrate for encrusting and sessile fauna remains still largely 76 unexplored (Galgani, 2015; Katsanevakis et al., 2007). This aspect could be of particular relevance as 77 seafloor litter can enhance the overall complexity of the habitat, particularly in those otherwise flat 78 and sandy bottoms, paradoxically enhancing local biodiversity (Katsanevakis et al., 2007; Song et al., 79 2021).

81	can take advantage of seafloor plastic debris for settlement (Santín et al., 2020).
82	C. rubrum is an octocoral, endemic to the Mediterranean sea and its neighbouring Atlantic
83	coasts, belonging to the Corallidae family. Its peculiar features can be summarized as follow:
84	i) the species suffers from extensive harvesting since ancient times (Garrabou et al.,
85	2017) and, being one of the most valuable but also vulnerable marine resources
86	(Cannas et al., 2019), its management is still largely debated (Carugati et al., 2020;
87	Follesa et al., 2013; Tsounis et al., 2013);
88	ii) from a management and ecological point of view, shallow and deep populations may
89	be distinguished into populations dwelling above and below ca. 50 meters depth
90	(Cannas et al., 2016; Cattaneo-Vietti et al., 2016; Costantini et al., 2013);
91	iii) <i>C. rubrum</i> has a life span can exceed one century (Benedetti et al., 2016; Lartaud et
92	al., 2017; Santangelo et al., 2007) and reaches sexual maturity earlier than other
93	octocorals (Santangelo et al., 2003; Gallmetzer et al., 2010; Torrents et al., 2005);
94	iv) red coral is particularly vulnerable to climate change-related disturbances, including
95	water warming and acidification (Bramanti et al., 2013; Cau et al., 2018b; Cerrano et
96	al., 2013; Torrents et al., 2008);
97	v) the use of non-plastic artificial substrates failed to provide good practices for stock
98	restoration, due to high juvenile mortality and low recruitment on those substrates
99	(Bramanti et al., 2012, 2007; Santangelo et al., 2012).
100	We report here the first evidence of plastic items colonization by Corallium rubrum (L. 1758)
101	either in the wild or in controlled conditions in the laboratory.
102	Samples collected in the wild were obtained during professional, licensed, diving activities for
103	red coral harvesting conducted along the Northern coast of Sardinia (Western Mediterranean Sea,
104	Santa Teresa di Gallura, Strait of Bonifacio) (Fig. 1; Tab. 1). Samples of seafloor litter colonized by
105	red coral colonies were collected in 2012 at depths comprised between 80 and 90 meters, during

Also long-lived and protected species with naturally low recruitment rate and high juveniles mortality

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106 official harvesting season (15 May – 15 October) by licensed red coral scuba divers. The collected

107 plastic items showed dfferent polymer composition and various shapes: a plastic bag fragment (PB,

polyethylene; Fig. 2A); a plastic tape fragment (BT, polyvinyl chloride; Fig. 2B) and two ropes (R,
polypropylene; Fig. 2C).

110 The synthetic nature of collected items was confirmed by attenuated total reflectance (ATR) 111 Fourier transform infrared spectroscopy (FTIR), using a Spectrum Two spectrometer (PerkinElmer) 112 equipped with the Universal ATR accessory and operating with Spectrum 10 Software. The IR spectra 113 were acquired with a resolution of 4 cm⁻¹ in the Middle Infrared region (wavenumber range = 4000-114 600 cm⁻¹) with 4 scans after a background scan. The identification of polymers was performed by 115 comparison with both commercial libraries of standard spectra (PerkinElmer®) and custom-made 116 libraries. Polymers matching with reference spectra for more than 70% were validated 117 (Supplementary Figure 1).

118 Additionally, we observed few cases of C. rubrum recruitment on plastic substrates 119 (polypropylene) during a settlement experiment carried out at the Observatoire Oceanologique in 120 Banyuls sur Mer (France). Approximately 660 larvae were maintained in closed circuit, oxygenated 121 and temperature-controlled aquaria, to test for substratum preferences (see Zelli et al., 2020 for 122 details). Under these conditions, 5% of the larvae (n=33) settled on plastic substrates (Fig. 3A) and 123 survived for at least 60 days. Production of sclerites was observed in all newly settled individuals 124 (Fig. 3B) and, for some of them, a second polyp was observed next to the primary one, suggesting a 125 good health status of the young settlers.

Maximum height (cm) and basal diameter (mm) of wild samples were measured and colonies
were divided in two morphological categories: branched (B) and non-branched (NB). Basal diameter
was used to estimate the age of the samples on the basis of the relationsip between age and basal
diameter (Priori et al 2013).

