

## Article

# Diversity Loss in Coralligenous Structuring Species Impacted by Fishing Gear and Marine Litter

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**Abstract:** Coralligenous structuring species (CSS) form a group of marine megabenthic species with an engineering capacity. Since they are highly vulnerable to anthropogenic activities, they have been selected for the Marine Strategy Framework Directive (MSFD) monitoring programs. The pressure and impact of fishing gear and marine litter on these species were evaluated through the image analysis of 54 remotely operated vehicle (ROV) routes along the Campania coasts (Tyrrhenian Sea, Italy). CSS density was calculated as the number of colonies/100 m<sup>2</sup>. Anthropogenic pressure was estimated as the frequency of frames showing longline, nets, other gear, plastic objects, metal objects, and other litter; while the impact was expressed as the frequency showing necrosis/epibiosis, broken/upturned and covered/entangled colonies. Cnidaria dominate in the Napoli, Campanella and Capri areas, while Bryozoa dominate in Cilento N and Cilento S areas. Campanella and Capri appeared to be the least heterogeneous despite their higher CSS densities, which was possibly related to the dominance of a few species. These areas were the most affected by showing the highest numbers of fishing gear (longlines) and marine litter (metal objects) recorded, amongst which longlines are the most abundant. In addition, these fishing areas are either close to a large urban center or located along popular touristic routes. In all the areas, colonies with necrosis/epibiosis (CNE) impact are present with low-moderate values, while the category gears covering/entangling (GCE) impact prevails in the Campanella and Capri areas, and this is strictly connected to the high presence of fishing gear.

**Keywords:** mesophotic reefs; ROV; anthropogenic pressure; megabenthos; mechanical damage; epibiosis; necrosis

**Citation:** Ferrigno, F.; Appolloni, L.; Donnarumma, L.; Di Stefano, F.; Rendina, F.; Sandulli, R.; Russo, G.F. Diversity Loss in Coralligenous Structuring Species Impacted by Fishing Gear and Marine Litter. *Diversity* **2021**, *13*, 331. <https://doi.org/10.3390/d13070331>

Academic Editor: Bert W. Hoeksema

Received: 20 May 2021

Accepted: 14 July 2021

Published: 19 July 2021

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## 1. Introduction

Coralligenous bioconstructions are heterogeneous structures of biogenic origin typical for the benthic Mediterranean habitats, developing under dim light conditions [1]. These mesophotic reefs occur at depths ranging from 30–40 m and extending to the limit of the photic zone, which may be up to 200 m depth [2–5]. Coralligenous assemblages mostly grow on hard substrates, but they can also develop on soft bottoms starting from the aggregation of free-living coralline algae, named rhodoliths [6,7]. Crustose coralline algae (mostly of the genera *Mesophyllum*, *Pseudolithophyllum* and *Lithophyllum*) build up a multi-layered substrate on which sponges (e.g., *Chondrosia reniformis*, *Axinella* spp., *Ircinia* spp., *Sarcotragus* spp., *Petrosia ficiformis*, *Aplysina cavernicola*, *Spongia* spp., *Haliclona mediterranea*, *Oscarella lobularis* and *Agelas oroides*), cnidarians (e.g., *Leptopsammia pruvoti*, *Caryophyllia* spp., *Epizoanthus arenaceus*, *Eunicella cavolinii*, *E. singularis*, *Corallium rubrum* and *Paramuricea clavata*), and bryozoans (e.g., *Schizomavella* spp., *Pentapora fascialis*,

*Myriapora truncata* and *Turbicellepora avicularis*) grow in addition to other engineering taxa [1,8,9]. Indeed, they form complex three-dimensional structures, hosting more than 10% of the known Mediterranean marine benthic species and provide suitable habitats for several endemic, vulnerable and protected species [1,10–12]. In particular, some megabenthic species, quite easily recognizable by images, represent the coralligenous structuring species used for the Marine Strategy Framework Directive programs (MSFD, 2008/56/EC and its 2017 updated version) [13]. Moreover, coralligenous reefs are an important “hot spot” of species diversity in the Mediterranean Sea and supply a variety of valuable ecosystem services [14], boasting also their economic relevance [15,16] and offering important fisheries grounds and aesthetic seascapes for diving tourism [1,17,18]. To these latter ecosystem services, it is also important to add carbon stock and sequestration by carbonate structures producing the bioconstructions [19].

Mesophotic reefs are threatened by several anthropogenic impacts, such as fishing, pollution, introductions of alien species and climate change [20,21]. These impacts may lead to structural and functional changes, altering trophic webs and transferring organic and inorganic contaminants to the food chain, thus influencing ecosystem functioning and ultimately human health [22,23]. Fishing gear, such as trawling nets and longlines, has a high impact on benthic communities by entangling organisms, mechanically damaging the erect species, and increasing turbidity and sediment accumulation [24–30]. Specifically, the entangled colonies may be broken and upturned by gear reaching out to tissue necrosis and subsequent epibionts overgrown [31–33]. The recovery of coralligenous species from damage can negatively affect the regenerative process, the growth, the reproduction, the resistance to diseases, and the competitive ability of the colonies [34,35].

Marine litter may favor hitch-hiking organisms and the spreading of alien species [36], as well as producing toxic effects in marine animals [37]. In particular, plastic litter lost in the marine environment can fragmentize into smaller pieces, becoming microplastics (<5 mm diameter) [23] and they can then be ingested, causing problems both for feeding and digestive activities and favoring the spread of xenobiotics [38]. The litter ingestion by fishes also has potential consequences for human health [39–41]. Overall, the accumulation of lost fishing gear and litter is considered among the major causes of marine environment degradation, and for this reason, several studies are trying to characterize and assess their impacts on marine ecosystems [32,42–46].

In a classical ACI (after control impact) experimental design, the impacts due to anthropogenic activities have determined a decrease of  $\beta$ -diversity. This is explained by a decrease in the number of species adapted to natural edaphic factors and an increase of the most resilient ones [47]. Therefore, differences in species composition on a large scale can reveal impacts. Indeed, at a small scale (differences among replicates of a single station), the high heterogeneity of community species composition is associated with a less impacted environment [48]. Some European legislation for the protection of the marine environment and natural resources and sustainable use of marine waters, such as the MSFD, have measures and monitoring programs aimed at studying, protecting and restoring sensitive habitats in order to maintain the biodiversity in the Mediterranean Sea [49]. Particular attention has to be paid to some remarkable habitats that play an important role in marine ecosystems dynamics and biodiversity. Indeed, among coralligenous species, there are some regarded as vulnerable and considered for protection according to international agreements (Habitat Directive 92/43/CEE; SPA/BIO Protocol; Barcelona Convention; Berna Convention).

