



Visual faunistic exploration of geomorphological human-impacted deep-sea areas of the north-western Mediterranean Sea

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Visual faunistic exploration of geomorphological human-impacted deep-sea areas of the north-western Mediterranean Sea

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19 *Abstract*

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33 **Key-words:** north-western Mediterranean; ROV; submarine canyons; seamounts; landslides;
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INTRODUCTION

The identification of deep-sea “essential habitats” is currently a major focus of European Community research programs with the aim of furthering the conservation and management of benthic biodiversity (Salomon 2009). In this context, faunistic surveys in cold seeps, mud volcanoes, seamounts, and canyons as “hot spots” for local biodiversity are of strategic relevance in the global context (Orejas *et al.* 2009, Fabri *et al.* 2014, Angeletti *et al.* 2015). In this scenario, *in situ* video observations of Mediterranean deep-sea fauna are still much reduced in comparison to those conducted to date in other oceans (Cunha de Jesus & Cancela da Fonseca 1999, Stein *et al.* 2005, Buhl-Mortensen & Buhl-Mortensen 2008). Nevertheless, the deep Mediterranean Sea hosts a complex collection of geologically and ecologically relevant environments that can vary across the short geographic scale of a few kilometers, hence resulting in a potentially highly variable faunal composition (Orejas *et al.* 2009, Cartes *et al.* 2009, Papiol *et al.* 2012, Fanelli *et al.* 2013, Mecho *et al.* 2014) that remains, to date, largely unknown in several areas, including the north-western (NW, hereafter) Mediterranean Sea (Danovaro *et al.* 2010).

Three distinctive geomorphological structures mostly occur in the NW basin: canyons, seamounts and open slopes. Large submarine canyons, deep incisions in the continental margin, occur just a few miles off the coastline in close proximity to each other. Canyons concentrate and then funnel downward all sediment, including organic particles (Puig *et al.* 2003, Canals *et al.* 2006, Company *et al.* 2012), hence affecting the local current regimes (Flexas *et al.* 2008, Bahamon *et al.* 2011). Their biodiversity has been the object of intense research in the past two decades in various oceans (Company *et al.* 2008, McClain & Barry 2010, Duffy *et al.* 2014). Seamounts, defined as topographic structures that rise above the surrounding seafloor, also occur in the Mediterranean basin (Acosta *et al.* 2003). Typically, their morphology is characterized by an exposed hard substratum that makes them ideal spots

63 for sessile filter-feeder fauna (Koslow 1997, Samadi *et al.* 2006, Howell *et al.* 2010). Finally,
64 a third type of structure is represented by muddy landslides which occur on continental
65 shelves and slopes, resulting in mud plains with several outcrops (Camerlenghi *et al.* 2010).

66 A broad knowledge of species distribution and biodiversity within these various
67 different geomorphologies is still poor for the NW Mediterranean, with some areas (e.g.
68 certain canyons or, in general, the slopes) more studied than others, relative to the commercial
69 trawl fisheries. In these areas, scientific surveys have been conducted in an attempt to achieve
70 faunal data for the integrated management of exploited stocks (Abelló *et al.* 2002, De Mol *et*
71 *al.* 2008, Bahamon *et al.* 2009). In general, one should bear in mind that most of NW
72 Mediterranean areas are presently threatened by a highly diversified typology of
73 anthropogenic impacts. These are not only related to the commercial fishery itself (e.g.
74 trawling as well as lost or discarded gears and longlines: Martín *et al.* 2008, Ramirez-Llodra
75 *et al.* 2010, Puig *et al.* 2012, but also from the accumulation of litter (Galgani *et al.* 1996,
76 Hess *et al.* 1999, Ramirez-Llodra *et al.* 2013), whose decomposition acts on the metabolism
77 of species and on the dynamics of the resulting trophic webs (Koenig *et al.* 2012, 2013a,
78 2013b). For all these reasons, anthropogenic impacts on deep-sea ecosystems are presently a
79 source of concern for both the science community and policy makers everywhere (Miyake *et*
80 *al.* 2011, Ramirez-Llodra *et al.* 2011, Woodall *et al.* 2015).

81 ROV video-imaging surveys have increased worldwide in recent years as an efficient
82 survey methodology, delivering key faunistic data on species composition, ethology, and
83 overall anthropic impacts (Galgani *et al.* 2000, Miyake *et al.* 2011, Ramirez-Llodra *et al.*
84 2011, Fabri *et al.* 2014, Mecho *et al.* 2014), in an ecologically more ethical manner (i.e. with
85 no damage to the explored environments, unlike trawling). In this context, the objective of the
86 present study is to describe, by means of ROV imagery, the megabenthic communities of
87 various deep-sea geomorphologic areas within the NW Mediterranean. Fauna from one

canyon, two seamounts and two landslides were observed and quantitatively described. In addition, we quantified anthropogenic impact within each area, reporting at the same time relevant ethological observations, as an important ecological by-product of this exploration.

MATERIALS AND METHODS

Data collection

The ROV *Max Rover II* of the Hellenic Centre of Marine Research (HCMR) was used to conduct visual observations during the research cruise EUROLEON, which was conducted in October 2007 on Mediterranean Spanish waters aboard the *R.V. BIO Hespérides*. The ROV was equipped with two wide-angle color CCD cameras with a resolution of 3.2 Mpixel, 1Gb, offering a frontal and a lateral view, plus a third with a macro-zoom. Lighting was provided by 2 x 100 W HID lights and 4 x 150 W quartz lights. The ROV speed and height above the seabed during filming operations were approximately 1.2 knots and 1.5-2.0 m, respectively. The resolution was constant along transects. The limit of detection depended on the ROV distance to the bottom. In some cases, the presence of mud clouds could result in a diminution of the limit detection.

Seven transects (here after termed as “dives”) were conducted for a total of 14.5 km surveyed (equivalent to a total of 30 hours of video; Table 1). Three different NW Mediterranean distinct geomorphological zones were inspected (Figure 1): the continental margin off Blanes, the Gulf of Valencia, and the Eivissa Channel (also known as Ibiza Channel). In particular, dives occurred as follows (see Table 1): dives 1 (41° 38' N - 02° 52' E) and 2 (41° 39' N - 02° 53' E) at the head of the Blanes canyon; dive 3 on an unreported seamount in the Gulf of Valencia (39° 30' N - 00° 17' E); and dives 4 through 7 in the Eivissa Channel. In particular, for this latter area, two dives (4 and 5; 38° 39' N - 00° 55' E) were

conducted along a small flat-topped seamount, and the other two (dives 6 and 7 (38° 41' N - 00° 50' E) were performed close to the escarpments of two large submarine landslides (named *Jersi* and *Ana*; Berndt *et al.*, 2012; Lafuerza *et al.*, 2012; Lastras *et al.*, 2004).

