

First *in situ* observations of soft bottom megafauna from the Cascais Canyon head

GOMES-PEREIRA, J.N., I. TOJEIRA, L.P. RIBEIRO, R.S. SANTOS, F. DIAS & F. TEMPERA



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We report the first *in situ* observations of soft bottom megafauna from the Cascais Canyon head. Observations were collected opportunistically during three technical dives with the ROV *Luso* between 460-805 m at two locations distanced 1,230 m. The habitats were classified as upper bathyal fine mud. The soft bottom fauna was dominated by burrows of *Nephrops norvegicus* reaching up to 2.9 burrows/m², a common habitat along the Portuguese continental margin. To our knowledge, densities are the highest ever reported for depths below 300 m. The ichthyofauna at the upper Cascais Canyon is a mixture of lower shelf and upper bathyal species, including *Phycis blennoides*, *Scyliorhynchus canicula*, *Coelorhynchus labiatus/occa* and *Chimaera monstrosa*. Bait release attracted *Myxine glutinosa*. Surveys in other geological settings of the Cascais Canyon are required to understand more comprehensively the diversity of its sessile and vagile biodiversity.

Key words: Atlantic, Canyon, soft bottom megafauna, ichthyofauna, *Nephrops norvegicus*

Gomes-Pereira¹, J.N., I. Tojeira, L.P. Ribeiro, F. Dias, Task Group for the Extension of the Continental Shelf (EMEPC), Rua Costa Pinto 165, Paço de Arcos 2770-047, Portugal. R.S. Santos & F. Tempera ¹MARE – Marine and Environmental Research Centre, IMAR, Department of Oceanography and Fisheries of the University of the Azores, 9901-862 Horta, Azores, Portugal.

INTRODUCTION

Canyons play key roles in geological and biogeochemical processes (Gardner 1989; Arzola et al. 2008). In the last decade there was an increasing acknowledgment of the importance of submarine Canyons for biodiversity (Morell 2007; Tyler et al. 2009). In southwestern Europe, the Portuguese continental shelf and slope are cut by several canyons, with the Nazaré Canyon amongst the largest and most studied in the world (Tyler et al. 2009).

The Cascais canyon is located approximately 120 km to the south of the Nazaré Canyon, cutting into the Portuguese shelf edge at depths of ~175 m. It is the steepest of the Portuguese Canyons and one of the shortest attaining the abyssal plain, with water depths exceeding 2500 m, in a mere 20 km (Lastras et al. 2009). In contrast with

the Nazaré canyon, which has no rivers of significance in its vicinity, the Cascais canyon is located off the Tagus mouth – the longest river of the Iberian Peninsula and the one draining the second largest Iberian basin (e.g. Milliman et al., 1995). This makes the Cascais Canyon more prone to anthropogenic impacts originating both from pollutants collected throughout the Tagus drainage basin and from human activities based on the neighbouring densely populated and industrialized Lisbon metropolitan area.

Despite this proximity to a major northeastern Atlantic European capital (i.e. Lisbon), the megafaunal communities from the Cascais canyon, like other canyons on the Portuguese continental margin, have been poorly studied, with few studies reporting *in-situ* observations of its biological communities.

We report benthic megafaunal observations recorded opportunistically with an ROV during technical operations in the upper reach of the Cascais Canyon, including a description of habitats and a list of invertebrate megafauna and fish fauna.

METHODS

STUDY AREA

The head of the Cascais Canyon is located at 38.507°N; 9.480°W approximately 23 km to the south of the town of Cascais. Its head has a 4.2 km width, a V-shaped morphology, and is incised down to 1350 m (Lastras et al. 2009).