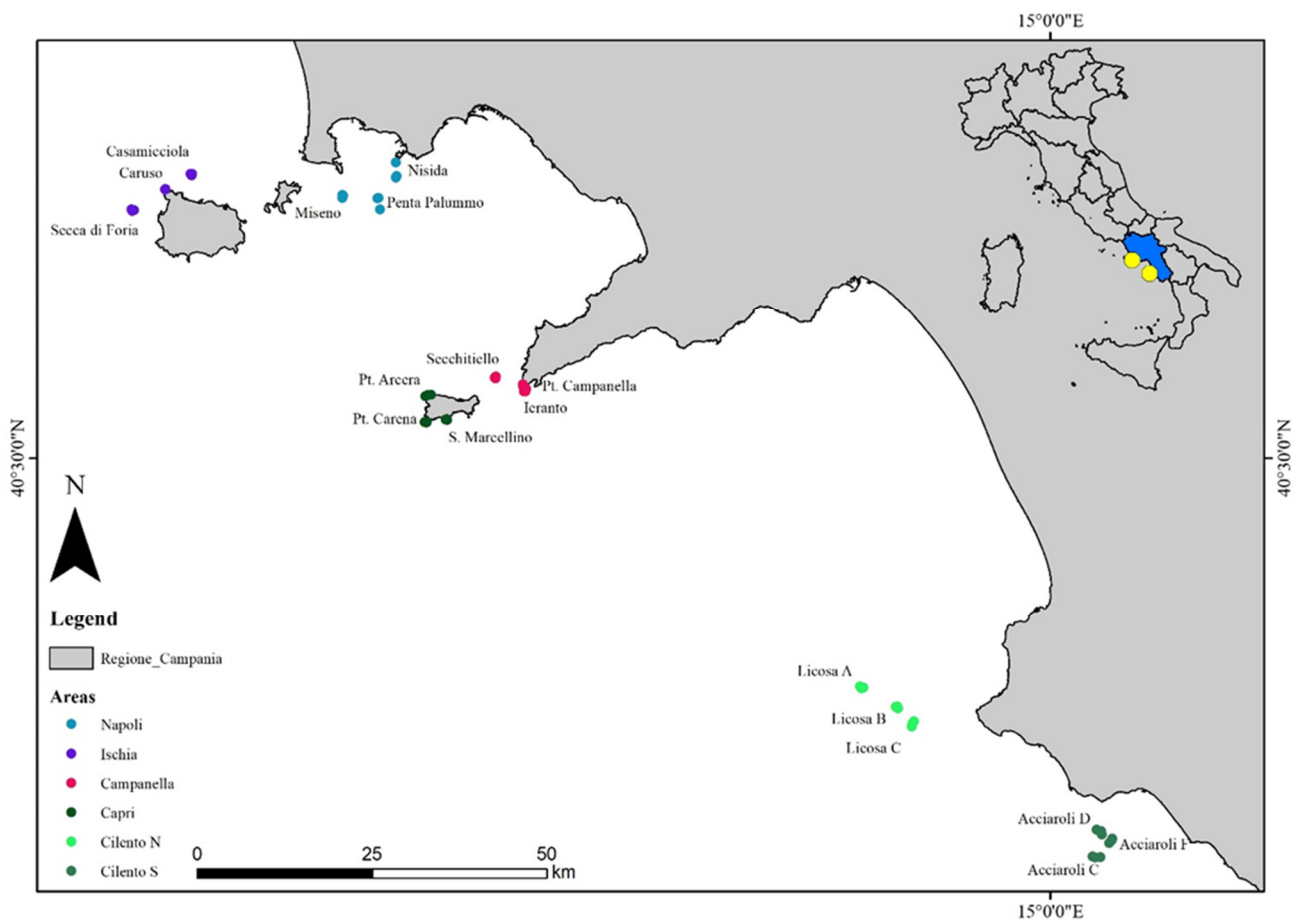
The aim of this study is to evaluate, by remotely operated vehicle (ROV) imaging technique, the effects of anthropogenic pressure type and abundance, represented by fishing gear and marine litter (D10—marine litter, MSFD), on coralligenous structuring species diversity (D6—seafloor integrity, MSFD), along the Campanian coasts (Tyrrhenian Sea, Italy). These coasts are characterized by high levels of urban population and maritime traffic for tourism and fishing activities, mostly represented by artisanal

fishing, in which trawl nets, gill nets, and longlines are used as fishing gear [50], even though there are still not enough available data on the characterization and impact of fishing gear and marine litter on the coralligenous sensitive habitats [4,12,32,45,51,52]. Such information may be a useful tool for the implementation of marine resources management plans in the Mediterranean Sea, aimed at the reefs' biodiversity and their associated species conservancy and the linked socioeconomic effects on fishing and tourism.

## 2. Materials and Methods

### 2.1. Study Area

The study areas (Figure 1) are located along different parts of the Campanian coasts (south-central Tyrrhenian Sea, Italy). A total of six areas, and three randomly replicated sites for each area, were investigated: “the Napoli area” (Nisida, Penta Palummo and Miseno sites), between 20 m and 73 m depth; “the Ischia area” (Secca Forio, Caruso and Casamicciola sites), between 26 m and 60 m depth; “the Campanella area” (Ieranto, Secchitiello and Punta Campanella sites) between 34 m and 102 m depth; “the Capri area” (S. Marcellino, P. Carena and P. Arcera sites), between 30 m and 102 m depth; “the Cilento N area” (Licosa A, Licosa B and Licosa C sites), between 40 m and 95 m depth, and “the Cilento S area” (Acciaroli C, Acciaroli D and Acciaroli F sites), between 49 m and 78 m depth.



**Figure 1.** Study areas with sampling sites.

The Napoli area consists of a sand/mud substrate with scattered coralligenous banks; Ischia island is mostly characterized by a hard bottom due to the presence of cliffs and rocky outcrops of volcanic origins; the sites located in the Campanella area are

represented by two rocky cliffs (P. Campanella and Ieranto) and a shoal (Secchitiello); the Capri area also consists of steep rocky cliffs; finally, the Cilento N and Cilento S areas are characterized by a succession of rocky outcrops and coralligenous banks [53–55].