Data processing and analysis

All video footages considered for animal taxonomic identification and counting were obtained with the frontal camera and inspected in a *time-lapse* mode (i.e. at 50% of acquisition rate). Video analysis was conducted using the software application Intervideo WinDVD 9.0 (Windows). All observed organisms larger than 5 cm were identified as faunistic entries (i.e. smaller animals were not visible), being classified to the lowest taxonomical level as possible. For a more precise taxonomic determination, digital frames were extracted after video partitioning. Classification was accomplished by the use of current taxonomic guides for the Mediterranean (Zariquiey 1968, Riedl 1983, Mercader *et al.* 2001).

Data on faunal composition were annotated according to their timing of occurrence in the video footage (hence allowing correlation with ROV navigation data for a precise geographic positioning) along with concomitant annotations on the substrate type, classified as mud, rock, sand, and coral rubble, as well as on anthropogenic artifacts.

Data analyses were carried out considering faunal entries grouped within classes, to avoid those classification mistakes that may occur in ROV studies when attempting a more precise classification when no concomitant sampled specimens are available for comparison. Faunistic comparisons among different substrate types and depth ranges were carried out by grouping class entries by 100 m of ROV navigation track distance. Then, faunal data were compared across different geomorphologies. The same analysis was performed for anthropogenic impact.

Although all our statistical analyses were performed with class-level data (see below), for a better visualization of faunistic spatial trends, the numbers of individuals were plotted each 100 m according to the five most frequently observed phyla (Porifera, Cnidaria, Echinodermata, Brachiopoda and Chordata) and subphyla (i.e. Crustacea) and represented along the dive in the Appendix section. Finally, behavioral observations were reported and classified when occurring in videos more than twice (Stoner *et al.* 2008).

Statistical methods

The level of similarity of class sampling composition among 100 m splits within a dive and among dives in the same or different geomorphologic areas was assessed using the Non-metric Multidimensional Scaling (NMDS) method (Minchin 1987). The function metaMDS in the ‘vegan’ library in R (Oksanen *et al.* 2013) was used to find both non-parametric relationships and Bray-Curtis dissimilarities between classes. To fit the area parameters (gradients of depth, type of substrate and anthropogenic impact) to taxa ordination, two functions in the vegan library were used. The function ‘envfit’, based on permutation tests, allowed fitting centroids of the levels of the factor variables “sediment type” and “study area” into the ordination of the taxa. The variable “anthropogenic impact” was not significant. Therefore, it was not plotted onto the taxa ordination. Finally, the function ‘ordisurf’, based on thinplate splines (Wood 2003) with cross-validation selection of smoothness (Marra & Wood 2011), allowed fitting smooth surfaces for the continuous variable “depth” onto the taxa ordination using restricted maximum likelihood (REML) as smoothing parameter estimation method.

RESULTS

General remarks

We observed a total of 4534 individuals, considered different faunistic entries (Table 2) in the various geomorphological areas surveyed (i.e. canyon, seamount, and landslide) (see Figure 1). A comprehensive list of these entries, classified to the species level (when possible), is provided in Appendix 1. The fauna belonging to the classes Actinopterygii, Malacostraca, and Anthozoa were the most abundant, representing 24%, 20%, and 14% of all observations, respectively (see Figure 2). The class Demospongiae was less abundant (12%), with an occurrence similar to those of Rhynchonellata (11%) and Scyphozoa (9%). The abundance of all other remaining invertebrate classes was less than 3% each.

NMDS results showed the presence of a significant effect of depth on species ordination, taking all the inspected areas both together and within each area (see Figure 3). Area and sediment type were significantly related to the class ordination only when areas were considered together (Table 3; see Case 1). No significant effect of anthropogenic factors was found. When we considered all the classes in the three areas taken together (see Figure 3A), we observed that Asteroidea, Echinoidea, and Holothuroidea were associated with shallower sandy areas, whereas Ophiuroidea, Crinoidea, and Cephalopoda occurred primarily in deeper zones on muddy flat slopes.

Canyon head

A total of 792 faunistic observations were made on the western flank of the Blanes canyon head (see Table 2 and Figure 1). Both dives were similar in setting, with the exception of the southern dive 2, which crossed an area with a steeper slope in its deepest section. Two types of substrates were observed: a muddy area in the deepest part and a sandy area with strong tanathocenosis (i.e. assemblages of dead shells within the sediment), this latter on the shallower part of both dives 1 and 2. Globally, the class Anthozoa was the most reported in

the Blanes Canyon head, with 31% of the total observations (Figure 4). This group had also the highest number of individuals per group (i.e. the Anthozoan *Pennatula* spp. with 158 observations, see Appendix 1). Class Malacostraca represented 26% of the total observations, most of them corresponding to the infraorder Brachyura (i.e. crabs). Malacostraca was followed by Actinopterygii (20%) and Rhynconellata (7%), with all the remaining classes less than 5% each.

On the western flank of the Blanes canyon head, we could distinguish 3 different faunistic distributions (Appendix 2) coinciding with depth and slope changes. The deepest part surveyed (450 - 250 m) showed a low number of observations and a high number of classes. In general, canyon dives showed a two-step slope change at 250 - 300 m and 150 m depths. The deeper areas with steep muddy slopes were dominated by crustaceans. From 150 to 60 m depth, the seafloor is relatively flat and was dominated by the phyla Cnidaria (class Anthozoa, mostly *Pennatula* spp.) and Echinodermata, primarily the class Asteroidea, with the species *Anseropoda placenta* (Pennant, 1777), and the Holothuroidea, with *Parastichopus regalis* (Cuvier, 1817).

A NMDS analysis of the observations from the canyon revealed a significant effect of depth on class ordination (see Table 3, Case 2) but not of sediment typology and anthropogenic impact. At the faunistic level, there is a highly similar group composed by Actinopterygii, Malacostraca, Anthozoa and Elasmobranchii. This group of taxa is dissimilar to Demospongiae, Hydrozoa, Gastropoda and Thaliacea (see Figure 3B).

Seamount

Two seamount dives were analyzed (see Figure 1), one in the Gulf of Valencia and the other in the Eivissa Channel (see Table 1). The first seamount presented a conical morphology surrounded by a muddy plain. A total of 10 hours of images were recorded at this

212 site. The second seamount, in the Eivissa Channel was surveyed separately on its eastern
213 flank and on its flat top. The results are described separately below for each seamount, and a
214 general analysis is then presented for both seamounts.

215 The Gulf of Valencia seamount rises from a depth of 800 m (Appendix 3). Its top is at
216 450 m. It was characterized by two types of substrates: a rocky area constituted by steep
217 slopes combined with rocky substrata (from 450 to 600 m depth) and a large muddy plain
218 surrounding the rocky area, from 600 to 800 m depth. A significantly denser concentration of
219 benthic fauna was observed in the shallowest rocky areas (Appendix 3A), in contrast with a
220 drastic diminution of that fauna toward the deepest muddy zones (Appendix 3B). The
221 seamount presented 2 well separated faunistic distributions, which were related to these
222 substrates and depth. The rocky substratum was located on the flank of the seamount
223 (Appendix 3A) and presented a fauna composed basically of benthic species of the classes
224 Demospongiae (31% of the total observations within the rocky area), Anthozoa (25%, benthic
225 species such as corals, anemones and gorgonians) and Brachiopoda (28%). The second
226 substratum, the muddy plain surrounding the rocky area (Appendix 3B), was dominated by
227 crustaceans of the class Malacostraca (33% of the total observations), the class Actinopterygii
228 (32%), and Anthozoa (mostly deep-sea anemones of the genus *Cerianthus*, 22%). In the case
229 of the muddy plain, the distribution of the benthic communities was patchy along the dive and
230 was related to subtle changes of slope and substrate (Appendix 3B).

