UNIVERSITAT DE BARCELONA

Ecology and biodiversity of the deep-sea meiobenthos from the Blanes Canyon and its adjacent slope (NW Mediterranean)

Ecología y biodiversidad del meiobentos profundo del Cañón de Blanes y su talud adyacente (NO Mediterráneo)

Sara Román Moreno

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Memoria presentada por Sara Román Moreno para optar al grado de Doctor por la Universidad de Barcelona

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A mis padres,

mi hermana y a Rober

<<En una época en que todos consideran el espacio como la última frontera, no debemos olvidarnos de que ahí abajo, en la Tierra, nos quedan montones de cosas por descubrir>> Robert D Dallard.

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¡La Aventura continua!

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1

General Introduction

1.1 DEEP-SEA FLOOR: A glimpse to the environment

The deep-sea is the largest ecosystem on Earth, including about 90% of the seabed (Gage and Tyler, 1991; Ramirez-Llodra et al., 2010b). It is the portion of the ocean beyond continental shelf below 200 m water depth, including both the water column and the seafloor (Gage and Tyler, 1991) (Fig. 1.1). Due to its remoteness and difficulties in observing and sampling, only a 5% of the deep-sea floor was explored until 2010 (Ramirez-Llodra et al., 2010b). Nevertheless, the development of, and easier access to new technologies, such as remote operated and autonomous underwater vehicles (i.e., ROVs and AUVs), fibre optic communications and bottom crawlers and landers, together with the refinement of multibeam acoustics have allowed recognizing a greater habitat complexity and new ecological interactions in deep-sea environments, suggesting that continental margins likely have the most heterogeneous seafloors conditions for its biota.

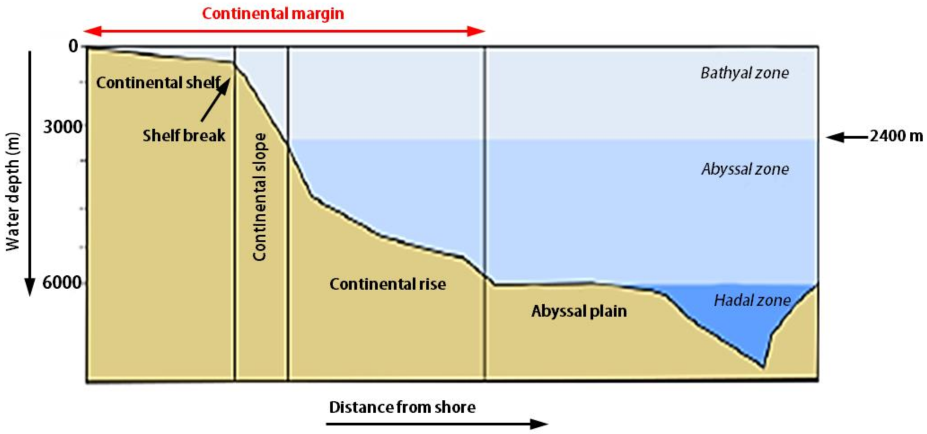


Figure 1.1. Diagrammatic cross-section of the ocean floor showing the physiographic and ecological depth zones. Based on Thistle (2003).

The deep-sea floor is an extreme environment that has been characterized as a physically stable (Sanders, 1968), this giving rise to the image of a supposedly monotonous submarine seascape. At 200 m depth, photosynthesis stops and, at 1,000 m depth, normal sunlight disappears. Without light and under high pressure (increase of 1 atm per 10 m), low temperature (<4°C on average), rather constant salinity and PH, and scarce food inputs (Gage and Tyler, 1991; Thistle, 2003), life as we know seems originally to be impossible. That is why in 1844, the naturalist Edward Forbes postulated his “Azoic Theory” (Forbes, 1844). Indeed, the apparent lack of variability in the physical parameters affecting the deep sea made early ocean scientists to believe that the bathyal was a monotonous cold and dark environment (Thomson, 1873). Nevertheless successive expeditions and, especially, that expedition of the *H.S.M Challenger* around the world (1872-1876) demonstrated that there was life at all depths, from the coast to the abyss. The Forbes's theory of a lifeless deep ocean was disproved and, spectacular bathyal and abyssal life forms were discovered. Since then our knowledge has evolved in parallel to the advances in technology. Indeed, the idea of a biological impoverished deep-sea environment was completely abandoned by the 1960s, and diversity in the deep sea was found to be as great as or possibly even greater than in shallow-water environments (Hessler and Sanders, 1967; Ramirez-Llodra et al., 2010b).

But how do so many organisms thrive in the "harsh" conditions of such an "extreme environment? With the exception of chemosynthetically-driven communities from hydrothermal vents or cold seeps amongst others (Van Dover, 2000; Tunnicliffe et al., 2003), the lack of primary production forces the deep-sea ecosystems to be heterotrophic. The food web depends ultimately on the arrival of organic matter produced in the photic zone by photosynthesis, with only a small fraction of the surface production arriving at the seafloor (Gage, 2003), being subsequently largely controlled by sedimentation and degradation rates in the water column (Fabiano and Danovaro, 1999). The biological debris, i.e. the decaying material that falls from higher in the water column and form aggregates, is also known as *marine snow* (because it looks a little bit like white fluffy bits). Food inputs arrive to the deep seafloor through different pathways (Gage, 2003) (Fig. 1.2), including i) active biological transport by vertical migration of organisms (Sardou et al., 1996; Sutton et al., 2008), ii) passive fall of organic particles of different sizes, from large falls such as animal carcasses and terrestrial plants (Smith and Baco, 2003) to fine particulate matter composed by faecal pellets and phytoplankton (Billet et al., 1983; Lampitt, 1985), and iii) downward lateral advection on continental slope and in canyons (Canals et al., 2006; Tesi et al., 2010). During its fall in the water column, faecal pellets, sand, dust, and particulate organic matter (POM) from dead planktonic animals, and terrestrial input particles, get progressively degraded by bacteria (Azam et al., 1983). *Marine snowflakes* grow as they fall, some reaching several centimetres in diameter. Some flakes fall for weeks before finally reaching the ocean floor. This continuous rain of *marine snow* provides food for many deep-sea creatures (Gage, 2003). Many animals in the deep-sea ocean filter *marine snow* from the water or scavenge it from the seabed.

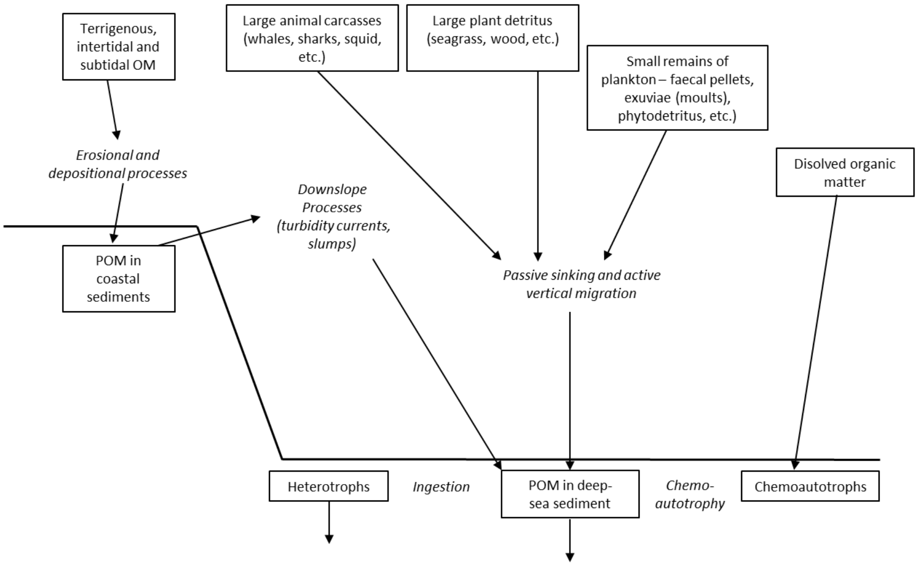


Figure 1.2. Categories of food inputs to the deep-sea bed ecosystem. From Gage (2003).

Once on the seafloor, *marine snow* entrains the benthic food web, being thus essential in regulating how species interact with food sources and between them (Levin et al., 2001; Smith et al., 2008). The small percentage of material not consumed in shallower waters becomes incorporated into the muddy blanketing the ocean floor, where it is further decomposed through biological activity. How organic matter (OM) is distributed when arriving at the seafloor is shaping density, biomass and diversity of benthic species. Physical factors, such as seabed topography, disturbance, boundary constraints, and hydrodynamics (McClain and Schlacher, 2015), as well as the biological factors like bioturbation, population dynamics, and dispersal (Gage et al; 1997) may influence on how (quantity, quality and velocity) this OM reaches and gets distributed along the seafloor.

1.2 CONTINENTAL MARGINS

The most geologically diverse components of the deep seafloor are continental margins, the focus for this thesis. Continental margins mark the transition between the continents and the ocean (~ 200 to ~4,000 m water depth) and represent around the 15% of the seabed (Fig. 1.1). The landward part of the margin, the continental shelf, stops at the shelf break, where a marked increase in the downward bottom gradient indicates the continental slope. On the ocean side, the continental slope is bordered by the continental rise (steeper than the slope), which connects the continental slope with the abyssal plain, the largest part of the deep-sea bottom (~76%, Ramirez-Llodra et al., 2010b).

Continental margins can be divided into active and passive, which mainly differ in morphology attributed to the processes governing their formation. Active margin morphology is controlled by tectonic/magmatic processes, shows a steeper slope, and includes ocean trenches. In turn, erosion and deposition processes control passive margins, which show a wider, less sloppy shelf and include continental rises (Harris and Whiteway, 2011).