## 2.2. Field Activities

The investigation was carried out during several surveys in 2017 and 2018, aimed at assessing the ecological status of coralligenous habitats, following the MSFD protocols [56]). The study was carried out through a remotely operated vehicle (the ROV Ageotec, mod. Perseus), equipped with an HD camera (DVS-3000 high definition), a navigation camera with an underwater positioning Ultra Short Base Line System (USBL) interfaced with the on-board navigation system, two spotlights and two laser pointers providing a 14.5-cm scale for the measurement of the frame area and the size of the organisms. ROV moved at approximately 0.3–0.4 knots and at a distance of about 0.5–1 m from the sea bottom, with an average visual width of 0.5 m.

Overall, 6 areas with 3 sites each (18 sites in total) were monitored; within each site three 300 m routes were performed (54 routes in total).

## 2.3. Data Management

The ROV videos were analyzed using the VisualSoft® software, rendering the HD videos overlapped with the navigation data. The frames used for the analysis, were obtained by video defragmentation each 10 sec with the DVDVideoSoft® software. Taking into account the length of each route (around 300 m), a number of 74 images per route was chosen for uniformity between the different routes. Only high-quality images were analyzed.

The density of coralligenous structuring species identified by ROV-imaging technique (CSS, *sensu* MSFD protocols) was calculated as the number of colonies of each species on 100 m<sup>2</sup> (n. CSS colonies/100 m<sup>2</sup>) and compared with CSS frequency (percentage times that CSS appeared in each route of the total images number with coralligenous habitat), in order to assess the spatial distribution of species [57]. Moreover, quantitative dominances (Di), as the ratio of a specific CSS density with total density, were also calculated for each area.

Anthropogenic pressure on coralligenous habitats was assessed by frequencies of coralligenous bioconstructions presenting fishing gear or marine litter, calculated as a percentage of frames. Likewise, the anthropogenic impacts were assessed as frequency, calculated as a percentage of frames with damaged coralligenous structuring species.

The main anthropogenic impacts on the coralligenous bioconstructions were represented by three categories of fishing gear: “longlines”, “nets” (such as trawl nets, gill nets or trammel nets) and “other gears” (pots, ropes, moorings, anchors, etc.), and three categories of marine litter: “plastic objects” (bottles, bags, containers, etc.), “metal objects” (such as cans and bins) and “other litter” (textile, glass, wood, tires, etc.).

Anthropogenic activities impacting structuring species colonies can be direct or indirect and they were classified into three categories [34]: the direct impacts are BUC (broken/upturned colonies) and GCE (gears covering/entangling), while the indirect impact is CNE (colonies with necrosis/epibiosis).

## 2.4. Statistical Analyses

The experimental design, with fishing gears and marine litter not causing damage to the coralligenous communities as the null hypothesis, used statistical analyses involving two factors: area (Ar: fixed, six levels) and site (Si: random, 18 levels) with  $n = 3$  (routes per site). Prior to the analyses, abundances were square-root transformed to reduce the weight of very scanty CSS.

Univariate and multivariate analyses were performed on total densities and single species densities, respectively. In particular, following the Terlizzi et al. (2007) methodology [58], univariate analyses of variance PERMANOVA [59] based on Euclidean distance,

was conducted on CSS densities, while the Bray–Curtis similarity was used for the remaining analyses [60]. Subsequently, a pairwise test (corresponding exactly to Gosset's original t statistic in PRIMER v.6 + PERMANOVA (software) [61] was performed in order to detect differences among areas.

Multivariate patterns were visualized through the multivariate canonical analyses of principal coordinates (CAP) [62] of areas elements. Species showing a Pearson correlation index > 0.4 were also shown in CAP plot. Distance-based permutation multivariate analyses of variance (PERMANOVA) were carried out on densities in order to test for differences in assemblage CSS structure among areas and sites.

Permutation tests of multivariate dispersion, a  $\beta$ -diversity variation measure (PERMDISP [63]), were also performed to investigate small-scale heterogeneity (i.e., among replicates) of CSS assemblages.

Anthropogenic pressures patterns were visualized by the multivariate three-dimensional non-metric multidimensional scaling (3D-nMDS) of Areas centroids. CSS densities were related to them through the multivariate distance-based linear modelling (DistLM [64]), using Step-wise as section procedure and adjusted R2 as a selection criterion.

Finally, fishing gears (independent variable) being significantly correlated with CSS densities were related by linear regression (ordinary LS) to direct impacts (GCE + BUC), indirect impacts (CNE) and heterogeneities (dependent variables).

All the statistical analyses were carried out through PRIMER v.6 + PERMANOVA software [61], except the linear regressions, which were performed with Past4® software [65].

### 3. Results

In total, an area of 8851 m<sup>2</sup> was investigated by ROV, with an average of about 164 m<sup>2</sup> per route. A total of 12 CSS (Table 1) was detected for the density calculation, while the frequencies of species, anthropogenic pressures and impacts were calculated using 74 images per route, for a total of 3996 images of about 0.2 m<sup>2</sup>.

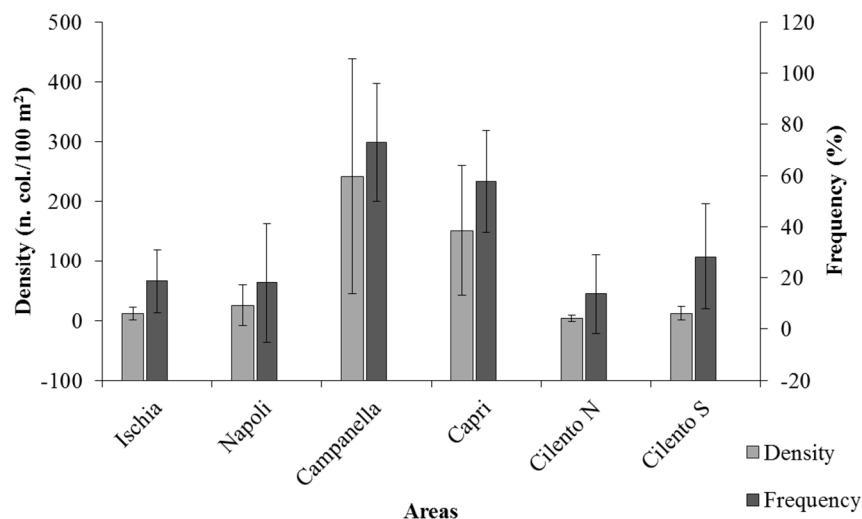
**Table 1.** Coralligenous structural species (CSS) densities, present in each investigated area and identified by ROV-imaging technique.