231 On the Eivissa Channel seamount (see Figure 1), two areas were studied: the upper
232 slope (flank) and the flat top (Appendix 4). At its bottom, we observed a flat area mainly
233 composed of mud with boulders (Appendix 4A). This area was dominated by motile fauna
234 such as classes Malacostraca (24% of the total flank observations) and Actinopterygii (22%),
235 but included also sessile fauna (24%, as benthic cnidarians on cobbles). Moving upwards, the

flank was constituted by rocky outcrops dominated by the benthic classes Demospongiae (14%) and Rhynchonellata (8%).

Dive no. 5, over the flat top of the Eivissa Channel seamount encompassed only one substratum type, a bioclastic sand with sparse rocky outcrops (Appendix 4B). This transect covered the shallowest parts (196 – 250 m depth) of the surveyed area, and it was dominated by motile fauna, including Actinopterygii (48%), class Scyphozoa (26%, mainly *Pelagia noctiluca* (Forsskal, 1775)) and Holothuria (14%, one species, *Holothuria (Holothuria) tubulosa* Gmelin, 1791). The large number of fish schools observed over the rocky areas of the top of the seamount was noteworthy.

A total of 2290 faunistic observations were reported from both seamounts over a distance of 6.5 km (see Table 2). The most commonly observed groups were the benthic classes Demospongiae (24%) and Rhynchonellata (19%) (see Figure 4B). These groups were followed by Actinopterygii (16%) and Anthozoa (15%). The classes Malacostraca, Scyphozoa and Holothuroidea were less abundant in the seamount dives, representing 16%, 8% and 2%, respectively of the total observations.

The NMDS analysis conducted in the seamount showed that the factors depth and sediment were significantly affecting the distribution of the classes (see Table 3, Case 3). Classes Holothuroidea, Polychaeta, and Thaliacea were associated with shallower sandy areas, while Rhynchonellata and Demospongia were associated with medium depths and rocky areas. In contrast, Crinoidea, Gastropoda, and Ophiuroidea showed a preference for deeper muddy areas (see Figure 3C).

Submarine landslide

Two submarine landslides (*Jersi* and *Ana*) were surveyed in the Eivissa Channel (see Figures 1 and Table 1). The landslide scars were made up by consolidated sediments and, in

the *Jersi*, even rocky pebbles and coral rubble were observed. The depositional areas were instead composed of mud, similar in gross morphology to the undisturbed upslope area (i.e. above the scars). As for the seamount, the results are described first separately for each landslide and then in general terms (including both landslides).

When we considered the landslides separately, we found *Jersi* dominated by the classes Malacostraca (60% of the total observations of this landslide) and Actinopterygii (19%). None of the other groups exceeded 8% in this landslide. We observed an increase of crustaceans on the scar area in front of the depositional area. Nonetheless, this landslide presented a generally constant faunal composition along all its surveyed area (Appendix 5A).

The substratum along the *Ana* landslide was mostly mud (Appendix 5B). The sediment along the scar appeared more consolidated. Actinopterygii (44% of the total observations of this landslide) dominated that area, followed by the classes Malacostraca (24%), Scyphozoa (19%), and Ophiuroidea (10%). The latter class was more abundant here than on the mud plain.

Considering both landslides together, the most representative groups were the classes Actinopterygii and Malacostraca, representing 40% and 32% of the total observations, respectively (see Figure 4C), followed by Scyphozoa and Ophiuroidea (respectively 14% and 9%). The other classes represented 5% of all the observations.

The different faunal groups identified fit well with the topographic features recognized on the bathymetry (see Appendix 5A, B). Landslide scars, deposits and undisturbed seafloor had different phyla compositions and abundances. The most observed fauna in the scars were the crustaceans of the class Malacostraca. Pelagic cnidarians (order Coronatae) and fishes dominated the landslide deposits. Finally, crustaceans and ophiurans (brittle stars) dominated the undisturbed seafloor upslope of the landslides. These observations were supported by an NMDS analysis. In the landslide area (see Figure 3D), this analysis indicated that depth

significantly influenced class composition (see also Table 3). In contrast, both the substrate type and anthropogenic impact did not influence the detected faunal distributions.

Anthropogenic impact

A noticeable level of anthropogenic impact was observed in all studied zones, with 158 recorded artificial objects of various types detected. These items included plastic bags, cans, and bottles (see Figure 5A). Trawl marks were also consistently observed (see Figure 5B). Finally, lost or discarded fishing gears were also detected, including longlines (see Figure 5C) and the remains of hauling fishing nets (see Figure 5D).

Overall, litter was the most abundant observation (39%), followed by trawl marks (30%) and longlines (28%), with lost or discarded nets being less abundant (3%) (Figure 6A). In the canyon head, plastic bags and bottles represented 79% of the total observations, whereas longlines represented only 14%. A minority of the observations (7%) were related to trawl marks. No fishing nets were detected (see Figure 6B). On the seamounts and their surrounding areas, 58% of the anthropogenic impact referred to the presence of longlines, with a significant amount of other litter (22%), trawl marks (16%), and only 4% of discarded fishing nets (see Figure 6C). On the landslides, approximately half (45%) of the total anthropogenic observations were represented by trawl marks and other litter (44%), with longlines (9%) and fishing nets (3%) less representatives (see Figure 6D).

Behavioral observations of identified species

Several behavioral observations were made for motile fauna during the ROV surveys. Within the class Malacostraca, individuals of the family Galatheoidea were observed to maintain their positions, extending forward their claws as the ROV approached, suggesting the performance of territorial and aggressive defense behavior. Burrowing behaviour was

observed in an isolated individual of Norway lobster (*Nephrops norvegicus*, Linnaeus, 1758) at 670 m depth (see Figure 7A). This animal showed motile activity in relationship to the patrolling of different burrow entrances, entering and exiting from them. Another behavior displayed by Malacostraca was related to camouflage. This was observed in six individuals of the crab *Paromola cuvieri* (Risso, 1816), which were carrying white plastic bags and other artifacts on their carapace (see Figures 7B, C).

Fish behavior was also noted in relationship to their reactions to the approaching ROV. Evasion was typically observed in individuals of the family Macrouridae (see Figure 7D), while other fishes (i.e. order Scorpaeniformes) did not show alterations in their behavior. Schooling behavior was reported for *Trachurus* sp. (Linnaeus, 1758), *Pagellus bogaraveo* (Brünnich, 1768), *Capros aper* (Linnaeus, 1758), and *Lepidopus caudatus* (Euphrasen, 1788) (see Figure 7E).