Margins can be seen as ocean edges covered by large accumulations of mud and sand. Most often, these mainly consist of terrigenous debris that have been eroded from the nearby continents and shaped by ocean processes into thick sedimentary wedges. Biogenic oozes such as planktonic shell remains increases with distance of shore and mainly appear in the deeper areas. Overall, the 90% of the ocean´s carbon burial occurs in the margins, forming the planet´s largest long term sink for carbon form land and shelf (e.g., carbon converted in petroleum or methane reservoirs). Far from the previous perception of deep continental margins as monotonous mud slopes, relatively recent seafloor mapping and direct observations have revealed a huge heterogeneity, with a mosaic of habitats and ecosystems linked to geomorphological, geochemical and hydrographic features that are certainly influencing the biotic diversity (Levin and Sibuet, 2012 and references therein). In fact, continental margins comprise the most geologically diverse areas of the deep-ocean floor and display a considerable degree of habitat heterogeneity (Levin and Dayton, 2009, Levin et al., 2010, Menot et al., 2010; Levin and Sibuet, 2012) (Fig. 1.3). They encompass geomorphological (e.g., seamounts, canyons, muddy slopes, channels, gullies, pockmarks), hydrographic (e.g., major ocean water masses, cascading flows, internal tides), geochemical (e.g., oxygen minimum zone, gas hydrates, fluid seepage) and biogenic features (e.g., deep water coral reefs, sponge and tubeworm gardens, mussel beds) (Levin and Sibuet, 2012).The high habitat diversity host different faunal communities, thereby contributing to the high biodiversity found on continental margins (Levin et al., 2001; Vanreusel et al., 2010; Levin and Sibuet, 2012).

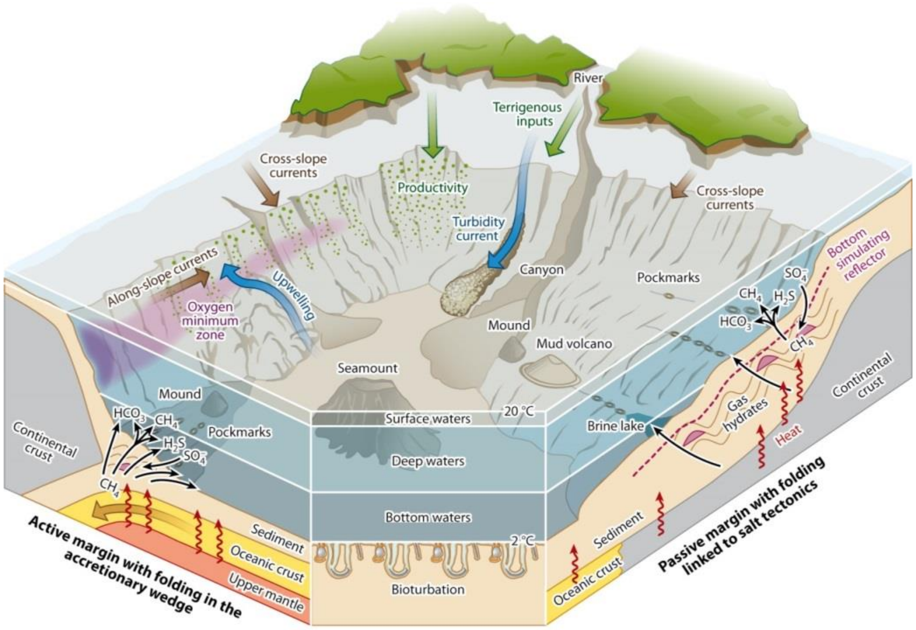


Figure 1.3. Scheme indicating the geological, chemical, and biological sources of habitat heterogeneity on continental margins. Not drawn to scale. From Levin and Sibuet (2012)

1.3 SUBMARINE CANYONS

Submarine canyons are major topographic deep-sea features along continental and island oceanic margins around the world, which connect continental shelves to deep ocean basins (Shepard and Dill, 1966) (Fig. 1.4). The existence of submarine canyons was first reported over 150 years ago by Dana (1863), who described a valley in the Bay of New York. But it was not until the 1960s when the interest on submarine canyons started to increase, leading to the Shepard and Dill (1966)´s detailed description.

Submarine canyons are characterized by flanks with a more or less pronounced slope and a central channel. They may either be shelf-incising, with or without direct connections to rivers, or blind (i.e., wholly confined to the continental slope, below the depth of the shelf break) (Harris et al., 2014; Huang et al. 2014) (Fig. 1.4). A recent worldwide inventory has estimated that there are around 9,477 distinct canyons covering 11.2% of the continental slopes (2% of the total ocean floor); 2,076 of which are shelf-incising, 7,401 blind, with only few of them being more than 2 km deep and hundred km long (Harris et al., 2014). Among them, the present thesis focusses on a shelf-incising canyon.

Canyons are found more frequently on active (44.2%) than on passive margins (38.4%), being, shorter, more dendritic and more closely spaced on the former active than on the latter (Harris and Whiteway, 2011). Despite being considered as widespread and ubiquitous features, there have been relatively few investigations on canyons, compared to other deep-sea environments such as seamounts and hydrothermal vents (Ramirez- Llodra et al. 2010b).

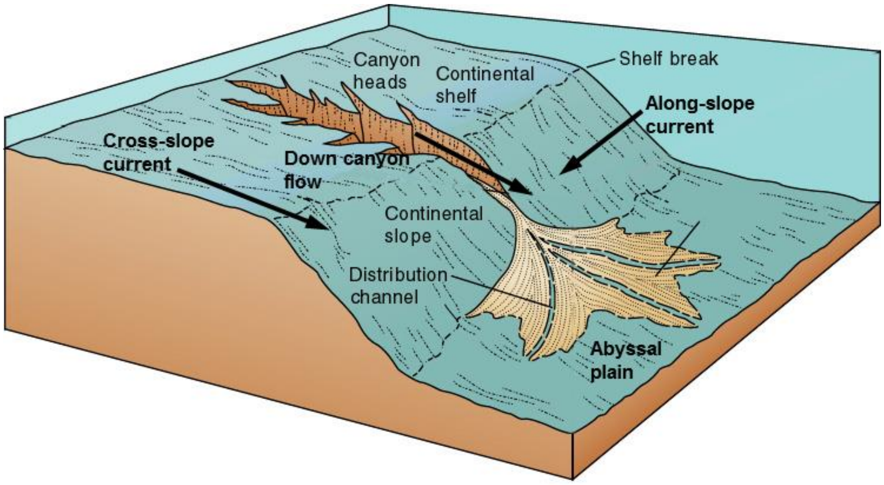


Figure 1.4. Schematic diagram of a submarine canyon.

1.3.1 Ecological role of submarine canyons

Submarine canyons harbour a wide variety of substrate, including mud, sand, hardground, gravel, cobbles, pebbles, boulders, and rocky walls, occurring either separately or in various combinations, which provides a heterogeneous set of habitats (Baker et al., 2011; De Leo et al., 2014). The global distribution and high frequency of canyons cutting the continental slopes supports that they play an important role in the connectivity between the continental shelf and the abyssal plain.

Canyons introduce significant habitat heterogeneity on continental margins due to their extreme topography and the corresponding effects on current regime, substratum types, sedimentation, OM fluxes, and detrital funnelling from the continental shelf (Levin et al., 2001). Active canyons are believed to be very unstable environments. Their abrupt topography influences the hydrodynamic regime by interrupting, accelerating, and redirecting flows, which intensify mixing and amplify currents (Martín et al., 2006), but it also enhances sediment fluxes relative to open slopes (Heussner et al., 2006; Zuñiga et al., 2009).

Current modifications may result in local upwelling, which elevate nutrients to the euphotic zone stimulating primary production (Ryan et al., 2005), but also in developing closed circulation cells and canyon downwelling, which results in an enhanced trap capacity for particles transported by long-shore currents (Granata et al., 1999; Palanques et al., 2005; Allen and Durrieu de Madron, 2009). Thus, canyons play a major role as conduits or transport pathways between shelf and deep ocean environments by trapping, accumulating, concentrating and funnelling sediments, OM and nutrients (Puig et al., 2014 and references therein) (Fig.1.4). Moreover, they also act as morphological shortcuts, accelerating the transit of particles from the productive coastal zone and inner shelf environments toward the deep sea (Allen and Durrieu de Madron, 2009; Puig et al., 2014). All in all canyons play also a major role as sedimentary deposit area (Epping et al., 2002; Masson et al., 2010).

Shelf matters are transported either through canyons´ main axis or through lateral contributions, the later increasing mass transport with depth. Down canyon transport may occur through bottom nepheloid layers, gravity flows and/or turbidity currents. However, it is not constant and unidirectional and there may be periods of active transport and resuspension, alternating with passive intervals in which sediments accumulate at the bottom of the central channel. Furthermore, canyons´ presence amplifies the effect of external environmental forces, such as large storm waves, hyperpycnal flows, dense shelf water cascades (DSWC: will be introduced in more detail further in **Chapter 2**), and earthquakes, among others, which trigger mass failures of unstable deposits within canyon heads and on the shelf-edge areas of shelf-incising canyons (Canals et al., 2006; De Stigter et al., 2007; Puig et al., 2014 and references therein). Overall, particle fluxes and sediment accumulation rates have been found to be much larger inside submarine canyons than in the adjacent non-dissected margin at comparable depths (Martin et al., 2006; Zuñiga et al., 2009; Lopez-Fernandez et al., 2013).