CSS	Ischia	Napoli	Campanella	Capri	Cilento N	Cilento S
<b>Porifera</b>						
<i>Axinella cannabina</i>					0.25 ± 0.25	
<i>Spongia lamella</i>					0.10 ± 0.29	
<i>Sarcotragus foetidus</i>			0.31 ± 0.46		0.04 ± 0.11	0.05 ± 0.14
<b>Cnidaria</b>						
<i>Corallium rubrum</i>			44.94 ± 103.86	2.56 ± 7.68		
<i>Eunicella cavolinii</i>	5.80 ± 5.67	12.98 ± 26.18	127.39 ± 100.06	104.43 ± 71.09	0.98 ± 2.22	0.77 ± 2.31
<i>Eunicella singularis</i>	0.61 ± 1.30	9.45 ± 21.06	14.14 ± 26.95	0.37 ± 0.82	0.30 ± 0.91	
<i>Savalia savaglia</i>					0.08 ± 0.25	
<i>Leptogorgia sarmentosa</i>		0.11 ± 0.33				0.04 ± 0.12
<i>Paramuricea clavata</i>	0.63 ± 1.60	1.75 ± 5.09	54.12 ± 56.72	31.64 ± 43.45	0.67 ± 2.00	
<i>Cladocora caespitosa</i>	0.21 ± 0.63					
<b>Bryozoa</b>						
<i>Myriapora truncata</i>	3.81 ± 4.78	0.58 ± 1.08	0.56 ± 0.72	9.59 ± 22.17	1.14 ± 1.54	10.58 ± 10.84
<i>Pentapora fascialis</i>	0.57 ± 1.17	0.12 ± 0.25	0.10 ± 0.29	2.32 ± 5.74		0.20 ± 0.40

Only 39% of these images showed CSS: in particular, Ischia presented 6 CSS with a frequency of 18.63 ± 12.35%, Napoli presented 6 CSS with a frequency of 18.12 ± 23.24%, Campanella presented 7 CSS with a frequency of 72.98 ± 23.03%, Capri presented 6 CSS with a frequency of 57.79 ± 19.92%, Cilento N presented 6 CSS with a frequency of 13.75 ± 15.42%, Cilento S presented 7 CSS with a frequency of 28.38 ± 20.61% (Figure 2). Values

ranged from a maximum of  $88.67 \pm 5.36\%$  in a route for Campanella, to a minimum of  $7.52 \pm 6.75\%$  in a route for Cilento S.

Average densities of total CSS (n. colonies/100 m<sup>2</sup>, Figure 2) show significant differences among areas ( $p = 0.0016$ ): Ischia  $11.63 \pm 10.79$  col./100 m<sup>2</sup>, Napoli  $25.00 \pm 34.22$  col./100 m<sup>2</sup>, Campanella  $241.56 \pm 197.06$  col./100 m<sup>2</sup>, Capri  $150.92 \pm 109.34$  col./100 m<sup>2</sup>, Cilento N  $3.21 \pm 5.28$  col./100 m<sup>2</sup>, and Cilento S  $11.81 \pm 10.98$  col./100 m<sup>2</sup>. In particular, CSS range between a maximum value of  $422.23 \pm 213.21$  col./100 m<sup>2</sup> at Campanella, and a minimum value at Cilento S, with  $0.70 \pm 0.61$  col./100 m<sup>2</sup>. Results of Pairwise tests, performed to detect differences among areas, are shown in Table 2.

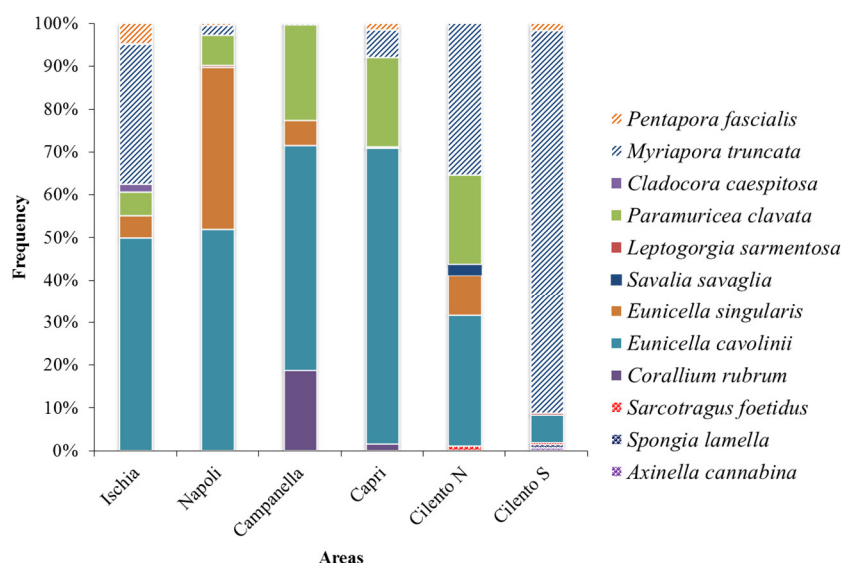


**Figure 2.** Average values of density (n. col./100 m<sup>2</sup>) and frequency (%) of coralligenous structuring species (CSS) in each investigated area. Bars represent SD.