Finally, a peculiar observation was reported in relation to jellyfishes, mostly *Pelagia noctiluca* (Forsskal, 1775) and specimens from the order Coronatae, which were observed swimming a few centimeters over the seabed. In particular, small groups of *P. noctiluca* were observed touching the seafloor over the top of the flat seamount in the Eivissa Channel (see Figure 7F).

DISCUSSION

We conducted ROV video-observations of the benthic communities inhabiting a group of diverse geomorphological areas in the NW Mediterranean Sea for which poor faunistic information is to date available. Classes' composition was mostly related to canyon, seamount, and landslide in a site-specific manner. Anyway, depth was the principal parameter that shaped the zonation in our faunistic observations as already reported for other

Mediterranean areas (D'Onghia *et al.* 2003). This parameter constrained the presence of some species at certain locations, in a fashion that appeared to be independent of the different geomorphological character of the surveyed area and the type of substrate. For example, some shallow-water species (i.e. the anthozoan *Pennatulula rubra* (Ellis, 1761), the asteroid *Anseropoda placenta* (Pennant, 1777), and the holothurian *Parastichopus regalis* (Cuvier, 1817), were never observed in deeper areas, even when suitable substrata were available, confirming species distribution ranges as observed by trawling (Sardà *et al.* 1994, Moranta *et al.* 1998, D'Onghia *et al.* 2003). Similarly, deep-living and highly motile species such as shrimps of the genus *Plesionika* spp. or fishes belonging to the order Stomiiformes and those within the family Myctophidae were only observed below a depth threshold.

Substrate type also plays a strong role in driving species composition in different geomorphological areas within a certain geographic region. Recurrent species composition across geography is of importance for the establishment of canyons, open slope, seamount, and landslides as valid seascape units (Longhurst 1998, Levin *et al.* 2010). According to these considerations, we decided to discuss our results separately for each geomorphological zone.

Canyons

The majority of the observations in the Blanes canyon corresponded to sessile fauna such as anemones, sea pens and fans, or tubeworms. All these taxonomic groups are suspension feeders and are common in canyons of the Catalan margin (Ramirez-Llodra *et al.* 2008, 2010, Company *et al.* 2012). A variable topography and physical characteristics has profound effects on the community structure within the canyon itself and in the surrounding slope areas (Genin 2004, Tecchio *et al.* 2011, 2013, Papiol *et al.* 2012, Company *et al.* 2012, Fanelli *et al.* 2013). In the specific case of Blanes canyon, an internal downstream flux of

sediment takes place at a rate three times higher than on the surrounding open slope (Zúñiga *et al.* 2009).

Seamounts

Faunistic differences between the Gulf of Valencia and the Eivissa Channel seamounts were observed. These differences are related to their topographic characteristics and depth, in turn influencing substrate types, local hydrography, and, most likely, food availability. The seamount of Valencia, with its conical shape, presented a sponge's community and a hard coral fauna, related to the abundance of hard substrate. On the other hand, the flat and shallower (195-250 m) topped seamount in the Eivissa Channel presented a dominance of motile fauna such as crustaceans and fishes, most likely associated with the shallow depth and the bioclastic sand.

Landslides

On the Eivissa Channel, two small submarine landslides and pockmarks were reported (Lastras *et al.* 2004). We considered them to be mud plains or slopes with escarpments because too old in geological time to presently still affect the community colonization (Lastras *et al.* 2004). Crustaceans and fishes dominated the faunal assemblages of both landslides, corroborating the preference of motile fauna for these types of geomorphologies. In fact, our results agree with those proposed by previous studies employing different sampling strategies (e.g. otter and Agassiz trawls) in these areas, highlighting these groups as the most abundant in terms of biomass (Stefanescu *et al.* 1993, Sardà *et al.* 1994, Abelló *et al.* 2002). Moreover, a high proportion of predators (fishes and cephalopods) were observed in both areas.

Anthropogenic Impact

The Mediterranean Sea has been a human thoroughfare since pre-history time and hosts some of the most ancient coastal settlements along its coastlines, which are currently densely populated (Longhurst 2007). As a result, it has been affected by all types of anthropogenic impacts for a longer time than other seas (Ramirez-Llodra *et al.* 2013). Here, we observed noticeable levels of human impact, not only in relation to commercial fishery activity, but also to littering. We noticed several trawl marks as a proxy of intensive and repetitive fishing activity on canyon walls between 400-700 m. That activity produces a resuspension of sediment, which is mobilized towards deep areas with a potential significant impact on deep-sea communities (Palanques *et al.* 2006, Martín *et al.* 2008). The continuous trawling over the seafloor on the Catalan slope has had a ploughing effect on the seafloor, resulting in a change of the seabed geomorphology and characteristics (Puig *et al.* 2012).

Recent studies in this region reported biodiversity and community composition differences between fished and non-fished areas, with a decrease of sessile species in impacted zones (Ramirez-Llodra *et al.* 2008, 2010). Flat-topped sea hills and seamounts may present a modified faunal composition in relation to a previous undisturbed status (Clark *et al.* 2010), primarily caused by the impact of commercial fishing activity (Pham *et al.* 2014). In the present case, trawl marks were also observed at the top of the flat seamount. In our study area, we observed evidences of a differential fishing activity on both seamounts (not quantified here). There was a high amount of lost longlines (targeting fishes) tangled on the rocky substrate of the Gulf of Valencia seamount. Differently, the flat topped Eivissa Channel seamount presented a higher abundance of trawl marks (targeting mostly decapod crustaceans as the Red shrimp, *Aristeus antennatus* (Risso, 1816) (García Rodríguez & Esteban 2008).

Floating litter was observed in the Eivissa Channel landslides. Plastic bags accumulated in depressions such as pockmarks. This floating litter was also observed in

Blanes canyon, where currents usually transport them from shallower to deeper areas. The impact of marine litter on deep-sea habitats is being addressed by several international initiatives (Galgani *et al.* 2000, Ramirez-Llodra *et al.* 2011, 2013, Pham *et al.* 2014). These studies provide a distribution of marine litter and its potential effects on the habitat and fauna, such as suffocation, physical damage to fragile sessile fauna (e.g. sponges, cold water corals) or the ingestion of microplastics in the NW Mediterranean Sea. Other studies have addressed the chemical contamination on deep-water fauna (Rotllant *et al.* 2006, Koenig *et al.* 2013a, 2013b) and sediments (Abi-Ghanem *et al.* 2011). The presence of lost or discarded fishing nets is also often observed (Vertino *et al.* 2010, Ramirez-Llodra *et al.* 2013), resulting in ghost fishing for long time periods.

Behavioral observations

In this study, schooling behavior of fishes was observed near seamounts, as reported in similar studies in other oceans (Clark 1999). Conversely, on the muddy open slope, isolated individuals were usually detected. The reaction of fishes to the ROV approach varied depending on the species. As a first instance, all avoidance reactions could have been generated by a combination of strong illumination from lamps, water displacement around the ROV and vehicle-generated noise. In relation to the absence of behavioral reaction detected in some species at ROV approach (i.e. *Polyprion americanus*; Atlantic wreckfish), some questions arise about the ecological value of that passivity (Herring *et al.* 1999). Behavioral observations for fishes are becoming abundant as ROV studies increase, since species are well visible, being often the focus of these surveys (Trenkel *et al.* 2004, Davis & Chakrabarty 2011, Ayma *et al.* 2016). Several studies in the Atlantic ocean compared trawl data with ROV video-surveys to evaluate biases produced by both sampling methods (Lorance & Trenkel 2006). These studies showed that fish reaction and response to both ROV lighting and net

approach generates a different bias-dependent effect on observations. In our case, ROV does not seem perceived as a potential threatening stimulus by some species.