This is particularly evident for shoreline-close canyons, which are directly influenced by river inputs, severe coastal storms or dense shelf water cascading

(Canals et al., 2006; Tesi et al., 2010; Pedrosa-Pàmies et al., 2013). The variation in the frequency of these events, as well as the pulses of materials and energy, highly influence the structure and functions of the benthic assemblages (Cunha et al., 2011; Duros et al., 2011; Paterson et al., 2011; Pusceddu et al., 2013; Ramalho et al., 2014).

Subsequent consequences derived from canyons´ role as conduits for sediment and OM transport to the deep-sea, include carbon storage regulation (Canals et al., 2006; Masson et al., 2010) through carbon burying from surface to deep sediment layers. Hence, they may also play a major role in climate regulation. Also, they may act as larval settlement and recruitment, nursery, and refuge grounds for demersal and benthic organisms like fish and crustaceans (Sardà et al., 1994b; Fernández-Arcaya et al., 2013), in which the higher food availability and attractiveness of habitat structures (i.e., rocky walls, boulders and detritus patches) play a major role (Stefanescu et al., 1994, Vetter and Dayton, 1999; Fernandez-Arcaya et al., 2013). Faunal concentration is also favoured by motile organisms leaving the adjacent slope to concentrate in the nearby canyon, in an attempt to evade visual predators by hiding within the complex canyon topography (Farrugio, 2012).

In short, canyons play an important ecological role, where topography, oceanic currents and substrate heterogeneity have intense consequences for the diversity, functioning, and dynamics of both pelagic and benthic communities.

1.3.2 Human activities impacting canyon ecosystems

An additional consequence of their particular characteristics is that submarine canyons often provide ecosystem goods and services sustainable supporting human wellbeing (Thurber et al., 2014). Consequently, they are also subjected to different anthropogenic stressors involving, for instance, fisheries, oil and gas exploration and exploitation, and pollution. The enhanced conditions for marine life in and around submarine canyons often results in converting these ecosystems in important areas for commercial pelagic and demersal fisheries (Würtz 2012), including highly destructive practices like bottom trawling (e.g., Company et al., 2008; Martin et al., 2008; Puig et al., 2012), with those above 1500 m water depth being particularly more vulnerable destructive fishing practices (bottom trawling). Physical disruption of habitat by trawling, overexploitation, and fish removal as bycatch, coupled with slow growth and reproduction rates by target species, have led to systematic depletion of deep-sea fish populations (e.g., Smith et al., 2008).

Fisheries, and particularly bottom trawling has induced both direct and indirect effects on canyon ecosystem functioning, which will be introduced in more detail further in this introductory chapter.

1.4 MEIOFAUNA: SIZE MATTERS

The deep-sea benthic systems, once considered azoic (Forbes, 1844), are now known to support an abundant and complex life (e.g. Hessler and Sanders, 1967; Sanders, 1968). Deep-sea fauna appear to be broadly distributed across the seafloor, even in highly patchy and isolated environments, indicating the presence of numerous cosmopolitan taxa across most habitats (e.g. Vanreusel et al., 2010; McClain and Hardy, 2010). Exceptions occur, such as in methane seeps and large food-falls, which usually sustain a more restricted range of faunal diversity (McClain and Hardy, 2010). Deep-sea is believed to be mostly shaped by the same factors as shallow waters: resource availability, sediment heterogeneity, hydrodynamic regimes, oxygen availability, and disturbance. But the effect of these environmental drivers depends on the studied organism, as well as on the approached spatial scale.

Meiofauna (i.e., the meiobenthos) is an important component of virtually all benthic ecosystems from fresh water to marine (Thiel, 1975; Vincx et al., 1994). The study of the small organism called meiofauna started since the early days of microscopy (i.e., the aplacophoran Chaetoderma by Lovén (1844) in Giere, 2009), before the term "meiofauna (meiobenthos)" was coined for this group by Molly M. Mare (1942).

The term meiofauna refers to the organisms size (i.e., larger than microfauna but smaller than macrofauna) rather than its taxonomy. Meiofauna include both benthic Protozoa (e.g., foraminiferans) and metazoans, although in this thesis we have not considered the protist component. The meiofaunal size boundaries were arbitrarily defined based on the standardised sieve widths, with 500 - 1000 μm and 44 - 63 μm as upper and lower limits, respectively (Higgins and Thiel, 1988; Giere, 2009). In deep-sea studies, the upper and lower limits are most often 1000 μm and 32 μm, which retain quantitatively sound samples even for the smallest meiofaunal organisms, especially nematodes (Soltwedel, 2000; Leduc et al., 2010).

Meiofaunal organisms include the most phylogenetic diverse fauna on Earth. Almost the entire 35 modern animal Phyla are represented, while Gastrotricha, Gnatostomulida, Kinoryncha, Rotifera, Loricifera and Tardigrada are considered exclusively meiobenthonics taxa. The organism can be *permanent* (i.e., whole cycle a meiofauna) or *temporal* (only larvae or juveniles occur in the meiofauna).

Despite them being invisible to the human naked eye and being just comprised as a methodological definition, the meiofaunal organisms have in fact a major ecological relevance and play an important role in benthic ecosystems. Meiofauna contribute to sediment stability (e.g., increase sediment permeability, cohesion and pore space, grazing and secretion of EPS*, bioturbation and construction of mucus line burrows); nutrient cycling (e.g., their secretions and excretions provide matrix and inorganic nutrient which are easily metabolized by microorganisms, stimulating their activity and growth); waste breakdown and removal and, food webs dynamics (e.g., contribution to carbon processing through ingestion, respiration, defecation and production as a prey for secondary producers). Indeed, meiofauna can provide up to 80% of the diet of predators; being essential for juvenile fish (e.g., scald fish and dab) and omnivore meiofauna (Schratzberger and Ingels, 2017, see references therein). Also they well represent the biochemical conditions: being small-sized, meiofaunal organisms may penetrate proportionally deeper into the sediment than other benthic organism, so that they can trace the environmental variability by shifting in species composition along the sediment profile.

1.4.1 Deep-sea meiofauna

Metazoan meiofauna is the most abundant infauna in deep-sea sediments, with nematodes being particularly dominant (Giere, 2009; Heip et al., 1985; Vanreusel et al., 2010; Pape et al., 2013a, b; Rex et al., 2006). The ecology of deep-sea meiofauna beyond the continental shelf (up to 567 m depth) was studied for the first time by Wigley and McIntyre (1964), being carried out by using a semi-quantitative anchor dredge and an epibenthic sledge. Instrumental limitations account for the relatively late appearance of dedicated studies. Since then, however, the development of suitable sampling gears (mainly based on combinations of corers) has made the deep-sea floor more accessible, enabling quantitative sampling of deep-sea meiobenthos and leading to substantial increases in our knowledge on these organisms in a variety of habitats (Vincx et al., 1994).

***EPS:** Extracellular polymeric substance is a kind of mucus secreted during the construction of burrows.

The quantitative composition of deep-sea metazoan meiofaunal assemblages is relatively stable. Both in poor and rich areas, free-living nematodes dominate the meiofaunal component of the benthos in terms of abundance (80-95%) and biomass (50-90%), when the Foraminifera are excluded (e.g. Heip et al., 1985; Giere, 2009), and their importance often increases with water depth (Giere, 2009). The next most abundant meiofaunal group is the harpacticoid copepods, followed by nauplii larvae and polychaetes. Other groups such as kinorhynchs and tardigrades, etc., occur in much lower numbers (Giere, 2009).

Despite the overall decrease of faunal abundance and biomass with the increasing water depth, the meiofauna tends to increase in importance along the bathymetric gradient compared to macro- and megafauna (Rex et al., 2006), which led these organisms to be the most abundant metazoans in some abyssal environments.

Density declines along the vertical sediment profile are probably the most pervasive gradient observed in marine sediments (e.g., Ingels et al., 2009; Vanaverbeke et al., 1997). Both density and diversity are typically higher in surface sediments and lower in deeper ones, where nematodes become dominant (e.g., Danovaro et al., 2002). At the same time, the most abundant and diverse surface communities are also the most exposed, being thus more influenced by physical disturbances.

Deep-sea meiofauna also displays a remarkable degree of biological and ecological adaptation to the prevailing conditions (e.g. limited and often seasonal food supply, low constant temperatures, etc.). Their life strategies may be controlled by the need for energy conservation, which could translate into slow growth, long life span with low maintenance expenditure, and a reduction of average body size (Giere, 2009). Hence, various contrasting environmental conditions are expected to impact the structure and function of this benthic group more clearly in deep than in shallow waters.

Due to their small size, meiofaunal are dependent on the biochemical characteristics of the sediment (i.e. composition and oxygen availability) (Ingels and Vanreusel, 2013; Leduc et al., 2012c; Tyler, 1995; Vanreusel et al., 1995), food input from the surface (Smith et al., 2008), small-scale habitat heterogeneity, and biotic interactions (i.e., bioturbation and predation), which are the factors believed to be the most important drivers of meiofaunal community attributes (Levin et al., 2001).

In the case of deep-sea meiofauna, the negative correlation between meiofauna abundance and depth is mostly related to the decreasing OM supply (Thiel, 1983; Tietjen, 1992; Rex et al., 2006; Tselepides et al., 2004). However, specific local hydrographical characteristics or topographical features may also play an important role in structuring the meiobenthic communities, with the latter being particularly relevant in the highly heterogeneous submarine canyons. Deep-sea meiofauna are probably dispersal though passively via bottom currents (Thistle et al., 1985) as, due to their size, active dispersal over large distances could likely be discarded.