**Table 2.** Pairwise tests among investigated areas for CSS densities. The significant  $p$ -values are shown in bold.

	Ischia	Napoli	Campanella	Capri	Cilento N	Cilento S
Ischia						
Napoli	0.68					
Campanella	<b>0.04</b>	<b>0.02</b>				
Capri	0.06	<b>0.03</b>	0.43			
Cilento N	0.21	0.24	<b>0.01</b>	<b>0.01</b>		
Cilento S	0.50	0.70	<b>0.02</b>	<b>0.02</b>	0.50	

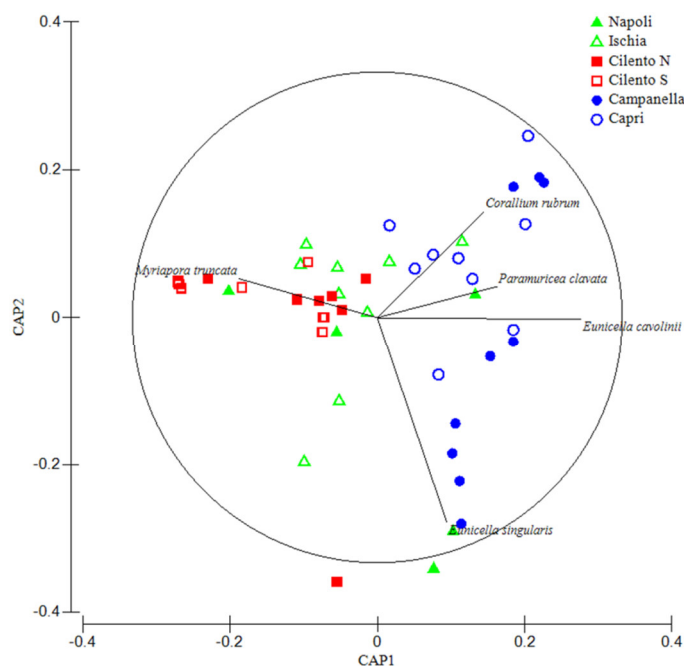
Figure 3 shows that the phylum Cnidaria mainly characterizes Ischia, Napoli, Campanella and Capri areas. In particular, *E. cavolinii* was a dominant species (respectively 50, 52, 53, and 69% of images containing CSS), followed by *E. singularis* in Napoli (38%), *P. clavata* and *C. rubrum* in Campanella (22% and 19%, respectively), and *P. clavata* in Capri (21%). In Ischia, the bryozoan *M. truncata* was the second dominant species (33%). This last species results the dominant species in Cilento N (35%), followed by *E. cavolinii* and *P. clavata* (31% and 21%, respectively), and Cilento S (90%).



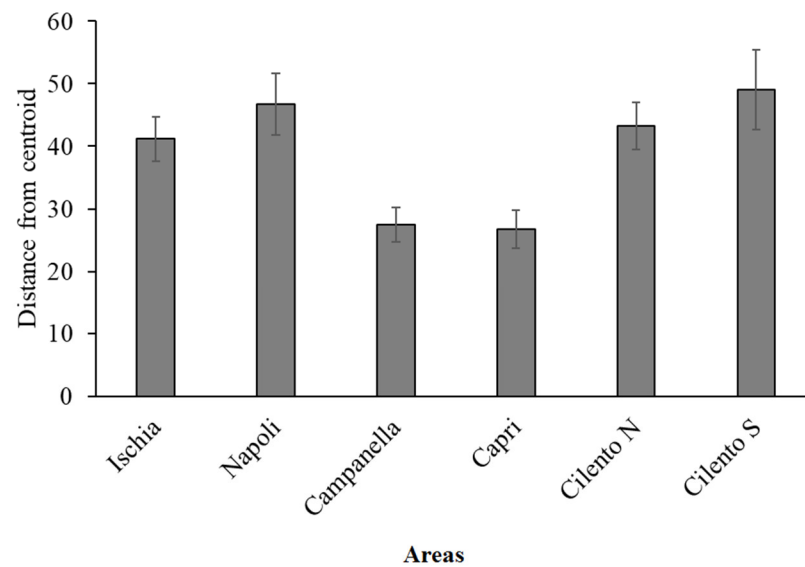
**Figure 3.** Species Dominance ( $D_i$  = frequency of  $i$ -th species on the total of CSS) in each area.

Such findings were confirmed by the CAP analysis (Figure 4), showing: a cluster composed by Campanella and Capri areas, polarized in the positive part of the CAP 1 axis, strongly related with cnidarian species, and a cluster composed by Cilento N and Cilento S areas, polarized in the negative part of the CAP 1 axis, strongly related with bryozoan species. Napoli and Ischia areas are scattered along the whole CAP 1 axis.

Multivariate PERMANOVA analysis on the species densities dataset showed significant differences both among sites ( $p = 0.0002$ ) and areas ( $p = 0.0066$ ); in particular, pairwise tests show that Cilento N and Cilento S were significantly different from Campanella and Capri ( $p < 0.05$ ). Indeed, the PERMDISP analysis ( $p = 0.0076$ ), and measure of  $\beta$ -diversity in terms of small-scale heterogeneities, showed the lowest values for Campanella and Capri and the highest values for the other areas (Figure 5).

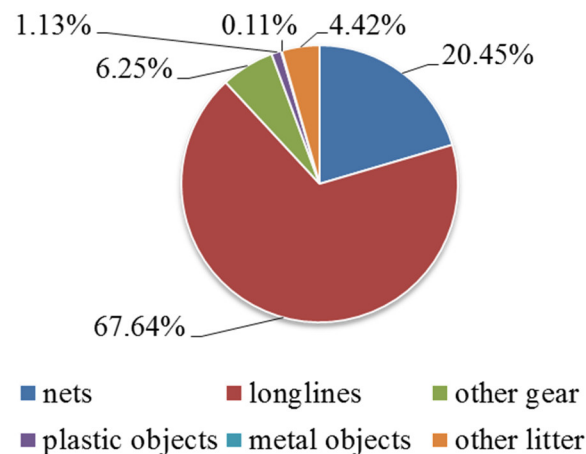


**Figure 4.** The CAP analysis on CSS densities; straight lines are vectors of species whose orientation and length are proportional to the most correlated areas. Only species with a correlation Pearson index  $> 0.4$  are shown. The circle represents a 95% confidence interval.



**Figure 5.** Mean ( $\pm$  SE) multivariate dispersion (PERMDISP) of replicates around areas centroids, calculated on CSS densities. High distances correspond to high heterogeneity and consequently high  $\beta$ -diversity in each area.