We observed 4 individuals of *Paromola cuvieri* as carrying human artifacts, as already reported in other areas (Braga-Henriques *et al.* 2011). This behavior in the Mediterranean populations could be the result of the availability of litter in deep-sea areas. Plastic bag camouflage reported for the genus *Paromola* can be considered as a common behavioral trait for several other species of crabs (Bedini *et al.* 2003), although they usually use gorgonians as camouflage (Wicksten 1985).

We observed seabed aggregation of the pelagic jellyfish *Pelagia noctiluca* according to previous findings (Cartes *et al.* 2013). This species is known to have nycthemeral (alternated water column day and night) migrations (Franqueville 1970), and individuals were observed near the bottom on the top of seamounts, probably in relation to those movements (Boehlert 1988). The presence of *P. noctiluca* in the benthic boundary layer indicates that this species, previously classified as fully pelagic has instead a benthopelagic life habit (i.e. animals enter contact with the seabed sediment once over the 24-h cycle; *sensu* Aguzzi & Company, 2010). Another interpretation could be that our observations were the result of some mass deposition of dead jellyfishes, probably resulting from some sort of schooling on the water column, which could be, potentially, a common behavior in these animals (Billett *et al.* 2006).

The different targeted environments showed faunal composition according to substrate, depth, and topography. This aspect justifies a seascape approach in further ecosystem studies within north-western Mediterranean deep-sea areas. Several canyons, seamounts and landslides with the same characteristics could be classified as seascape units because they share similar compositions and distributions of taxonomic groups. This would

allow faunistic predictions in other presently unexplored but similar western Mediterranean areas.

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For Review Only

747 APPENDICES:

748

749 **Appendix 1.** Number of observations for each species per area (canyon, landslide and
750 seamount).

751

752 **Appendix 2.** Blanes canyon head. Number of faunistic observations plotted by taxonomical
753 group every 100 m for dives 1 and 2 (A and B, respectively).

754

755 **Appendix 3.** Gulf of Valencia seamount. Number of faunistic observations plotted by
756 taxonomical group every 100 m for the seamount rocky area and the surrounding mud plain
757 (A and B, respectively).

758

759 **Appendix 4.** Eivissa Channel seamount. Number of faunistic observations plotted by
760 taxonomical group each 100 m for the flank (A, dive 4) and the flattop (B, dive 5).

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762 **Appendix 5.** Eivissa Channel landslides. Number of faunistic observations plotted by
763 taxonomical group each 100 m. for *Jersi* (A, dive 6) and *Ana* (B, dive 7).

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Table 1. Depth range (m) and surveyed seafloor (km) of the 7 ROV dives conducted in different geomorphological deep-sea zones of the NW Mediterranean.

Dive	Location	Area	Substrate type	Depth Range	Coordinates	Surveyed area
1	Blanes	Canyon	Mud+Sand	70-450	41° 38' N 02° 52' E	2.5
2	Blanes	Canyon	Mud+Sand	60-450	41° 39' N 02° 53' E	0.7
3	Gulf of Valencia	Seamount	Mud+Rock	450-800	39° 30' N 00° 17' E	3.7
4	Eivissa Channel	Seamount	Sand+Rock	280-500	38° 39' N 00° 55' E	0.6
5	Eivissa Channel	Seamount	Sand+Rock	196-250	38° 39' N 00° 55' E	2.2
6	Eivissa Channel	Landslide	Mud+Rock+CoR	575-600	38° 41' N 00° 50' E	2.0
7	Eivissa Channel	Landslide	Mud	650-700	38° 41' N 00° 50' E	2.8

Typologies of observed substrate are indicated as follow: CoR, coral rubble; Mud; Sand and Rock.

Table 2. Number of individuals by class observed at each geomorphological area.

Class	Canyon	Seamount	Landslide	Total
Demospongiae	5	546	5	556
Hydrozoa	1	26	4	31
Anthozoa	248	354	21	623
Scyphozoa	3	176	207	386
Polychaeta	40	29	8	77
Echiuroidea	5	14	3	22
Bivalvia	16	2	0	18
Gastropoda	4	21	3	28
Cephalopoda	0	0	12	12
Malacostraca	204	228	460	892
Rhynchonellata	56	429	2	487
Crinoidea	3	9	3	15
Asteroidea	17	1	0	18
Ophiuroidea	0	18	136	154
Echinoidea	8	4	0	12
Holothuroidea	21	56	8	85
Thaliacea	1	2	2	5
Elasmobranchii	3	2	0	5
Actinopterygii	157	373	578	1108
TOTAL	792	2290	1452	4534

Table 3. Summary of statistical validations for the connections between taxa ordination and environmental variables. Data type determined methods for calculating *p*-values (permutations test and restricted maximum likelihood - REML).

Study case	Environmental variable	Type of data	<i>p</i> -value method	<i>p</i> -value	<i>r</i> ²
1. Taxa in all habitats	Habitat	Factor	Permutations	0.036*	0.11
	Sediment	Factor	Permutations	0.002*	0.24
	Depth	Continuous	REML	<0.001*	0.31
	Anthropogenic impact	Vector	Permutations	0.759	0.01
2. Taxa in the canyon	Sediment	Factor	Permutations	0.788	0.02
	Depth	Continuous	REML	<0.001*	0.02
	Anthropogenic impact	Vector	Permutations	0.211	0.17
3. Taxa in the seamount	Sediment	Factor	Permutations	0.041*	0.33
	Depth	Continuous	REML	<0.001*	0.29
	Anthropogenic impact	Vector	Permutations	0.068	0.54
4. Taxa in the landslide	Sediment	Factor	Permutations	0.26	0.33
	Depth	Continuous	REML	<0.001*	0.46
	Anthropogenic impact	Vector	Permutations	0.078	0.56

* are the significant ($p \leq 0.001$) values.