1.4.2 Canyons´ meiofauna

Habitat heterogeneity (both in terms of hydrodynamic and geology) certainly influences meiofaunal distribution and biodiversity (e.g., Danovaro et al., 1999; Gambi and Danovaro, 2006; Ingels et al., 2009; Vanreusel et al., 2010) and this is particularly relevant in the case of canyons. This feature, combined with an enhanced food supply (i.e., through the accumulation of organic matter caused by the canyon´s physical and geological characteristics), has led to the idea of submarine canyons as hotspot of biomass and density (Levin and Dayton, 2009; De Leo et al., 2010; McClain and Barry, 2010) compared with the surrounding slope and abyssal plains. They are also believed to be sites of enhanced species turnover (beta diversity), leading to overall increases in regional diversity on continental margins for megafauna, especially fish (Vetter et al., 2010) macrofauna, particularly polychaetes (De Leo et al., 2014) and meiofauna, mainly nematodes (Ingels et al., 2009).

Meiofaunal abundance and biomass in canyon systems tends to be higher than on the adjacent slopes (Leduc et al., 2014; De Bovèe et al., 1990; Soetaert et al., 1991a; Ingels et al., 2009; Rosli et al., 2016), while biodiversity tends to be lower (e.g., meiofauna higher taxa an nematode genus/species) (Leduc et al., 2014; Ingels et al, 2009; Garcia et al., 2007), mainly due to the higher food concentration in canyons. However, some canyons did not show consistent changes in standing stocks and diversity between habitats (e.g., Soltwedel et al., 2005; Bianchelli et al., 2010; Danovaro et al., 2009; Gambi et al., 2010) or, even, have standing stocks being lower than the adjacent slopes (Flach, 2002; Garcia et al., 2007; Van Gaever et al., 2009). Low to moderate increases in productivity are thought to increase diversity

(Rosenzweig, 1995), while organic material funnelling may favour opportunistic species (Paterson et al. 2011; Ingels et al., 2009) and thereby act to depress diversity if levels are too high (Curdia et al. 2004; Ingels et al., 2009; Leduc et al., 2014). Moreover, the distribution may differ within the same canyon as well as between the canyon branches, due to large differences in sediment composition (including quantity and quality of food resources and biogeochemical composition) (Bianchelli et al., 2008). So, among the growing knowledge, there are also "contrasting" results dealing with different paradigms on the biodiversity, trophic conditions and functions of deep-sea submarine canyons (Danovaro et al., 2009; Tyler et al., 2009; Bianchelli et al., 2010; De Leo et al., 2010).
The drivers most frequently invoked to affect meiofauna standing stocks and biodiversity patterns are: OM supply, physical energy/levels of disturbance (e.g., current regimens and/or catastrophic disturbances), sediment loading and sediment bioturbation by macro- and megafauna (**Appendix 1**). However, they may act in different combinations and may be superimposed onto other local or regional conditions, causing unpredictable biotic responses (Levin et al., 2001).

One of the most important drivers affecting meiofauna patterns in canyons are OM inputs (sources, quality, quantity and bioavailability). This factor is mostly related to coastal detrital inputs (e.g., by river discharges) or pelagic productivity regimes (e.g., upwelling), as well as to the distance from shore. Organic enrichment often favours increasing infaunal density and biomass, and this is also valid for submarine canyons (Soetaert et al, 1991a; Vetter and Dayton, 1998, 1999). Moreover, it is also related with a decrease in oxygen availability that creates reduced conditions and, thus, may contribute to modulate the structure and composition of the sediment communities. Grain size has also been considered as an important parameter (Etter and Grassle, 1992), since it directly or indirectly reflects the local physical energy and sedimentation patterns, in addition to the local biochemical conditions. In turn, the physical disturbance regime, which generally includes high bottom currents on head or upper canyon regions, may impact the benthic communities. Accordingly, only specialised species may be capable to colonize and survive in these harsh conditions, but also the whole benthic communities may experience periodic cycles of disturbance, giving rise to recurrent defaunation processes, followed by recolonization and eventual community recovering (Company et al., 2008; Lins et al., 2013; McClain and Schlacher, 2015).

In short, meiofauna is intimately related to the sediment, the place where they live. Its physical properties will influence the type of meiofauna harbored by a given ecosystem, in a similar way as the currents are influencing the organization of the marine sediments (hydrodynamic effects) (Soyer, 1985). These factors are combined in submarine canyons in a way that certainly increases its interest, so that understanding the mechanisms controlling deep-sea biodiversity and distribution within and across these complex and at the same time rich environments, crucial to the functioning of the Global Ecosystem, will open new perspectives for their conservation and sustainable management.

1.4.3 Meiofauna taxa

The present thesis focuses on metazoan meiofauna. However, it's the high dominance and diversity of nematodes within the deep-sea meiofauna that lead to address particularly this taxon. On the other hand, kinorhynchs were also the subject of a dedicated study. The main characteristics, as well as the reasons explaining the protagonist role in the present thesis, of these two particular taxa is explained in the following sections.

1.4.3.1 Deep-sea nematodes

With ca. 700 deep-sea described species (Miljutin et al. (2010), free-living nematodes are the most abundant (Giere, 2009) and arguably the most diverse (Lambshead and Boucher, 2003) metazoan meiofaunal taxon of deep-sea benthos and their relative abundance often increases with water depth (Giere, 2009). They appear in almost all seafloor associated environments and show wide trophic range and flexibility (Giere, 2009; Vanreusel et al., 2010). In other words they occur everywhere. Even in relatively impoverished habitats, they may roughly reach densities of 10^5 ind · m^2, while in more productive habitats they may exceptionally reach 10^8 ind · m^2 (Lambshead and Boucher, 2003). Moreover, their ability to survive in some of the most extreme environments found on our planet is unmet by any other marine benthic metazoan taxon. Nematode abundance and diversity appear to be intrinsically correlated with food supply, sediment composition and habitat heterogeneity (Giere, 2009). Thus the study of their feeding ecology and role in the benthic food web and carbon flow may provide relevant information on the environments they inhabit. The abundance, community composition and diversity of nematode genera may vary considerably among habitats, along the sediment profile, and at different spatial scales (see Heip et al., 1985; Ingels and Vanreusel, 2013 Moens et al., 2014). Depending on the sampling scale, patterns in nematode communities can be linked to different ecological processes since these are scale dependent too. In other words, processes structuring nematodes communities at smaller scales are not necessarily identical to those operating at larger scales (Danovaro et al., 2013; Fonseca et al., 2010). In fact, small-scale (mm-cm) variability may be as high as at larger spatial scales, owing to local variations in microtopography, oxygen availability, physical structure of the habitat, patchy distribution of food sources, and biogeochemical characteristics of the sediments, as well as to interactions with other benthic organisms (Fonseca et al., 2010; Gallucci et al., 2009; Ingels et al., 2009). On a larger scale, nematode distribution patterns have been associated with differences in physical parameters (e.g. water mass characteristics, bottom currents), productivity regimes and habitat heterogeneity (e.g., Bianchelli et al., 2013; Lins et al., 2014; Moens et al., 2014; Vanreusel et al., 2010).

Marine free-living nematodes do not possess the planktotrophic larvae or resting stages that characterize other benthic invertebrates, but instead develop through four different moulting stages (Decraemer et al., 2013). A few nematodes show active swimming abilities, but most of them are deemed poor swimmers (Moens et al., 2014). Thus, their dispersion is considered to be a passive rather than active process where the hydrodynamic processes play a key role. In terms of dispersal capacity, its mainly endobenthic lifestyle has important consequences in unstable and heterogeneous environments such as submarine canyons.

In short, due to their occurrence in large numbers almost everywhere in the deep sea, and their high number of species, they are often considered as a useful tool for investigating the structure and diversity of the benthic compartment, as it is expected that the structure of their communities will reflect the habitat variability, disturbance and food availability present in a given ecosystem, including submarine canyons (Giere, 2009; Vanreusel et al., 2010; Ramalho et al., 2014).

1.4.3.2 Kinorhynchs

Kinorhynchs, known also as mud dragons, are an exclusively meiobenthic phylum of marine invertebrates (Higgins and Thiel, 1988; Kristensen and Higgins, 1991), known to occur from polar to tropical regions and from intertidal to abyssal depths. The deepest record was at 7,800 m depth in the Atacama Trench, South Pacific Ocean (Danovaro et al., 2002). Despite they are known to be highly diverse in the deep sea (Neuhaus and Blasche, 2006), they are usually reported at phylum level in ecological papers, or they are simply considered among the "rare" meiofauna (i.e., taxa representing < 1 – 2 % of the total deep-sea meiofauna in a given study). This certainly results in a loss of valuable information (e.g. Coull et al. 1977).

Even though the amount of available information on mud dragon diversity and distribution is gradually increasing, the biogeographical patterns are still highly incomplete and biased, usually reflecting sampling effort rather than real distributions. Also, the species distribution trend at a local scale and their relationships with the abiotic factors is still poorly known being this information practically inexistent for deep-sea environments. Most studies specifically focusing on mud dragons are basically taxonomic papers that either describe new taxa or report the presence of known ones in previously unknown regions. Among them, the studies focusing on the Mediterranean Iberian coast revealed Blanes Bay as a hot spot of kinorhynch diversity (Sánchez et al., 2012) which lead us to focus also on this particular taxa to assess whether this basically shallow-water statement could be also extended to the deep sea off Blanes and, particularly, to the Blanes Canyons system.

1.5 BACKGROUND

During the past ten years, the number of national and international projects focusing on submarine canyons and, in consonance, including studies on benthic communities, has increased substantially (e.g., HERMES, HERMIONE, INDEMARES, RECS, BIOFUN, PROMETEO, DOSMARES). Considerable funding is being invested in research dedicated to increase the understanding of deep-sea canyon system environments and their ecological functioning, in parallel with the increasing availability of new sampling and surveying technologies, leading to a significant increase of canyon-dedicated research. Among the 2,479 publications recorded to date, 1,453 dealt with geology, 1,115 with oceanography, and 1,011 with fauna (Web of Science, search term: submarine canyon). Moreover, technical efforts have not only been addressed to research. In fact, cabled observatories, such as the Ocean Network Canada´s in Barkley Canyon, are currently providing a window into the deep-sea for the general public by offering online, real time views of the canyon seabed (www.oceannetworks.ca). Nevertheless, the number of studies addressing to canyon meiobenthos during the same period hardy reached 50.