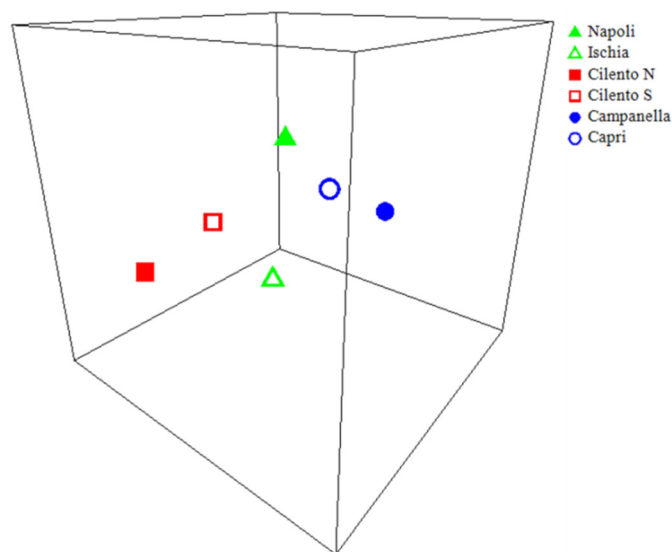
Anthropogenic pressure is mostly represented by longlines (67.6%), followed by nets (20.4%); while metal objects were the most frequent among marine litter (0.1%) (Figure 6). Capri is the most stressed area with the presence of gear and litter in the 32.7% of frames; while, Cilento N represents the less stressed site, with value of 4.6%. In particular, the medium values of fishing gear and marine litter for each area resulted: Ischia  $14.2 \pm 7.9\%$ , Napoli  $24.3 \pm 26.3\%$ , Campanella  $30.3 \pm 23\%$ , Capri  $32.7 \pm 23.4\%$ , Cilento N  $4.5 \pm 4.8\%$ , and Cilento S  $4.7 \pm 4.8\%$ .



**Figure 6.** Anthropogenic pressure is represented by frequencies of fishing gear and marine litter of total.

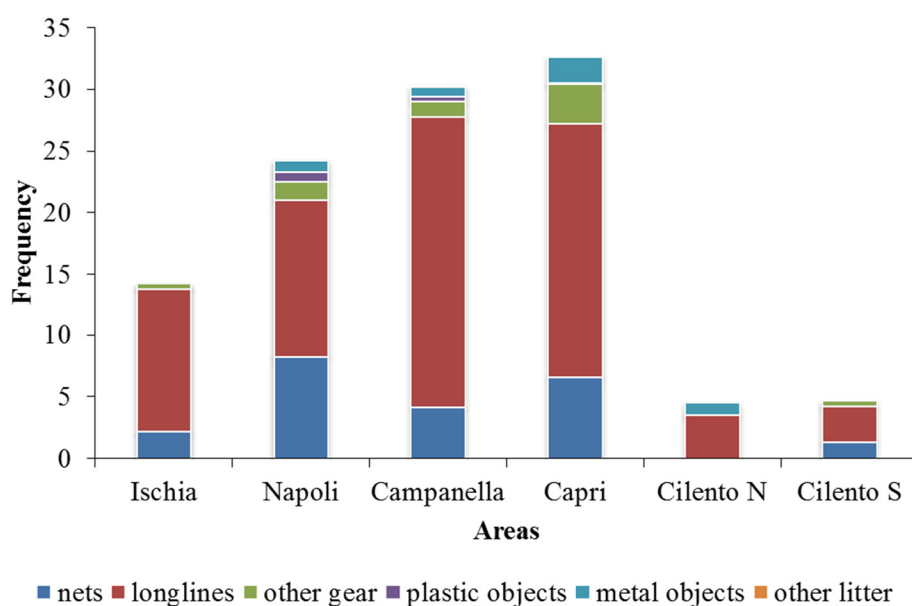
The nMDS ordination plot (Figure 7) performed on the different areas centroids show three evident clusters: Campanella–Capri, Cilento N–Cilento S, and in the middle Napoli–Ischia. Moreover, the distLM analysis highlights that longlines are the only gear significantly affecting coralligenous assemblages structures ( $p = 0.0004$ ,  $r^2 = 0.71$ ), explaining 94.5% of the total variation.





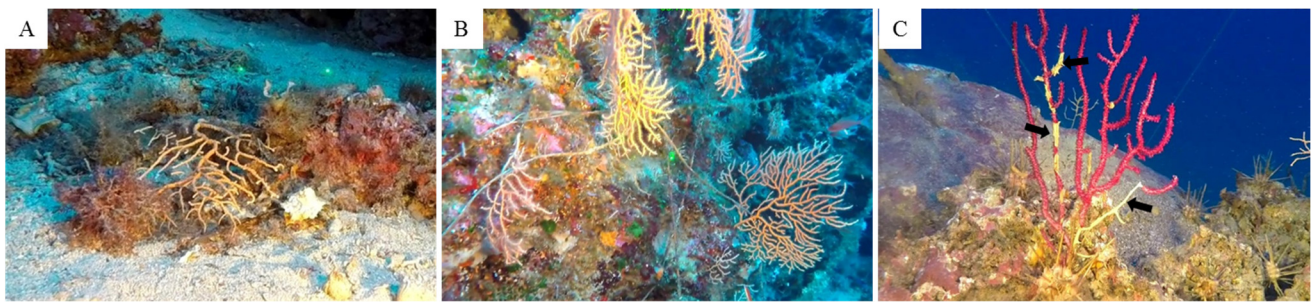
**Figure 7.** 3D-nMDS plot of anthropogenic pressure, from Euclidian distance matrixes. Points represent centroid of areas levels clusters in 3D multidimensional space, and the distance between any two points represents the difference between those two areas. The high quality of the ordination is indicated by a low-stress value (3D stress = 0.03).

Longlines are the most frequent gear detected in each area (Ischia 11.7%, Napoli 12.8%, Campanella 23.6%, Capri 20.6%, Cilento N 3.3% and Cilento S 2.9%). Second to longlines, nets are also frequent in all areas (Ischia 2.2%, Napoli 8.3%, Campanella 4.1%, Capri 6.6% and Cilento S 1.3%), except in Cilento N where metal objects were the second most frequent ones (1%), but their relation with assemblages structure was not significant (Figure 8). Areas with the higher values of marine litter results were Capri (2.2%), Napoli (1.8%) and Campanella (1.2%). Among marine litter, metal objects were the most abundant (4.4%), followed by plastic objects (1.1%) and other litter (0.1%).