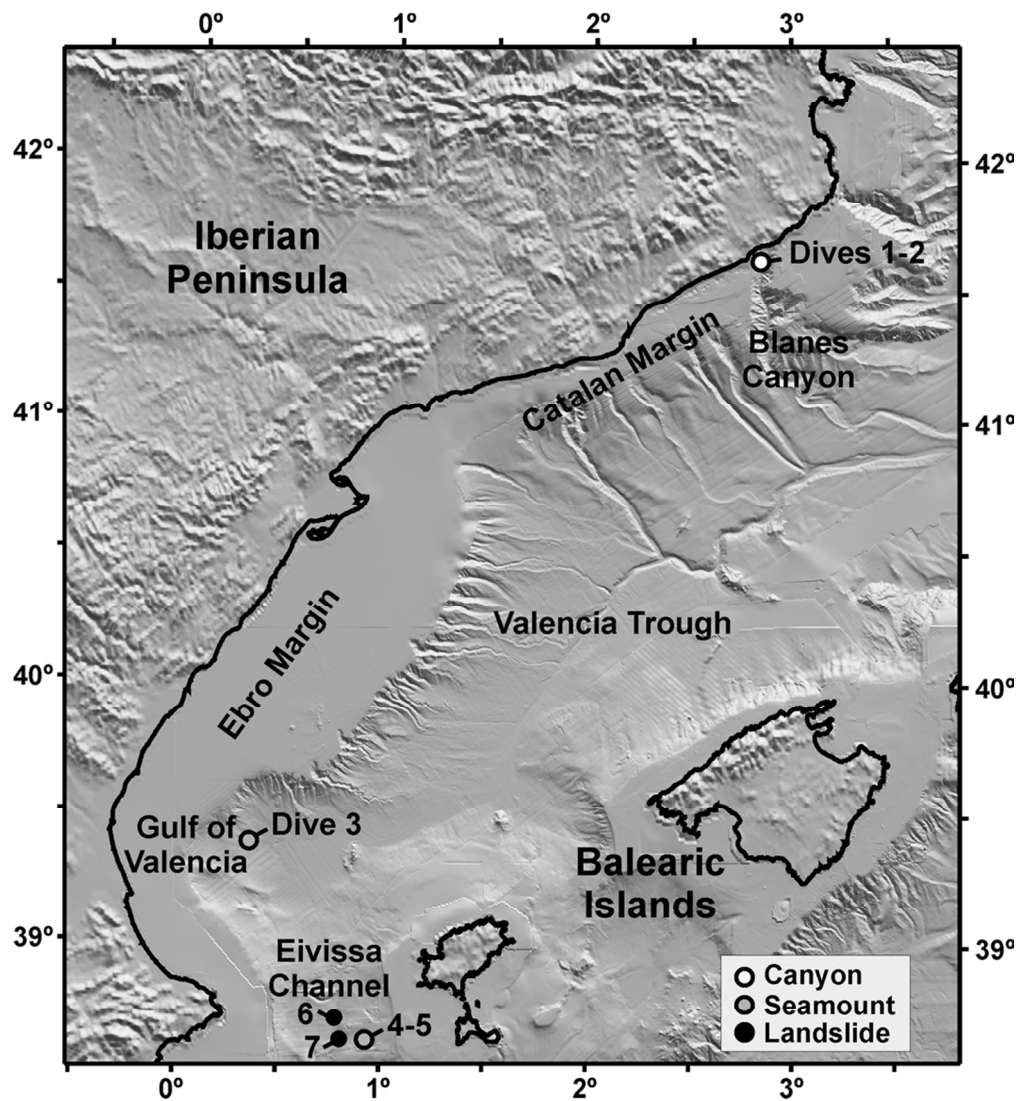


Fig. 1. NW Mediterranean area where ROV dives were conducted. Blanes canyon head in the Catalan continental margin, seamounts in the Gulf of Valencia and Eivissa Channel plus the landslides in the Eivissa Channel.

90x99mm (300 x 300 DPI)

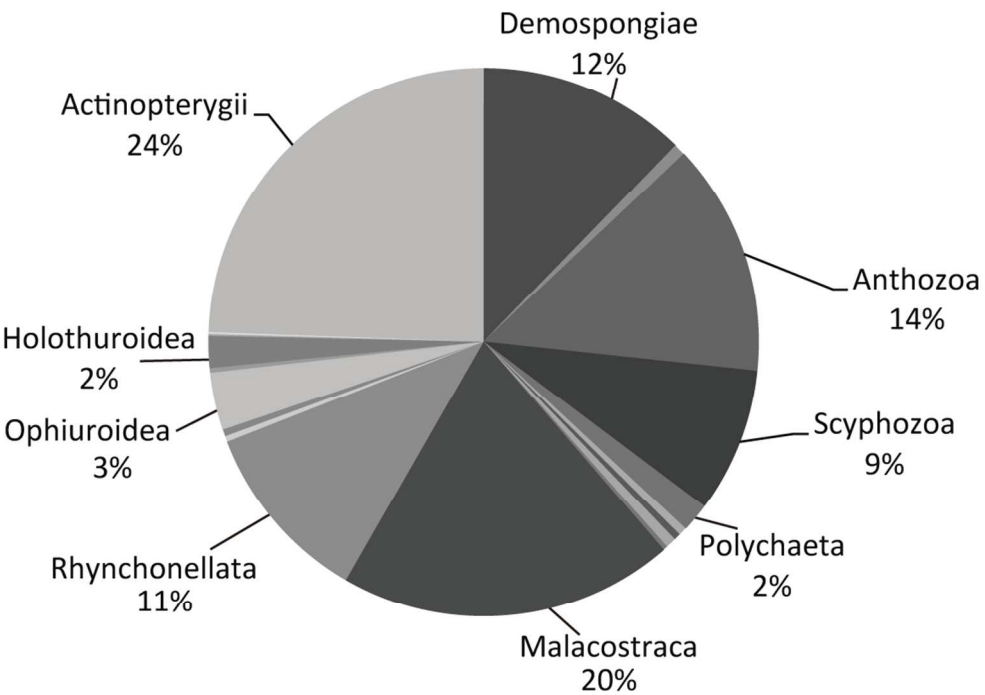


Fig. 2. Percentage per class of total faunistic observations.

99x71mm (300 x 300 DPI)