Overall, 20 juvenile colonies were observed on the different plastic items from the wild, all
alive at the moment of collection. Six out of the 20 colonies were classified as branched (Tab. 2).
The highest density was observed on the plastic bag (PB) fragment where 4 branched colonies
were found, along with 4 unbranched ones, over a surface of ca. 25 cm² (density 0.32 col cm⁻²; Fig. 4).
The basal diameter of colonies settled on PB ranged from 0.6 to 3.5 mm, with an average value of

135 $2.31 \pm 1.02 \text{ mm}$ (average \pm SD; Tab. 2), while the average maximum height was $1.41 \pm 0.98 \text{ cm}$, 136 ranging from 0.4 to 3.4 cm.

The two ropes hosted both branched and unbranched colonies with an encrusting growth pattern of the base (see Fig. 5A,B) and an average basal diameter smaller than that of colonies settled on PB. Colonies settled on the two ropes showed an average basal diameter of 1.87 ± 0.87 mm (min. 1 to 3.5 mm; Tab. 2) and 1.95 ± 0.60 mm (min. 1.2 to 2.6 mm; Tab. 2) for R1 and R2, respectively. The maximum height of juveniles settled on ropes ranged from 0.4 to 5.9 cm, with an average value of 1.32 ± 1.09 cm for R1 and 1.93 ± 2.66 cm for R2.

143 The estimated age of colonies retrieved from the wild varied between 0.2 and 7.1 years (Tab. 144 2), with an average of 2.9 ± 2.2 years and modal value of 3.6 years. Despite the uncertainty in age 145 estimation, our results indicate that most of the colonies from the wild may have an age of several 146 years. Our results, while confirming the very long persistence of small plastic debris once they reach 147 the sea bottom, pose the question on their mobility once deposited on the seafloor (e.g., BT and PB 148 samples). Seafloor litter can get partially buried due to sedimentation and remain blocked on the 149 substrate, which could explain how C. rubrum larvae could have settled and survive several years on 150 small plastic items.

151 According to the few studies available on the reproductive features of deep-dwelling 152 populations (Porcu et al., 2017; Priori et al., 2013), a fraction of colonies (*i.e.*, those with a basal 153 diameter >2.8 mm; 20% of the total) described in the present study are likely to be already sexually 154 mature. The relationship between basal diameter and age (Priori et al., 2013) suggests that age of 155 colonies is comprised between 0.2 and 7 years (Tab. 2), further corroborating this hypothesis since the 156 minimum age at the first reproduction of C. rubrum can vary between 6 and 10 years (i.e., 20% of the 157 colonies here described), according to recent age estimation based on the observation of annual 158 growth rings (Gallmetzer et al., 2010; Torrents et al., 2005). The fact that plastic surface can represent 159 a suitable substrate for fouling and epibionts is not a novelty and, very recently, also deep coral 160 settlement was documented to occur on floats from DFGs (Battaglia et al., 2019). Evidences are also 161 building up on the fact that seafloor litter, in certain circumstances, may occasionally become a 162 surface suitable to host benthic species of high conservation concern (Santín et al., 2020). Our results,

163 providing the first evidence of precious red coral settlement and growth on plastic debris, add a 164 species of high conservation concern to the list of those capable of colonizing plastic surfaces.

165 From an ecological perspective, beside the renown deleterious effects of plastic and 166 microplastic contamination of marine biota (Angiolillo and Fortibuoni, 2020; de Oliveira Soares et al., 167 2020), the paradox of the "plastic benefits" gained attention at the time when seafloor waste was 168 proved to favour the settlement of non-indigenous species (Mordecai et al., 2011), which can use litter 169 objects as a transport vehicle for their dispersal (Kiessling et al., 2015). This paradox, however holds 170 true also for indigenous benthic species which can use the available surfaces provided by seafloor 171 litter as stepping stones for their dispersal and for enhancing connectivity between populations 172 (Sammarco et al., 2012). This could even have effects at the community level, as larvae of several 173 taxa preferentially settle on non-natural substrates rather than natural ones (Li et al., 2016; Pinochet et 174 al., 2020).

175 Anyway, while some fast-growing organisms can be effectively facilitated in their dispersal when 176 settled on litter (Katsanevakis et al., 2007; Zettler et al., 2013), this could not be the case for long-177 lived benthic species, with lifespan overpassing 100 years, such as C. rubrum. Indeed, the small 178 colonies we found (max basal diameter of 3.5 mm), despite being likely sexually mature, host a small 179 number of polyps and this, coupled with the generally low number of fertile polyps of wild colonies 180 (Porcu et al., 2017; Santangelo et al., 2007; Torrents et al., 2005), suggest that those colonies could 181 provide a very small contribution to larvae production. Moreover, plastic substrates like those 182 described in the present study are relatively small and thus their stability on the bottom is unlike; thus, 183 the risk of being moved by near-bottom currents (Kane et al., 2020) and/or being buried into 184 sediments is considerable. Whatever the case, it is likely that it would occur in a time frame that will 185 not allow red coral colonies to reach a highly reproductive size classes, with hundreds of fertile 186 polyps.