1.5.1 Deep sea Mediterranean

The present thesis has been based in the Mediterranean Sea, the largest semi-enclosed sea in the world (Tyler, 2003; Sardà et al., 2004) (Fig. 1.5). The Mediterranean spreads over 4,000 km (< 1% of the world ocean surface), with an average width of only 700 km and an average depth of 1,500 m, its maximum depth reaches 5,267 m at the Calypso trench in the Ionian Sea. It is divided into two sub-basins (western and eastern) of nearly equal size, which are separated by the strait of Sicily (400 m depth). The western sub-basin connects with the Atlantic Ocean by the Gibraltar Strait, while the eastern one connects to the Black Sea thought the Bosphorous Strait, and with the Red Sea by the Suez Channel. As a consequence of the configuration of the surrounding continents and its small extension, the Mediterranean, does not have large systems of outcrops or tides. The Mediterranean Sea displays complex thermohaline circulation patterns driven by the excess of evaporation linked to the cold and dry winds and in relation to the fresh water inflows by precipitation and fluvial runoff (Bergamasco and Melanotte-Rizzoli, 2010). The water exchange of this semi-closed sea is mediated by the Atlantic inflow of surface water (in the upper part of the Straits of Gibraltar; 300 m in depth) and the outflow of the deep, high-salinity, Mediterranean waters (in the deeper parts of the straits; Béthoux et al., 2002).

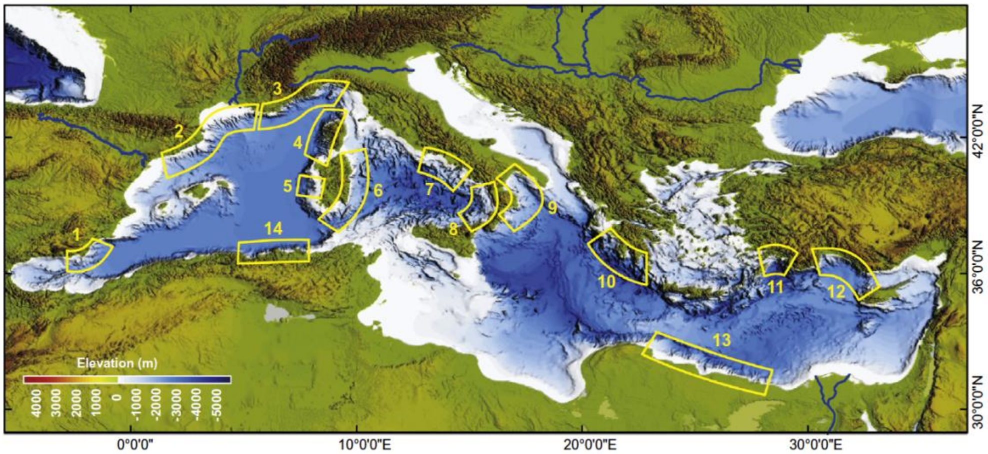


Figure 1.5. General bathymetric and topography of the Mediterranean region indicating the concentration of shelf-incised submarine canyons occur (yellow polygons). 1: Southeast Spain. 2: NW Mediterranean Sea. 3: Ligurian Sea. 4: West Corsica. 5: West Sardinia. 6: East Sardinia. 7: Campania margin. 8: West Calabria margin. 9: Taranto Gulf. 10: Peloponnese margin. 11: Fethiye margin. 12: Antalya-NW Cyprus margin. 13: West Egypt-East Libya margin. 14: East Algeria margin. From Canals et al. (2013)

Overall, the main physical features of Mediterranean deep waters are that, below 200 m depth, temperature is high and relatively stable (12-14°C), salinity is high (38.0-39.4 ‰) and oxygen levels are also high (4.5-5 ml/l), with the exception of specific areas such as the anoxic brine lakes of the central part (Tyler, 2003; Sardà et al., 2004).Moreover, there is a high decomposition rate of sinking OM, resulting in a lower quality of the major food source reaching the bathyal and abyssal seafloor, which are impoverished compared to those in other oceanographic regions (Sardà et al., 2004; Ramirez-Llodra et al., 2008). Accordingly, the deep Mediterranean Sea is one of the most oligotrophic regions in the world, showing many strong environmental gradients, mainly bathymetric but also including an eastward decrease in energy in terms of primary production and carbon export from the photic zone (Danovaro et al., 1999).

The deep Mediterranean seafloor presents a complex structure with markedly different habitats (Sardà et al., 2004; Danovaro et al., 2010) that include sedimentary slopes, submarine canyons and sea hills, deep basins, cold-water coral ecosystems, hydrothermal vents, cold seeps and deep anoxic basins (Danovaro et al., 2010; Company et al., 2012) (Fig.1.5). The presence of these systems results in a largely scale heterogeneity of the continental margin and deep basin (Buhl-Mortensen et al., 2010).

The Mediterranean Sea is considered one of the most explored areas since antiquity, and boasts a remarkable biodiversity from the coastal zones down to bathyal settings. The distribution patterns of the deep-sea Mediterranean meiobenthos are relatively well known (Danovaro et al., 1995; Gambi et al., 2010; Pape et al., 2013a; Sevastou et al., 2013), although most studies concern the North-Western (NW) Mediterranean (see Gambi et al., 2010 for review). Since the first study on deep-sea Mediterranean meiofauna by Soyer (1971) off Banyuls-sur-Mer (France), its knowledge has increased considerably. In general, meiobenthos density in the deep-sea Mediterranean is significantly lower when compared to the adjacent open Atlantic Ocean, and decreases eastwards (Gambi et al., 2010; Sevastou et al., 2013). Such traits are likely governed by differences in trophic regimes commented above, including the marked oligotrophy when compared with the Atlantic, and the subdued influence of the organic-rich Atlantic water masses moving towards eastern Mediterranean (Gambi et al., 2010, for review). Water depth and food availability (e.g., labile OM) largely explain the control of most meiofaunal descriptors (abundance, biomass and diversity) in the deep-sea Mediterranean Sea; and the importance of food decreases with increasing water depth (Gambi et al., 2010 and references therein). However, most of the variance remains unexplained suggesting that other factors such as the hydrodynamic forcing (e.g., episodic events, deep currents, other unexplored yet environmental variables), and/or topographic heterogeneity could be extremely important to explain the distribution of meiofaunal assemblages.

Concerning submarine canyons systems, the comparison between canyons and the adjacent open slopes did not reveal clear differences in terms of abundance, biomass and diversity, but also there is a lack of consistent bathymetric patterns for both habitats (Bianchelli et al., 2010; Danovaro et al., 2009). This underlines the importance of acquiring a better understanding of these peculiar topographic features. Furthermore canyons are also subjected to anthropogenic inferences tightly linked to impacts on deep-sea biodiversity (e.g., Puig et al., 2012; Martín et al., 2014b).

1.5.2 The NW Mediterranean/Catalan margin

The present research took place along the Catalan of the NW Mediterranean; a region is characterized by its narrow continental shelf, which represents most of the photic zone. This shelf also harbours a complex network of submarine canyons (i.e., Cap de Creus, Fonera, Blanes, Berenguera and Foix), which are among the largest of the ca. 700 submarine canyons identified in the Mediterranean (Harris et al., 2014) (Fig.1.6). These canyons originated during the inferior Oligocene-Miocene, and generated deep incisions in the shelf and slope (Tassone et al., 1994).

The Catalan margin is delimited by the Cap de Creus to the north and Ebre´s Delta to the south. The main north-Catalan submarine canyons are shelf incising, have their heads close to the shoreline and reach the base of the continental slope (Lastras et al., 2011). The largest one is Blanes Canyon, where the present study was conducted. It measures 184 km long and separates the region of the Gulf of Lyon from the Barcelona's continental shelf and the southern Catalan margin (Amblas et al., 2006) (Fig. 1.6).

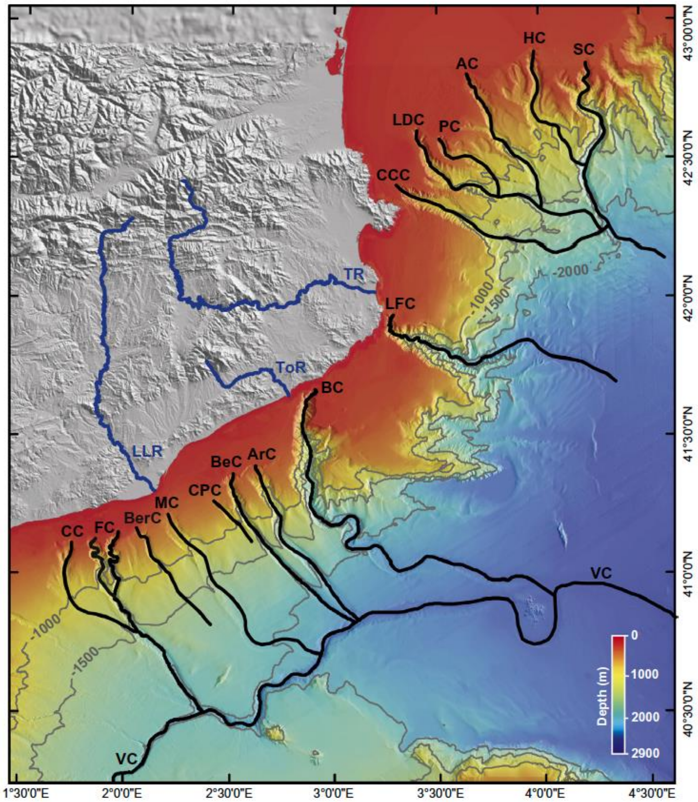


Figure 1.6. Detail of submarine network of the NW Mediterranean Sea including the Western Gulf of Lion and the Catalan margin. SC: Sete Canyon. HC: Herault Canyon. AC: Aude Canyon. PC: Pruvot Canyon. LDC: Lacaze-Duthiers Canyon. CCC: Cap de Creus Canyon. LFC: La Fonera Canyon. BC: Blanes Canyon. ArC: Arenys Canyon. BeC: Besos Canyon. CPC: Can Pallisso Canyon. MC: Morras Canyon. BerC: Berenguera Canyon. FC: Foix Canyon. CC: Cubelles Canyon.VC: Valencia Channel. CC. The main rivers are also indicated: TR: Ter River. ToR: Tordera River. LLR: Llobregat River. From Canals et al. (2013)