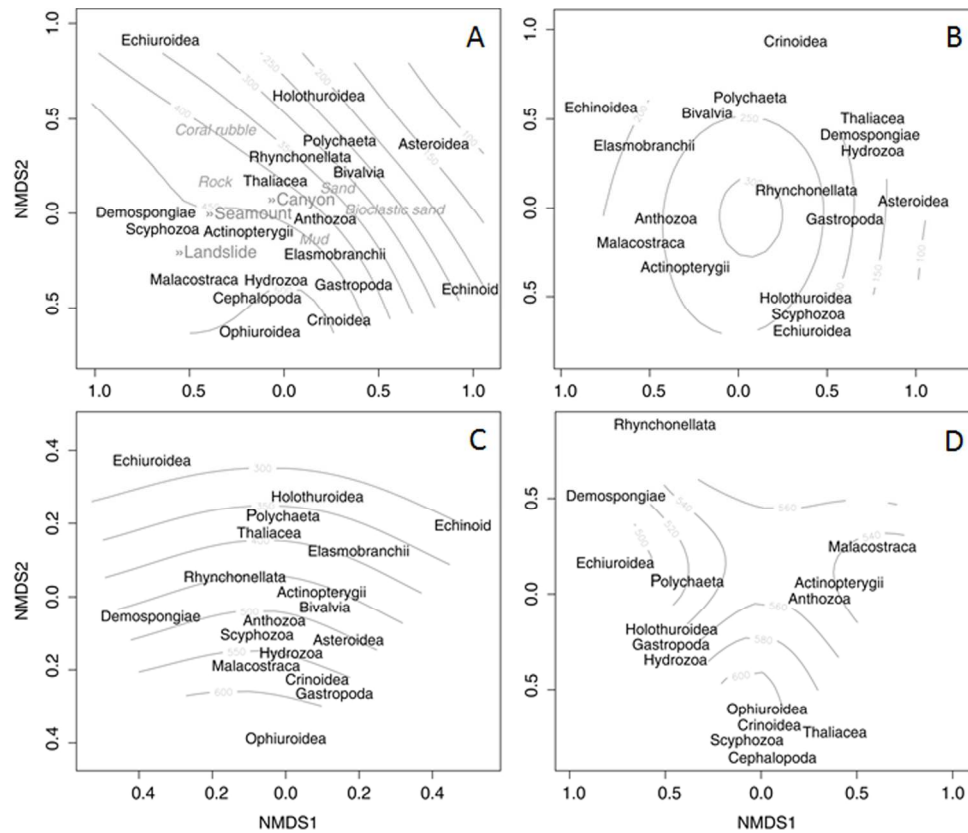


Fig. 3. Spatial ordination of class composition and abundances related to depth (m; grey curves), and sediment types, for (A) all the habitats together; (B) Canyon; (C) Seamount; (D) Landslide.

175x175mm (300 x 300 DPI)

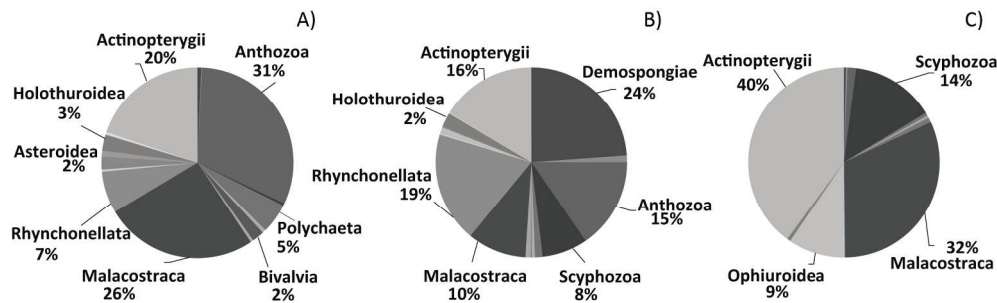


Fig. 4. Percentage of faunal observations at each geomorphological zone studied: (A) Canyon; (B) Seamount; (C) Landslides.

189x60mm (300 x 300 DPI)

Review Only

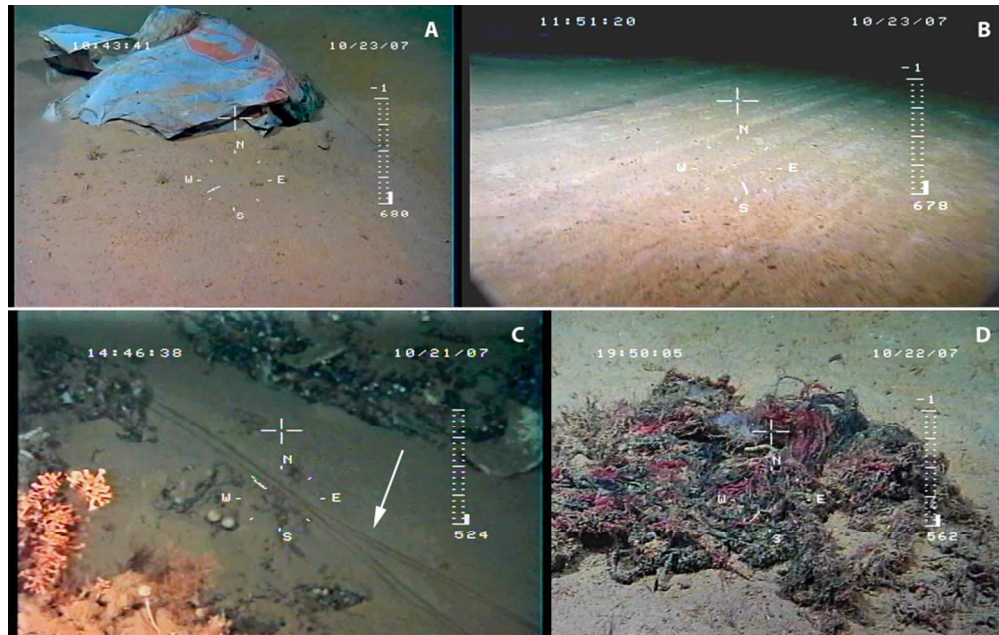


Fig. 5. Different types of anthropogenic impact observed. (A) Litter; (B) Trawl marks; (C) Longlines; (D) Fishing net.

90x56mm (300 x 300 DPI)

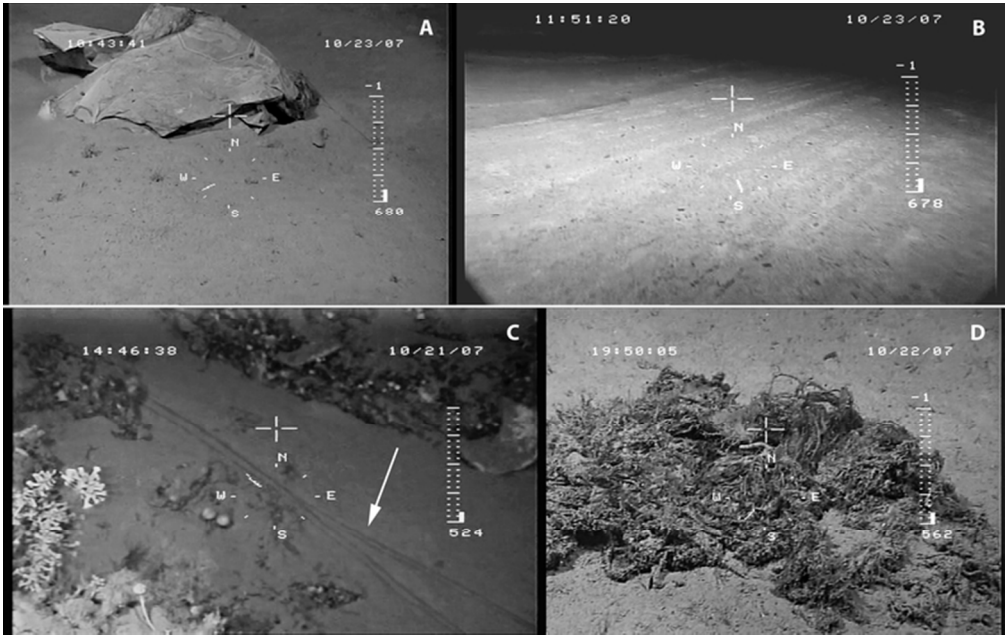


Fig. 5. Different types of anthropogenic impact observed. (A) Litter; (B) Trawl marks; (C) Longlines; (D) Fishing net.