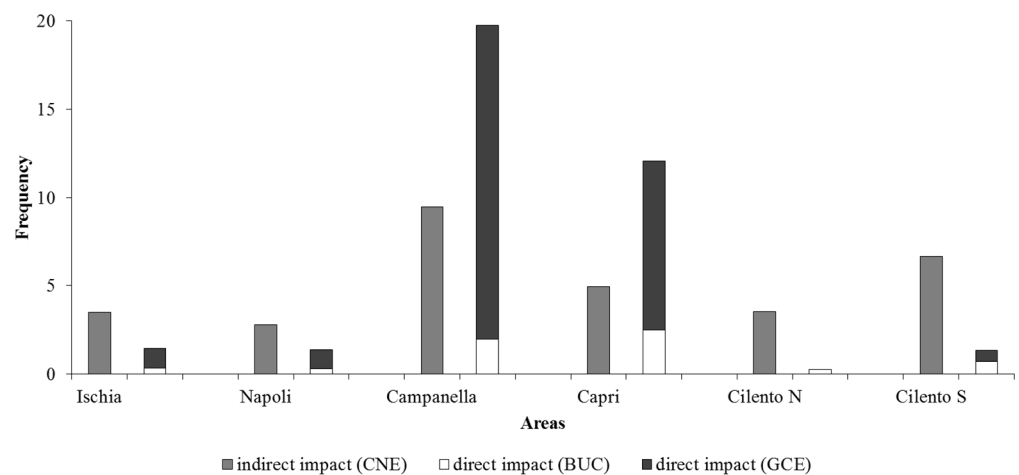
**Figure 8.** Frequencies composition of fishing gear and marine litter in each area.

Anthropogenic impact types direct (BUC = broken/upturned colonies and GCE = gears covering/entangling), and indirect (CNE = colonies with necrosis/epibiosis) are shown in Figure 9.



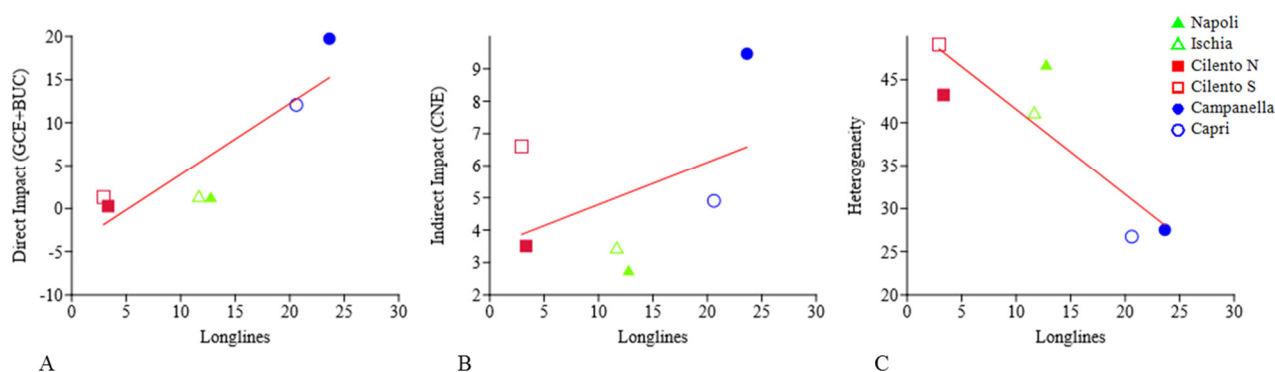
**Figure 9.** Examples of BUC (broken/upturned colonies, (A)), GCE (gears covering/entangling, (B)), and CNE (colonies with necrosis/epibiosis, (C)), black arrows show the necrotic parts).

In Figure 10, the frequencies of indirect and direct impacts detected in each area are shown. Campanella and Capri resulted in the most impacted areas (29.2% and 17% of frames), followed by Cilento S (7.9%), while Ischia, Napoli and Cilento N resulted in the less impacted areas (4.9%, 4.1% and 3.8%). In particular, CNE impact is the most frequent in Ischia, Napoli, Cilento N and Cilento S areas ( $3.5 \pm 3.3\%$ ,  $2.8 \pm 4.7\%$ ,  $3.5 \pm 8.8\%$  and  $6.6 \pm 5.2\%$ , respectively). Instead, GCE is the most frequent impact in Campanella and Capri areas ( $17.8 \pm 15.7\%$  and  $9.6 \pm 6.7\%$ , respectively).



**Figure 10.** Frequencies of indirect (CNE) and direct (BUC and GCE) impacts detected in each area.

Linear regressions between longlines and direct impact, indirect impact, and heterogeneity are showed in Figure 11 and Table 3. In particular, a significant positive relation ( $p = 0.0086$ ,  $r^2 = 0.77$ ) is detected between longlines and direct impact; while, no significant relation is detected between longlines and indirect impact. Finally, a significant negative relation ( $p = 0.035$ ,  $r^2 = 0.77$ ) between longlines and heterogeneity.



**Figure 11.** Linear regressions between longlines and direct impact ((A),  $p = 0.0086$ ,  $r^2 = 0.77$ ), indirect impact ((B), no correlation), and heterogeneity ((C),  $p = 0.035$ ,  $r^2 = 0.77$ ).

**Table 3.** Mean ( $\pm$  SD) of linear regression independent (longlines) and dependent variables (direct impact, indirect impact, and heterogeneity).

	Longlines	Indirect Impact (CNE)	Direct Impact (BUC + GCE)	Heterogeneity
Napoli	12.76 $\pm$ 13.13	2.77 $\pm$ 4.68	1.36 $\pm$ 1.73	46.78 $\pm$ 4.91
Ischia	11.67 $\pm$ 8.46	2.51 $\pm$ 4.77	1.44 $\pm$ 2.92	41.2 $\pm$ 3.57
Cilento N	3.35 $\pm$ 4.25	2.51 $\pm$ 4.77	0.26 $\pm$ 0.78	43.23 $\pm$ 3.78
Cilento S	2.93 $\pm$ 2.49	2.73 $\pm$ 4.69	1.33 $\pm$ 2.13	49.07 $\pm$ 6.41
Campanella	23.64 $\pm$ 18.51	3.39 $\pm$ 4.68	19.74 $\pm$ 18.19	27.52 $\pm$ 2.74
Capri	20.61 $\pm$ 14.11	4.27 $\pm$ 4.7	12.07 $\pm$ 9.07	26.74 $\pm$ 3.1

#### 4. Discussion

Campanella and Capri represent the areas with the highest number of organisms. In all areas, however, as evident in Figure 2, density and frequency of CSS show a regular trend, typical of a random spatial species distribution, possibly due to the absence of limiting factors controlling species eco-physiology (e.g., temperature, water, substrate type) [66].