ROV OBSERVATIONS

Three dives with the ROV *Luso* were performed in May 2015 in the Cascais Canyon head with technical objectives related to practicing the deployment and retrieval of equipment on the seafloor. ROV *Luso* is a working class Bathysaurus XL (Calado et al. 2008) from the Task Group for the Extension of Continental Shelf (EMEPC), and was operated from the NRP *Almirante Gago Coutinho* during the EMEPC/PEPC/LUSO/2015. Digital HD video was recorded and used to describe faunal occurrences. Faunal occurrences were described to the lowest feasible taxa (Hayward & Ryland 1995). The camera is an Argus HD-SDI camera, comprising a Argus HD Camera Housing with a Sony FCB-H11 camera (10x optical zoom; focal distance 5.1-51mm). Two parallel scaling lasers distanced ca. 63.5 cm apart projected on the seafloor, were used for measurements and area estimates. The software ImageJ was used for image analysis. The ROV dives were performed in two locations, distanced 1.230 m. Two dives were performed at site A, between 795-805 m (dives L15D01 and L15D03), and one dive in site B, ranging 460-470 m (L15D02) (Table 1, Fig. 1). A total bottom time of 7.5h was recorded.

The imagery collected during the ROV operations on the seafloor was annotated in order to extract scientific megafauna observations. Lists of sessile and mobile invertebrates as well as fishes were compiled for each dive. Habitat description notes were limited due to high turbidity and bottom currents. A video transect with the ROV

front camera in oblique view was executed at Site B to obtain densities of invertebrate megafauna. The ROV travelled at ca. 0.05m/s distanced 0.5 to 1.5 m from the seafloor. Area estimations were calculated using the parallel laser lines projected on the seafloor following (Dias et al. 2015). The video transect was sub-sampled by extracting still images at regular 10-second intervals between 11:07:00 and 11:11:20 (hh:mm:ss). Images of poor quality and with overlapping seafloor sections were excluded. A total of 25 images were analysed, representing on average 0.49 m² (STD=0.11) of seafloor per image.

Densities of *Nephrops norvegicus* were estimated by counting burrows on the seafloor (see Morello et al. 2007 for a review; Aguzzi & Sardà 2008). The burrows were ascribed to *Nephrops norvegicus*, which were observed at the burrow entrances in several occasions (Fig 2b). Other aspects considered were the shape and appearance of burrow openings, the size and angle of tunnels, the geometric relationship between openings, as well as the presence of features such as tracks next to the openings (Chapman & Rice 1971; Atkinson 1974; Chapman 1980; Tuck et al. 1994; Marrs et al. 1996). Generally, each burrow was assumed to be occupied and contain a single animal (Sardà & Aguzzi 2012). However, where the above-mentioned criteria suggested that clusters of openings were related and represented a single burrow system, individual openings were not enumerated and a single burrow was counted. Burrows of uncertain occupancy, in a state of collapse or with partially-blocked openings indicating abandonment were also ignored as it is known that unoccupied burrows rapidly degrade and collapse (Marrs et al., 1996). These criteria render conservative estimates.

In an attempt to record further fish species, fish attraction was stimulated by releasing bait and shutting down all the ROV's light and mechanical systems (dive L15D03; site A). The bait was composed of rotten *Trachurus trachurus* and was held in latex gloves. The bait was released using the ROV arm to rip the glove apart, and the lights were switched off for 30 minutes. The lights were then switched on, a second portion of the bait was released, and all ichthyofauna observed was registered.