66x42mm (300 x 300 DPI)

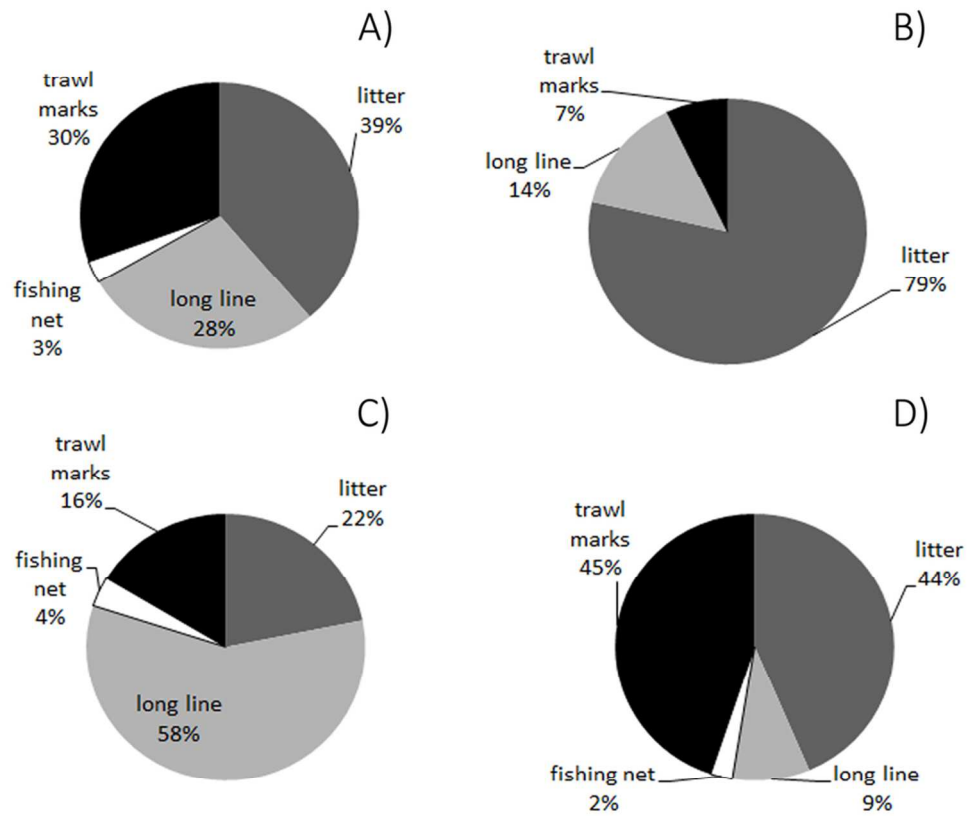


Fig. 6. Percentage of total anthropogenic impact observed in the study and in each area. (A) Total anthropogenic impact detected in all areas; (B) Canyon; (C) Seamount; (D) Landslide.

89x89mm (300 x 300 DPI)

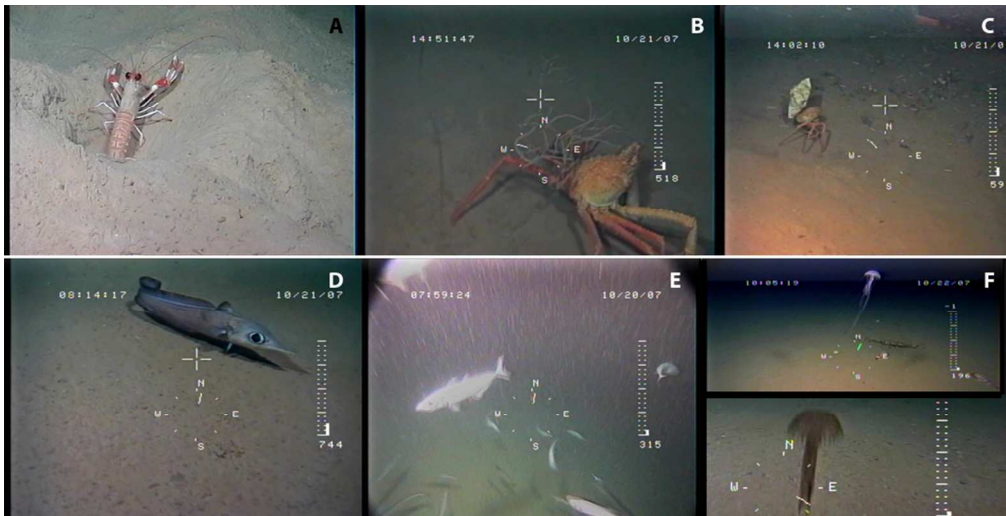


Fig. 7. Behavioural observations. (A) Territorial behaviour, in the Norway lobster, *Nephrops norvegicus*; (B-C) Camouflage behaviour from *Paromola cuvieri*; (D) The Macrourid *Trachyrincus scabrus* (Rafinesque, 1810), just before ROV evasion; (E) Schooling *Trachurus* sp.; (F) *Pelagia noctiluca* close to the bottom.

90x46mm (300 x 300 DPI)

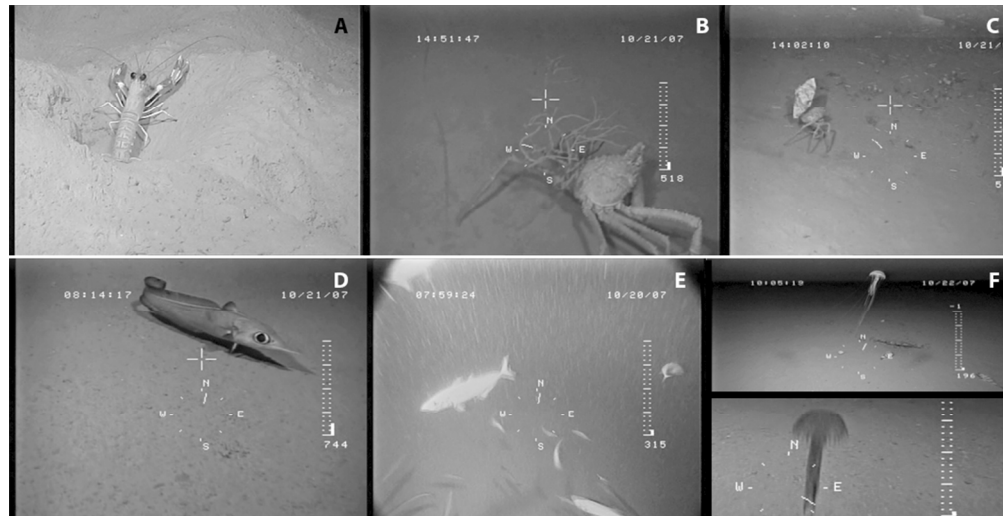


Fig. 7. Behavioural observations. (A) Territorial behaviour, in the Norway lobster, *Nephrops norvegicus*; (B-C) Camouflage behaviour from *Paromola cuvieri*; (D) The Macrourid *Trachyrincus scabrus* (Rafinesque, 1810), just before ROV evasion; (E) Schooling *Trachurus* sp.; (F) *Pelagia noctiluca* close to the bottom.

90x46mm (300 x 300 DPI)