Although the species richness in terms of CSS number among areas is consistent, changes in the species composition were observed; in particular, the phylum Cnidaria dominates in Napoli, Campanella and Capri, while the phylum Bryozoa dominates in Cilento N and Cilento S. This result may also depend on the different abiotic factors among areas, such as the substrate type, the hydrodynamic regime, the depth range, or the water trophism; for example, large gorgonians show a higher preference for trophic and hydrodynamic waters [67]. Coralligenous assemblage's structure and composition are strongly related to edaphic factors, which may act on their heterogeneity [51,68]. Some studies revealed that extreme environmental conditions may promote the growth of coralligenous assemblages and large suspension feeders [69]; indeed, the most abundant gorgonian species found in the study area grow in low-light conditions, induced probably also by the high turbidity and the prevalence of strong currents [57,70,71], that in the study areas may be near-bottom or upwelling, generating water turnover and promoting nutrient dispersion [72,73].

CSS densities in the Campanella and Capri areas are significantly different from those of the Cilento N and Cilento S areas, and appear to be less heterogeneous in terms of  $\beta$ -diversity, despite the higher densities values. This condition may be the result of high dominance values of few species, possibly due to anthropogenic impact [74]. Indeed, Campanella and Capri are mostly dominated by the large gorgonians *E. cavolinii* and *P. clavata* that are highly impacted, since their morphological structure may increase the likelihood to be entangled and damaged by fishing gear. On the contrary, Cilento areas are characterized by the presence of small bryozoans, such as *M. truncata* and *P. fascialis* that

may decrease the likelihood to be entangled. Several studies have demonstrated that low diversity is linked to a high ecosystem impact. For example, some authors, i.e., [75], showed that impacted assemblages on artificial bottoms present fewer taxa and lower values of diversity, suggesting that those support a less diverse and more homogeneous assemblage than natural substrates. Moreover, communities characterized by high  $\beta$ -diversity should be more resistant to disturbance than others, acting as refuges for neighboring patches [76,77]. High diversity levels make communities able to persist and absorb fluctuations, increasing the resilience potential and reducing the fragility of marine communities and ecosystems [47].

Campanella and Capri are the most stressed areas with the highest numbers of fishing gear and marine litter, followed by Napoli and Ischia, while Cilento N and Cilento S are the least stressed ones with very low values. Generally, longlines dominate in all the sampled areas, followed by nets, with very high numbers in Campanella and Capri. Nets are more abundant in the Napoli area, where the coralligenous bioconstructions are present in the form of banks surrounded by muddy substrate. Longlines are widespread because of their dual-source from both professional and recreational fishing, while nets should be considered mainly professional gear [78]. Moreover, longlines can be used on different bottom types, while the nets are usually set on soft bottoms, even though they may get stuck on hard bottom, with relevant damages for the benthic communities [32,79,80].

Marine litter is the most abundant along the coastal areas of Capri, Napoli and Campanella; in particular, the Napoli area is close to a large urban center, while Capri and Campanella areas are located along popular touristic routes. Despite the highest impact of fishing gear, linked to the developed professional and recreational fishing activities, the use of the coasts for recreational tourism purposes may be responsible for the accumulation of anthropogenic debris, as well as the rivers input [44]. In addition, recreational beach activities can possibly contribute to marine litter accumulation along the coastal areas [81].

CSS colonies with necrosis or epibiosis are present in all areas, with the CNE impact results the highest in Ischia, Napoli, Cilento N and Cilento S. However, they appear lower than 5% in Ischia, Napoli and Cilento S, and between 5 and 10% in Campanella, Capri and Cilento N. These values may be considered respectively low and moderate since the impact is usually considered elevated for values higher than 10% [52,82]. Fishing gear may either eradicate the large colonies or get entangled in the ramifications, scraping their coenenchyma with consequent necrosis tissues and favoring the development of epibionts, which may lead to a burdening of the colonies and to a greater mechanical stress, increasing their resistance to water movement [28,80]. Necrosis, and more extensively, colonies mortality, may also be caused by different stressing factors such as water temperature increase, possibly linked to climate change [83]. The heatwave of 2003 in Europe caused an anomalous warming of seawater, which played a key role in the observed mass mortality event of fan corals [84,85], as well as climatic anomalies, which can generally cause biodiversity loss and ecological shifts [86,87]. Necrosis may also be caused by a high sedimentation rate, as a consequence of resuspension caused by trawl-fishing [88], altering metabolic functions, enhancing respiratory and interfering with the prey-capturing apparatus, and also compromising the settlement [89]. For this reason, the identification of the effective cause of this indirect impact can be difficult.

Direct impact, mostly represented by GCE, prevailed in Campanella and Capri areas, where it is strictly connected to the high presence of fishing gear. Erect or branched organisms among Cnidaria, Porifera and Bryozoa are the most endangered by fishing gear since they are often broken and upturned [28,90–92]. Direct impact was mostly caused by the presence of abundant longlines, while no relation was found between indirect impact and longlines. Moreover, an indirect relation between heterogeneity and longlines was detected, demonstrating that fishing activity may cause severe species diversity loss [93–95].

A better knowledge of the anthropogenic activities impact on benthic communities of mesophotic reefs is increasingly needed in order to improve the management tools to protect one of the most fragile and valuable Mediterranean habitats. This should be reached through sustainable fishing techniques, adequate protection measures and long-term monitoring programs. Environmental protection, also through Marine Protected Areas, should respond not only to an ecological purpose but also to socioeconomic dimensions of sustainability [96].

**Author Contributions:** Conceptualization, R.S. and G.F.R.; data curation, F.F., L.A. and L.D.; formal analysis, F.F.; investigation, F.F., L.D., F.D.S. and F.R.; methodology, L.A.; software, L.A.; writing—original draft, F.F.; writing—review and editing, R.S. and G.F.R. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research received no external funding.

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Acknowledgments:** We sincerely thank the anonymous reviewers for their valuable comments and suggestions, which helped us to improve the quality of the manuscript. Thanks are due to the crew members and captain of R/V Cormorano V, ROV pilots and the informatics system technicians of Subonica Ltd. for field activities and data acquisition.

**Conflicts of Interest:** The authors declare no conflict of interest.

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