In situ observations from Cascais Canyon

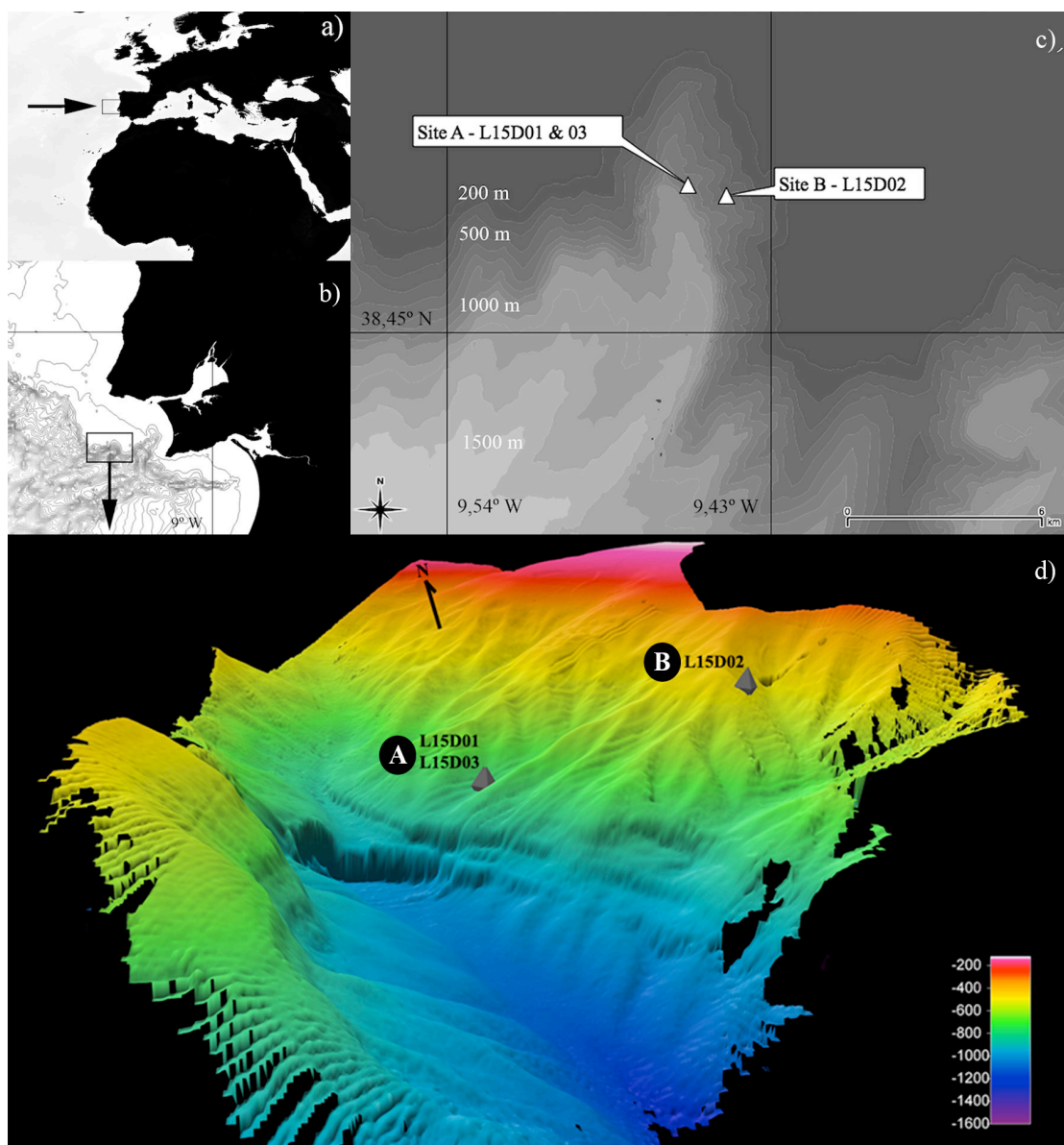


Figure 1. a-b) Cascais Canyon area on the European continental margin, Portuguese Coast; c-d) surveyed sites A and B; (image d) with vertical exaggeration of 2x; source: Estrutura de Missão para a Extensão da Plataforma Continental, EMEPC 2015).

Table 1. Location of Dives in the Cascais Canyon head

Site	DiveCode	Start		Start position		Depth (m)
		Date	Time	Lat;	Long	Min-Max
A	L15D01	25-05-2015	18:54:38	01:57:19	38,470042; 9,468358	795-802
A	L15D03	27-05-2015	09:27:20	01:39:18	38,470184; 9,468489	797-805
B	L15D02	26-05-2015	11:00:38	00:56:03 before bait 02:20:48 after bait	38,469550; 9,454483	461-491

RESULTS

The seafloor at both site A and B was dominated by clay. At the deeper site (site A) the sediment layer reached a thickness of at least 0.5 m (Fig. 2a). High suspension was generated by particulate matter constantly arriving to the seafloor.