1.5.3 Anthropogenic pressure in the Catalan Sea canyons

Blanes Canyon and their adjacent margins in particular, are important areas for trawling. The main target is *Aristeus antennatus*, the deep-sea red shrimp, which supports a specialized commercial fleet (~17 trawling-boats) that has been exploiting the area for over 60 years (Ramírez-Llodrà et al., 2010a; Sardà et al., 2009). Their captures represent the main economic income in Blanes occupying the first place of money collection (23.8% of total, 2.465.835 € in 2016), although their catches are not the highest (~3.3%, 92.118 kg of the total captures in 2016) (agricultura.gencat.cat).

This commercial benthic trawling has been performed constantly in the Blanes area (and in numerous NW Mediterranean canyon fishing grounds) down to 700-

800 m depth since the 1960s, causing a profound impact on the seafloor: By removing benthic biomass from the upper slope and around the margins of the deeply incised local canyons by physical reworking, by have caused significantly reduction of the seafloor heterogeneity (Martin et al., 2014a; Puig et al., 2012).

Such significant impact on the seafloor has been observed on the nearby La Fonera (or Palamós) Canyon, where trawling gears passing near and along the flanks (Puig et al., 2012). Trawling gears produce daily extensive sediment resuspension, erosion and organic carbon impoverishment, ultimately, resulting in enduring changes to seafloor morphology at the spatial scale of the entire continental margin (Puig et al., 2012; Martín et al., 2014a). In addition, intensive bottom-trawling activities severely impacted the biological communities. Particularly, the meiobenthic communities suffered from the continuous mixing of seabed soft sediments over the years that led to decreases in meiofaunal abundance and diversity of ca. 80% and 50%, respectively, while the nematode diversity decreased a 25% compared to similar areas where no trawling occurs (Pusccedu et al., 2014). Moreover, the impacts of trawling-induced sediments re-suspension are not restricted to fishing grounds, since they are advected towards greater depths, concentrated within nepheloid layers, and deposited into canyons though sediment-landen density floods flowing along the steeped canyon flanks (Palanques et al., 2006b; Puig et al., 2012; Martin et al., 2014c; Paradis et al., 2017).

Considering the long-term bottom trawling activities in the study zone, sedimentary alterations similar to those described in Palamós Canyon may also occur, having also comparable effects on faunal abundance and diversity, as well as on the structure and trophic status of the communities. In Blanes Canyon, the sea urchin *Brissopsis lyrifera* was found the dominant non-crustacean megafauna at 1500 m depth and was completely absent in the open slope (Tecchio et al., 2013). Similar results were also observed at shallower depths (Ramirez-Llodra et al., 2008; Ramirez-Llodra et al., 2010a). However, according to fishermen that operate benthic trawling in the area, this sea urchin was highly abundant also in the open slope in previous decades, suggesting a high impact of bottom-trawling activities on this low-mobility species, while the canyon may be acting as a refuge area. No similar studies have been conducted on meiofaunal communities, so that the direct and/or indirect trawling effects on the meiobenthos remains unknown to date in the Blanes Canyon system.

Blanes Canyon has also been affected by other anthropogenic pressures, because the characteristically intense hydrodynamic processes enhance the transport of litter (Tubau et al., 2015) and chemical pollutants (Castro-Jimenez et al., 2013; Koenig et al., 2013) from shelf to deep-sea, which they can affect its fauna.

1.5.4 Research framework

Blanes canyon is arguably one of the most intensively explored active canyons during the last years, together with its adjacent slopes. This included numerous disciplines, from basic geological and hydrological surveys to research on fish trawling impacts (e.g., Zuñiga et al., 2009; Lastras et al., 2011; Company et al., 2008; Ramirez-Llodra et al., 2010a). The intensive fisheries mainly targeting the red shrimp *Aristeus antennatus* is among the main reasons explaining such a growing scientific interest.

Several successive multidisciplinary Spanish research projects, RECS (300-1,500 m depth), PROMETEO (900-1,500 m depth), and DOS MARES (500-2,000 m depth) have been conducted in the Blanes canyon area during the years 2003-2004, 2008-2009 and 2012-2013, respectively, with their main focus on describing the canyon habitat, particularly for fish and macrofauna crustaceans of commercial interest. Particularly, the present thesis was conducted within the framework of DOS MARES project, covering the continental margin not sampled in the two previous projects. The specific goal of the DOS MARES project was to study the environmental factors and biological communities in the area of Blanes Canyon and its adjacent western slope to understand the biological response to the observed environmental variability in the main environmental drivers, which certainly including the meiofaunal responses.

As a result of this successive series of projects, a huge sampling effort has been conducted along the canyon flanks, axis, head and adjacent slope using either benthic trawls (otter-trawl Maireta system and Agassiz dredge) and/or epibenthic sledges. These studies revealed new findings on the spatio-temporal variability of biomass and diversity of mega- and macrobenthos (Tecchio et al., 2013; Almeida et al., 2017), the seasonal movements of deep-sea fish and invertebrate populations (Company et al., 2008; Aguzzi et al., 2013; Tecchio et al., 2013), the community succession in allochtonous wood of both xylophagain bivalves and microbial communities (Romano et al., 2013a; Fagervold et al., 2013; Bessette et al., 2014), the biodiversity of suprabenthic fauna (Almeida et al., 2017), and the reproductive patterns of fish and non-crustacean macrofauna (Fernández-Arcaya et al., 2013; Mecho et al., 2015). Most of them were included in a special issue of the journal Progress in Oceanography (Canals et al., eds., 2013).

As a result, the importance of the canyon role in shaping the benthic communities has been stressed. For instance, Ramírez-Llodrà et al., (2010a) (400-700 m depth) and Tecchio et al., 2013 (900 and 1500 m depth) found significant canyon-slope differences in community composition, suggesting habitat heterogeneity as a regulator of benthic diversity. Indeed, Tecchio et al. (2013) proved that the canyon megafauna formed a more diverse community than that in the outer continental slope, but with comparable levels of abundance and biomass, and seasonal fluctuations that seem to be driven by deep water-mass movements. More recently, Almeida et al. (2017) reported that the motile macrofauna (the so-called suprabenthos) shows lower diversity in the canyon than on the more sedimentary-stable adjacent open slope.

However, as in many other canyons, the meiobenthic component had been almost neglected in Blanes Canyon. In fact, there are only two studies dealing with the spatial and temporal patterns of meiofauna, as well as those of the nematode standing stocks, gender-life and feeding type. Both studies were based on samples collected at the canyon head (400 m), upper region (600 m), walls (900 m) and axis (1600 m), complemented with others collected at the eastern open slope (800 and 1500 m) during the RECS project (autumn 2003 and spring 2004) (Romano et al., 2013b; Ingels et al., 2013). Both studies revealed that differences between canyon and slope were observed in terms of abundance and community composition. However, due to the high heterogeneity and environmental variability inside the canyon, no consistent differences over time were identified.

The overall lack of detailed information on the role of the Blanes Canyon in shaping the associated meiobenthic assemblages triggered the interest of the researchers involved in the DOS MARES project. As a consequence, the project work plan included an intensive inter- and intra-annual sampling never performed before in the Blanes Canyon system, which also covered the entire central axis. The present thesis covers this sampling effort in terms on the meiofaunal assemblage paying special attention in describing its diversity (previously studied at a higher taxa level only) by analysing the composition of the nematode populations at the genus level, with complementary information on kinorhynchs.

AIMS AND STRUCTURE OF THE RESEARCH

The general aim of this Thesis was to increase the knowledge and further understanding of the patterns and trends on density, diversity and composition of submarine canyon fauna, as well as on its main environmental drivers, by focusing on metazoan meiofauna and, particularly, on the dominant nematode assemblages. The following specific aims were addressed:

To describe the spatio-temporal fluctuations, in relation with environmental variables.

To describe the distributional patterns of biomass, abundance and diversity.

To identify the most important environmental factors structuring the above described trends and patterns.

To assess the canyon role in comparison with the adjacent open slope.

To assess the importance of the scales of observation (i.e., regional, bathymetrical and sediment profile).