187 So, while plastic debris in the wild can provide favourable settlement substrates for *C. rubrum* 188 in otherwise unfavourable environments (*e.g.*, a plastic item that lay over soft bottoms or highly silted 189 environments), at the same time it could prevent larvae from settling on neighbouring favourable 190 substrates *e.g.*, rocky surfaces or crustose algae (Zelli et al., 2020). In this latter case, since the 191 lifespan of the settled colonies could be reduced, it could potentially affect the local population 192 demographical dynamics, particularly in environments accumulating huge amounts of plastic items, 193 including, for instance submarine canyons (Cau et al., 2017; Dominguez-Carrió et al., 2020). 194 On the other hand, the results obtained from the laboratory experiments (Fig. 3) suggest that in 195 the case of C. rubrum, for which the use of artificial non-plastic substrates and 196 transplanting/restoration trials provided scarce or null results, the use of plastic surfaces as a 197 settlement substrate appears a suitable and promising tool for future restoration actions. 198 However, other external cues are known to mediate coral larvae settlement (Heyward and 199 Negri, 1999), including either abiotic variables, such as light, colour, or sound (Lillis et al., 2018; 200 Mason et al., 2011), or biotic ones, such the presence of microbial films, CaCO₃ skeletons or crustose 201 coralline algae (Golbuu and Richmond, 2007; Negri et al., 2001; Nugues and Szmant, 2006; Webster 202 et al., 2004; Zelli et al., 2020; Jorissen et al 2020). Thus, we foster new experiments on settlement 203 preferences and larval behaviour of red coral and other species of conservation concern, for which 204 restoration measures are needed.

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206 **Declaration of competing interest**

The authors declare that they have no known competing financial interests or personal relationshipsthat could have appeared to influence the work here reported.

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223 Figures captions

- Figure 1. Map of the sampling sites along the Northern coast of Sardinia (Western MediterraneanSea).
- Figure 2. Overview of the *in-situ* samples: a plastic bag fragment (2A); a plastic tape fragment (2B)
 and two ropes (2C).
- Figure 3. Newly settled (45 days old) individuals of *Corallium rubrum* on plastic substrate. At the
- base of the two polyps it is possible to see the sclerites embedded in the coenenchyme. Single polyp
- 230 (A). A second small polyp is originating at the base of the big one (B).
- Figure 4. Overview of red coral colonies settled on the plastic bag sample (PB).
- Figure 5. Overview and details of red coral colonies settled on the first (A) and second rope (B).
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237 238 **Table 1.** *In-situ* samples: polymeric composition of *in-situ* samples, sites of collection, geographical coordinates, depth of collection. PVC = Polyvinyl chloride; PE = Polyethylene; PP = Polypropylene.

	Samples	Material	Sampling Site	Latitude (N)	Longitude (E)	Depth (m)
	Black Tape (BT)	PVC	1	41° 17.273'N	9°1.379'E	85
	Plastic Bag (PB)	PE	1	41° 17.273'N	9°1.379'E	85
	Rope 1 (R1)	PP	2	41° 16.923'N	9°1.882'E	84
	Rope 2 (R2)	PP	2	41° 16.923'N	9°1.882'E	84
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Table 1. *In-situ* samples: measured colonies code, basal diameter (in millimetres) and maximum243height (centimetres), the branching pattern (branched (B) / not-branched (NB) colonies) and the244estimated age, according to the growth rates by Priori et al., 2013 ($y = 1.3257 * x^{0.4947}$). NA = not245available.

Colorian	Basal Diameter	Max. Height	Duou ahim a	Estimated Age	
Colonies	(mm)	(cm)	Branching		
Black Tape (BT1)	2.5	1.7	NB	3.6	
Plastic Bag (PB1)	3.2	0.7	В	5.9	
Plastic Bag (PB2)	3.5	3.4	В	7.1	
Plastic Bag (PB3)	3.4	1.6	В	6.7	
Plastic Bag (PB4)	2.5	2	В	3.6	
Plastic Bag (PB5)	1.6	1.5	NB	1.5	
Plastic Bag (PB6)	0.6	0.4	NB	0.2	
Plastic Bag (PB7)	1.7	0.5	NB	1.7	
Plastic Bag (PB8)	2	1.2	NB	2.3	
Rope 1 (R1.1)	3.5	3.2	NB	7.1	
Rope 1 (R1.2)	1.6	2.1	NB	1.5	
Rope 1 (R1.3)	NA	NA	-		
Rope 1 (R1.4)	2.1	0.6	NB	2.5	
Rope 1 (R1.5)	1.5	0.7	NB	1.3	
Rope 1 (R1.6)	1.5	0.8	NB	1.3	
Rope 1 (R1.7)	1	0.5	NB	0.6	
Rope 2 (R2.1)	2.6	5.9	В	3.9	
Rope 2 (R2.2)	1.2	0.8	NB	0.8	
Rope 2 (R2.3)	2.2	0.6	В	2.8	
Rope 2 (R2.4)	1.8	0.4	NB	1.9	

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