Bonellia viridis was the most frequently observed epibenthic sessile species with densities up to 3.63 ind.m⁻² (average 0.224 burrows.m⁻²; SD=0.812). The seafloor was heavily bioturbated, particularly at the shallowest site (site B). Occupied and old burrows of *Nephrops norvegicus* made up a significant part of the major bioturbation signs observed. These reached 2.902 burrows.m⁻² (average 0.826 burrows.m⁻² with SD=1.168).

Henslow's swimming crabs *Polybius henslowii* were observed at the seafloor on both sites (Fig. 2c), as well as during the ROV descent in dive

L15D03. Euphasiacea were attracted to the ROV lights in the three dives, limiting the illumination of the seafloor.

Ichthyofauna records were the most varied, yielding at least eight taxa. All fish were considered to have been attracted to the ROV, as these occurred with the ROV inactive on the seafloor or appeared swimming in the ROV field of view (Lorance & Trenkel 2006). Exception was a one *Schyliorhinus canicula* individual that was observed resting on the seafloor (Fig. 2f).

After bait release, and 30 min. of light absence, the ichthyofaunal observed in the vicinity of the ROV was *S. canicula* and *Myxine glutinosa*. *S. canicula* was observed biting the latex glove with bait remains. *M. glutinosa* entered the ROV front drawer possibly in search or attracted by bait remains.

Table 2. Soft bottom megafauna (number of individuals) observed *in situ* in the Cascais Canyon head (TL – Total Length in cm; * faunal occurrences only after bait release).

Order	Lower taxa	Site A		Site B	TL (cm)
		L15D0	L15D03	L15D02	
Annelidae	<i>Cerianthus</i> sp.	-	-	1	n.a.
Crustacea	<i>Nephrops norvegicus</i>	-	-	2	n.a.
	Decapoda undetermined (shrimp)	-	-	1	n.a.
Echiura	<i>Bonellia viridis</i>	-	-	9	n.a.
Actinopterygii	Anguiliforme undet.	1	3	-	n.a.
	<i>Coelorhynchus occa/labiatu</i> s	1	1	1	40
	<i>Chimaera monstrosa</i>	-	1	1	79.5
	Macrouridae	-	-	1	n.a.
	<i>Myxine glutinosa</i>	-	-	1*	n.a.
	<i>Phycis blennoides</i>	-	-	1	33
	<i>Scyliorhinus canicula</i>	-	-	1+2*	44.5
	<i>Synaphobranchus kaupi</i>	1	-	-	n.a.
	<i>Synaphobranchus</i> sp.	-	1	-	n.a.

DISCUSSION

The soft bottom invertebrate fauna in the Cascais Canyon head is dominated by *Nephrops norvegicus*, inhabiting burrows excavated in the muddy sediment. This is a common habitat in the Portuguese shelf and slope areas between 90 and 800 m depth (Marta-Almeida et al. 2008). The species supports one of the most valuable fisheries from the Northeast Atlantic to the Mediterranean

(ICES 2012) including part of the Portuguese fleet operating on the shelf. It is worth noting that on none of the sites were these burrows associated with seapens as in the OSPAR priority list habitat “Seapens and burrowing megafauna communities” (Commission 2008). It is unclear if the absence of large epibenthic corals results from a lack of natural conditions for the development of these organisms.