Following this General Introduction (**Chapter 1**) and a generic Material and Methods section (**Chapter 2**), the detailed background information and methods, together with the results and discussions on the specific topics addressed in this Thesis, are presented in four different chapters, starting from an overall study of the meiobenthic assemblages (**Chapter 3**) and followed by detailed studies on nematodes (**Chapters 4 and 5**) and kinorhynchs (**Chapter 6**). **Chapters 3** to **6** have been structured as scientific papers in peer-reviewed journals, therefore are self-contained and can stand alone when prepared in the article-format and published (except for the general common methods, which are summarized in **Chapter 2**). Nevertheless, in this Thesis, the order of these chapters follows a specific thread. Each chapter is related to the others, providing a global and comprehensive vision of the meiofaunal communities living in the Blanes canyon system. In detail:

Chapter 3 investigate the spatio-temporal distribution of the metazoan meiofauna from Blanes Canyon and its adjacent western slope following a bathymetric gradient between 500 and 2,000 m depth, over two seasons (spring and autumn) and two years (2012 and 2013). Here there was an attempt to relate the observed differences in densities and higher taxa composition with natural (environmental variables) and anthropogenic (bottom trawling) drivers. Additionally, meiofaunal density patterns are discussed within the context of the actual knowledge on

Mediterranean and NE Atlantic submarine canyons. This chapter has already been published (Román et al., 2016).

Chapters 4 characterizes the nematode community structure, function and diversity along the Blanes Canyon axis, testing the effect of two major spatial scales (bathymetric and vertical sediment profile). The study of the relationships between environmental conditions and nematode communities allows to assess the overall canyon impact on meiobenthic communities. Additionally, nematode standing stocks (total biomass and density) and diversity are discussed in comparison to other the Mediterranean and NE Atlantic submarine canyons. This chapter has been submitted and is now under review (Roman et al., submitted).

Chapter 5 expands the basis laid out in the **Chapter 4** to focus on comparing canyon and slope habitats from the mid-deep and deepest stations (1,500, 1,750 and 2,000 m depth). The bathymetric trends and the effects of contrasting spatial scales (habitat, bathymetric and vertical sediment profile) on the nematode assemblages are assessed. Trends in community structure and structural diversity are discussed within the frame of the existing environmental conditions.

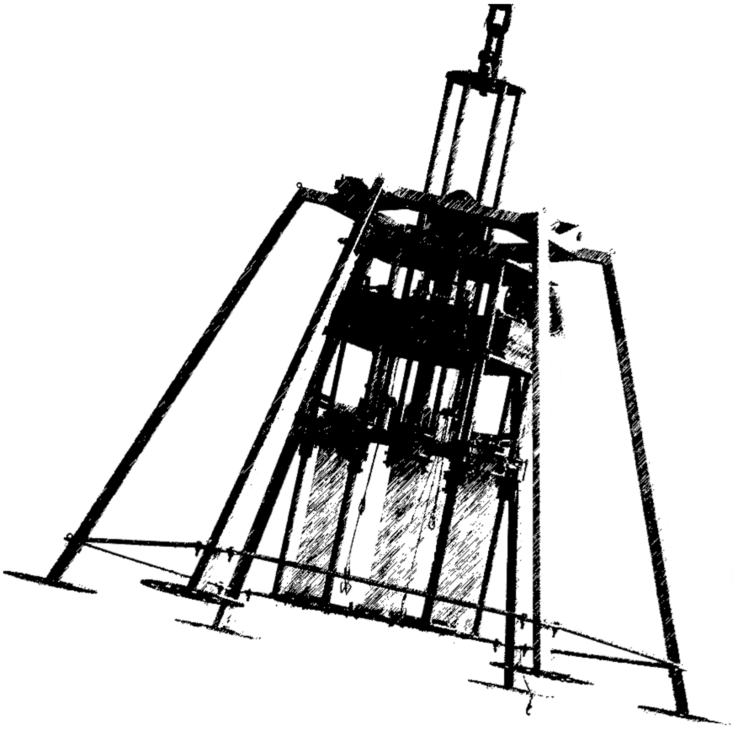
Chapter 6 investigates the phylum Kinorhyncha by comparing the density and diversity patterns along the canyon with those on the adjacent western slope. The obtained results increase the actual knowledge of often-neglected meiofaunal taxa. In fact, the chapter describes for the first time the quantitative local density and biodiversity patterns of a deep-sea kinorhynch community from a submarine canyon and its adjacent open slope, paired with bathymetric patterns and temporal trends.

The final chapter (**Chapter 7**) of this Thesis includes the conclusions summarizing the main results, together with the future directions of the studies dealing with meiofaunal assemblages from submarine canyon system.

Complementary information from some chapters is included as appendices, together with preliminary results on the temporal variation of the nematode assemblages from Blanes Canyon.

2

General Material & Methods



2.1 STUDY AREA

2.1.1 Morphology of Blanes Canyon

Blanes Canyon deeply cuts the continental shelf slope of the Catalan margin of the Mediterranean Sea. The canyon head incises the shelf at 60 m depth, only 4 km offshore the coastline, facing the Tordera deltaic area (Figs. 2.1,2.3). Its head first runs parallel to the coast to turn then offshore in a NW-SE direction. Its width increases with depth, reaching a maximum of 20 km wide at its deepest part, where it turns to a W-E course before outflowing to the lower Valencia Channel (Amblas et al., 2006; Lastras et al., 2011). The upper course characteristically shows the V-shaped cross-section, indicative of intense erosion processes, and is flanked by several gullies. The lower course has a U-shaped cross-section, indicative of high sediment deposition (Lastras et al., 2011). The canyon walls also show contrasting morphologies, with the eastern one being relatively smooth and the western one being sharper and heavily gullied (Lastras et al., 2011).

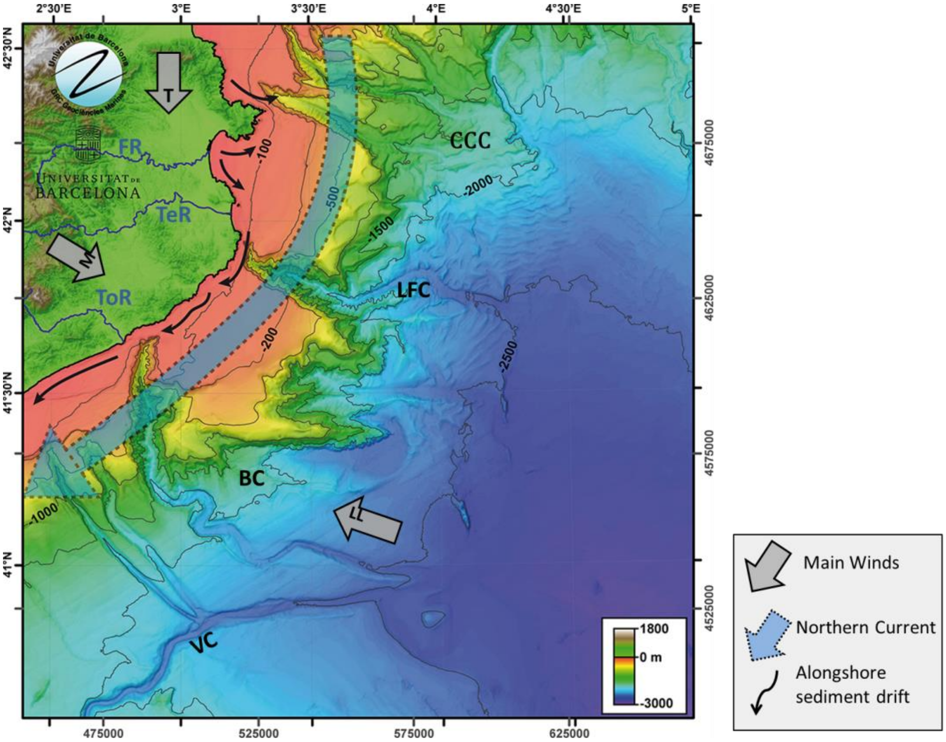


Figure 2.1. General bathymetric of the Catalan margin showing its main physiographic, oceanographic, hydrological and meteorological elements and location of the study area BC (Blanes Canyon). The most persistent winds: T: Tramuntana. M: Mistral. LL: Llevant. The main river are: FR: Fluvià River. TeR: Ter River. ToR: Tordera River. Modified from Tubau et al. (2015).

This particular topography plays an important role in the circulation of near bottom currents, which results in a highly variable flow at the eastern wall, and a prevailing offshore-directed flow over the western one (Zuñiga et al., 2009).

2.1.2 Oceanographic, meteorological and hydrological settings

The general circulation pattern in the NW Mediterranean is dominated by the cyclonic mesoscale flow of the Northern Current (NC). The NC comes from the Ligurian Sea, flowing south-westward along the continental shelf/ slope of the Gulf of Lions and the Catalan Sea, thus generating a dominant south-westward transport of suspended materials (Flexas et al., 2002) (Fig. 2.1). The NC is a permanent energetic flow involving principally: Modified Atlantic Water (MAW) at the surface and the Levantine Intermediate Water (LIW, 200-700 m) and the Western Mediterranean Deep Water (WMDW) at depth. The NC forms a 30 km wide sinuous stream that extends from the sea surface down to at least 400 m depth (Millot, 1999). Its movement is mainly forced by the AW entrance into the Mediterranean through the Gibraltar Strait, which extends from the surface down to 100–400 m depth (Millot, 1999). The NC tends to be faster and deeper in winter, while models hypothesize that upwelling and down-welling events associated with NC meanders crest and troughs occurs when passing over Blanes Canyon, so that the effects of the NC meanders may extend and propagate down to the deeper part of the canyon (Ahumada et al., 2013).

The main dominant winds in the NW Mediterranean and, thus, in the Blanes area, blow from the north over the European landmass, such as the dry *Tramuntana* (north) and *Mistral* (north-west), and from the eastern, such as the *Llevant* (south-east to east) that blow over the Mediterranean Sea and are thus humid (Fig. 2.1). The wind regime plays a pivotal role in driving the oceanographic dynamics of the NW Mediterranean Sea being also related with the two types of atmospheric forcing causing short-term high-energy events in this area. Among them, there are the east humid storms, characterized by intense precipitation, strong winds, and generation of high waves (Sanchez-Vidal et al., 2012), but also the dry northern storms, which include cold, dry winds that, during winter may gave rise to the formation of dense shelf water cascading (DSWC) events (Fig. 2.2)(Canals et al., 2006).