A series of works on Mediterranean canyons

In situ observations from Cascais Canyon

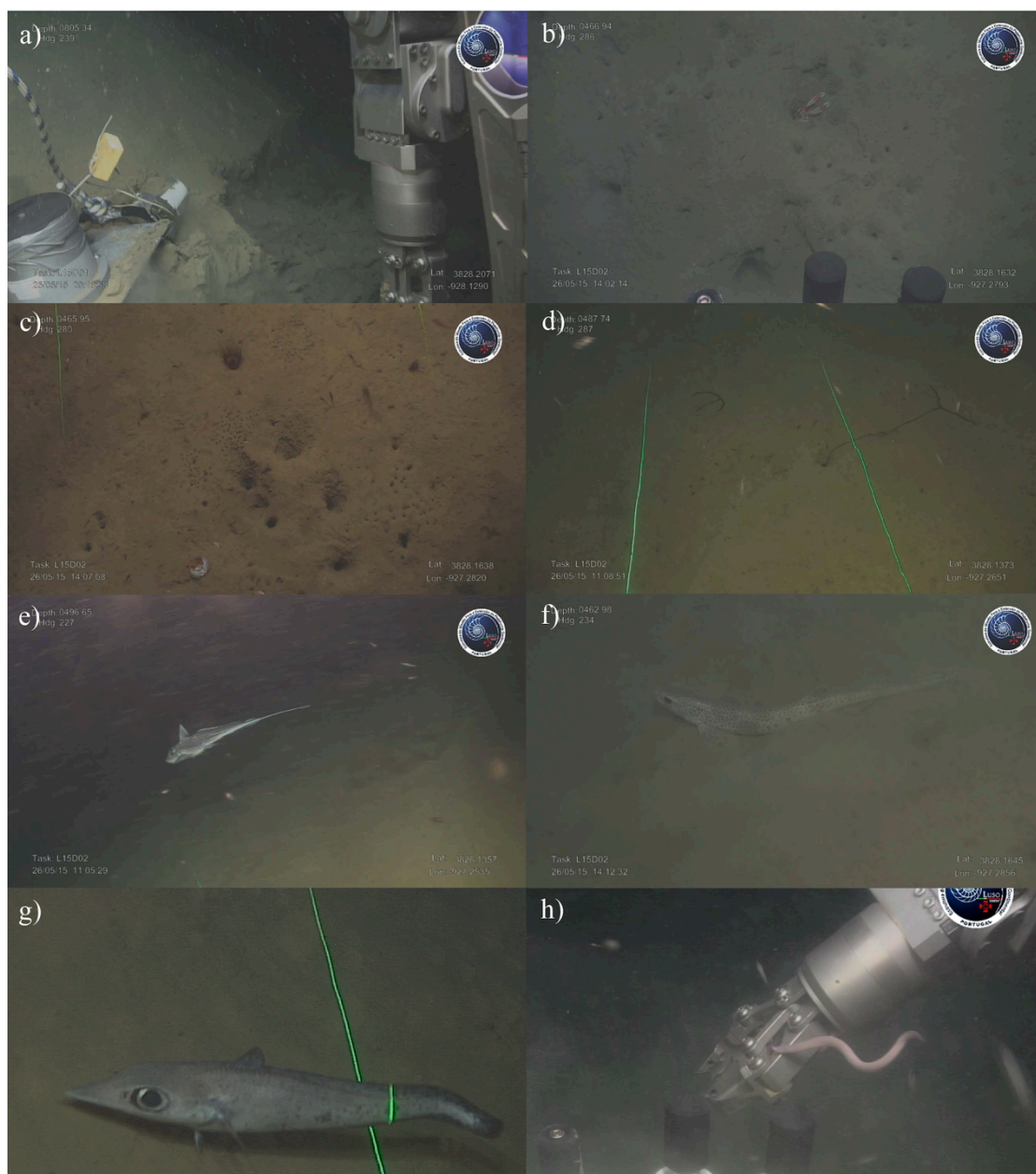


Figure 2. a) Site A, b) site B with *Nephrops norvegicus* at the entrance of a burrow; c) *Cerianthus* sp. (site A); d) two *Bonnelia viridis* (Site A/B), e) *Chimaera monstrosa* (Site B), f) *Scyliorhynchus canicula* (Site B), g) *Coeliorhynchus labiatus*, h) *Myxine glutinosa*.

demonstrated that deep-sea biodiversity and ecosystem functioning may also be impaired by commercial trawling fisheries frequently targeting the shelf areas upstream from submarine canyons and the canyon rims themselves (Palanques 2006;

Martín et al. 2008; Martín 2014). By enhancing the input of re-suspended particles into canyons and eventually triggering sediment gravity flows, such activities extend trawling impacts deeper from the fishing grounds, into bathyal sectors of

deep-sea canyons. From our observations, no trawl marks or litter items were observed at both sites.

The presence of suitable sediment is considered a key factor for *Nephrops* habitat selection and distribution. However the relationship between *Nephrops* burrow density and sediments appears to be non-linear and stock-specific (Campbell et al. 2009). Existing data generally show density declining with increasing depth (see review in Aguzzi et al. 2004). In fact, burrow densities at 460-470 meters depth in the Cascais canyon ($0.826 \text{ burrows.m}^{-2}$) are among the highest reported at such depths. Reported densities below 300 m are generally below $0.2 \text{ burrows.m}^{-2}$ (Smith et al. 2003).

The application of the area estimation method of Dias et al. (2015) represents an alternative suitable approach for surveying grounds of *Nephrops* (Morello et al. 2007). However, a combination of methods may still be most adequate (Sardà & Aguzzi 2012).

The ichthyofauna at the upper Cascais Canyon comprises a mixture of lower shelf and upper bathyal species. The distribution of *S. canicula* extends from shelf habitats down to 400 m (Whitehead et al. 1989), and was only observed at the shallower site. *Phycis blennoides* is also mostly a shelf species, ranging from 150-300 meters and was observed at 467 m. *Coelorhynchus labiatus/occa*, which normally range from 580-2,200 m (Whitehead et al. 1989), were observed at both shallow and deep locations (Fig. 2g). The species was observed in all dives on three consecutive days. In two occasions, *C. labiatus* approached the ROV just as it reached the seafloor, being strongly attracted to the platform. Other species were observed in the two dives, *Chimaera monstrosa* (on both shallow and deep sites; normal depth range 300-500 m), and anguilliform fishes, only present at the deeper site.

Rather sparse information is available on canyon benthic communities (Paterson et al. 2011; Huvenne & Davies 2014). This is true even for macrofaunal communities, the best studied group in the submarine canyons from northeast Atlantic continental margins with data restricted to a few Canyons: Capbreton Canyon (Rallo et al. 1994), Cap-Ferret (Sorbe 1999), Nazaré canyon (Curdia

et al. 2004; Paterson et al. 2011) and the Setúbal canyon (Gage et al. 1995; Lamont et al. 1995; Paterson et al. 2011). In the Cascais Canyon, Paterson et al. (2011) addressed macrofaunal diversity and assemblage structure using polychaetes with conclusions hampered by the small-scale heterogeneity derived from box corers. The megafaunal of canyon upper region has been addressed in other areas, such as in the South West UK (Davies et al. 2014), with bottoms of mud and sand composed mostly of burrowing fauna, including ophiuroids and cerianthids, with resemblances to the continental shelf communities.

This report adds to the growing efforts to understand canyon biodiversity and map marine habitats and associated species. The habitat observed conforms simultaneously to EUNIS level 3 *A6.5 Deep-sea mud* as well as to EUNIS level 4 *A6.81 Canyons, channels, slope failures and slumps on the continental slope* (<http://eunis.eea.europa.eu>). Such double nature is a good illustration of the ambiguity affecting many deep-sea habitats in the current version of EUNIS classification, where the attributed class depends on following the substrate-based path or the geomorphologic one (habitat complexes in section X of the classification).

The soft bottom fauna here reported should extend throughout the Canyon head, as similar geomorphology is expected throughout the canyon head. Some reliefs known to occur in the area where not explored, notably a ~80 m high escarpment crossing the canyon axis at 920 m water depth (Lastras et al., 2009). Different faunal settings are expected to occur as well as certain taxonomic groups not detected in this survey (Tecchio et al. 2013). Further studies should focus on different habitat features and deeper areas of the Cascais Canyon to extend current knowledge on species, habitats and canyon biodiversity.

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