The transfer of matter (especially, bioavailable OM) along the submarine canyon from the highly productive continental shelf to the deep basin is strongly controlled by hydrodynamic processes (i.e., storms, DSWC and deep convection) and annual bioclimatic events (e.g., primary production "bloom" in winter-spring). This causes strong differences of quantity and quality of food between the canyon and the adjacent slope, which may be mirrored by the infaunal organisms following trophic adaptations.

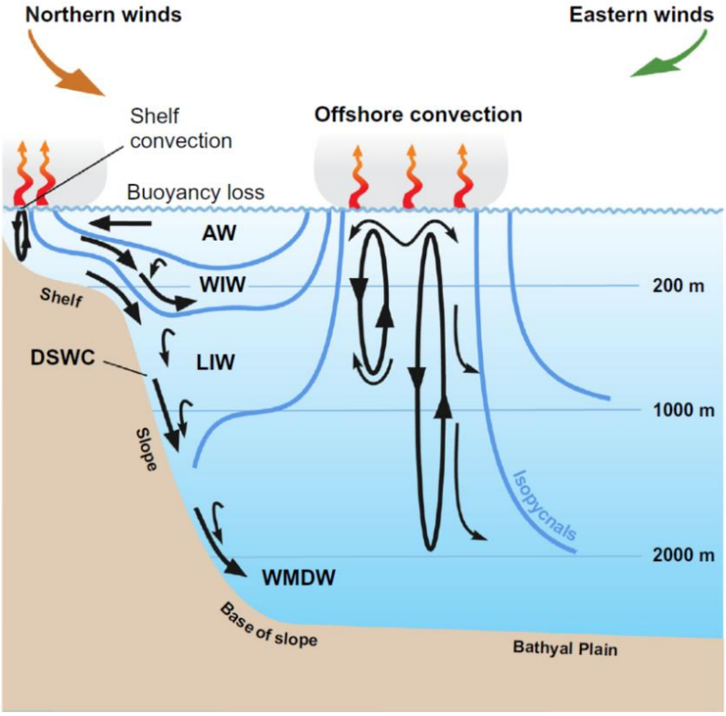


Figure 2.2. Schematic diagram of NW Mediterranean water mass circulation, dense shelf water cascading and open sea convection processes. From Canals et al., 2013.

DSWC and offshore convection tend to occur roughly synchronously (Fig. 2.2). They are triggered by cold and dry north winds (forced by the *Tramuntana* and *Mistral* winds) where they can cool the sea surface (Atlantic Water: AW), leads to the loss of buoyancy of surface waters and their cooling which becomes denser and sinks, producing the Western Intermediate Water (WIW). The WIW can be detected below the AW and over the LIW, mainly in winter. In short, surface waters increase their density until passing a threshold from where they start sinking down to the depth until reaching again density equilibrium. The large and wide shelf of the Gulf of Lion behave as shallow cooling platforms, which reduce water column stability so that densified surface water first sink to the shelf floor and then it stars moving over the bottom till the shelf edge to finally cascade downslope mostly along submarine canyons (Canals et al. 2006; Durrieu de Madron et al., 2013). In the NW Mediterranean Sea, cascading events occurs almost every year (Houpert et al., 2016), but also occurs in the Adriatic Sea (forced by the Bora wind), and the Aegean Sea (forced by the Etesian wind) (Pusceddu et al., 2010).

Overall, the oceanographic cycle in Blanes Canyon and nearby areas have two clearly defined seasonal periods: an intense winter, when DSWC, offshore convection and occasional eastern storms occur, and a calm summer, when the NC becomes the dominant influence, only interrupted by sporadic eastern storms (Lopez-Fernandez et al., 2013). The hydrodynamic and atmospheric processes play important role in driving the particle flows through the canyon, which in turn have a great importance for the biogeochemical cycles and marine ecosystems of the area. Particulate transport though the canyon also presents a well-defined seasonality (Heussner et al., 2006; Zúñiga et al., 2009; Lopez-Fernandez et al., 2013). During autumn-winter, mass fluxes increased in response to flooding of the Tordera River and the numerous nearby streams, as well as to major coastal storms. In spring-summer, they are mainly driven by the phytoplankton bloom (Heussner et al., 2006; Zúñiga et al., 2009; López-Fernandez et al., 2013). Maximum downward fluxes are related with storms, winter-river discharges, intensification of the NC and DSWC. The downward transfer of particles (and OM) through the canyon takes place along its axis but also by lateral inputs from a dense network of gullies carved in the canyon flanks (López-Fernandez et al., 2013). In turn, the DSWC events originated in the Gulf of Lion and particularly affecting the North Catalan Margin merits special attention. The particular topographic and hydrodynamic features of some submarine canyons can favour or even amplify the effects of DSWC (Allen and Durrieu de Madron, 2009), being able to have a great influence on the biodiversity and functioning of these ecosystems (Martin et al., 2006; Durrieu de Madron et al., 2000; Duineveld et al., 2001; Bianchelli et al., 2008; Company et al., 2008; Pusceddu et al., 2013). Moreover, as it has been demonstrated that the associated intense mixing of the water column may have a great influence on the functioning of these ecosystems and, thus, on their biodiversity (Martin et al., 2006; Durrieu de Madron et al., 2000; Duineveld et al., 2001; Bianchelli et al., 2008; Company et al., 2008; Pusceddu et al., 2013). For instance, the high speed near bottom currents (velocities close to 1 ms-1) funnel large amounts of organic matter to the deep sea (Heussner et al., 2006) reaching huge amounts of material down to the deepest bottoms of the continental margin (beyond 1,000 m depth) (Canals et al., 2006; Palanques et al., 2012; Durrieu de Madron et al., 2013). Moreover, they enhance ventilation (oxygen supply) of intermediate and deep-sea waters, thus contributing to the formation of bottom water masses in the western basin (WMDW) and modifying the morphology of the deep-sea floor (Canals et al., 2006; Puig et al., 2008; Lastras et al., 2007; Lastras et al., 2011). These high currents can diminish the benthic faunal abundance, as in the case of the deep-sea shrimp *Aristeus antennatus* whose fisheries may be temporary collapse (Company et al., 2008), but can also significantly affect the meiofaunal abundance and biodiversity, which are diminished, due to either being flushed away or buried in the canyons (Pusceddu et al., 2010; Pusceddu et al., 2013).

An additional but relevant source of particles contributing to the fluxes in the Blanes Canyon area, may derive from the Sahara dust combined with antrophogenically mediated resuspension processes linked to trawling activities (Lopez-Fernandez et al., 2013; Martin et al 2014b; Puig et al., 2012).

2.2 SAMPLING DESIGN AND COLLECTION

The research conducted in this thesis focused on the study of the deep-sea meiofaunal and their seasonal and bathymetric trends and patterns. To achieve this main objective (see **Chapter 1**), it was necessary to adopt different sampling and sample processing strategies, which are detailed here below.

Within the frame of the DOSMARES research project, sediment samples were collected in and around Blanes Canyon on board of the R/V García del Cid. The sampling design covered the whole bathymetric range of the Blanes Canyon and its adjacent western continental slope, from 500 to 2,000 m depth. Four seasonality cruises were carried out: DM-I (early spring 2012), DM-II (autumn 2012), DM-III (spring 2013) and DM-IV (autumn 2013) (Table 2.1, Fig. 2.3). Six stations were sampled along the canyon axis (BC500, BC900, BC1200, BC1500, BC1750 and BC2000) and five on the adjacent open slope (OS500, OS900, OS1500, OS1750 and OS2000).

2.2.1 Sediment sampling

Samples for meiofauna and sediment variables were obtained using a 6-tube multicore KC Denmark A/S (inner diameter 9.4 cm; length 60 cm), yielding samples with an intact sediment-water interface (Fig. 2.4A). Between 1 and 3 multicore deployments (replicates) were conducted at each sampling station depending on the cruise (Table 2.1). In total, there were 77 successful multicore deployments.

Table 2.1. Samples collected and studied in the present thesis, including the site, geographical position, depth range (minimum-maximum) and sampling intensity (number of replicates), indicating, year and period-days of the sampling campaigns. DM: DOS MARES campaign; BC: Blanes canyon, OS: open continental slope.

Site	Station	Latitude	Longitude	Depth (m)	Sampling intensity			
					2012		2013	
					DM-I	DM-II	DM-III	DM-IV
					11-15/03	07-12/10	5-11/04 22-23/04	26-28/09
Canyon	**BC500**	41°38´66´´N	02°52´75´´E	462-484	2	2	1	3
Canyon	**BC900**	41°34´28´´N	02°50´95´´E	835-903	2	3	3	3
Canyon	**BC1200**	41°30´93´´N	02°51´07´´E	1194-1258	2	2	3	–
Canyon	**BC1500**	41°27´37´´N	02°52´93´´E	1457-1520	2	3	3	3
Canyon	**BC1750**	41°21´51´´N	02°52´07´´E	1726-1785	2	3	3	3
Canyon	**BC2000**	41°14´90´´N	02°52´97´´E	1943-1980	2	3	–	3
Slope	**OS500**	41°19´10´´N	02°46´75´´E	493-509	–	–	3	–
Slope	**OS900**	41°16´29´´N	02°48´96´´E	887	–	–	1	–
Slope	**OS1500**	41°08´28´´N	02°53´75´´E	1451-1480	–	3	3	–
Slope	**OS1750**	41°06´79´´N	02°57´02´´E	1731-1751	2	3	3	1
Slope	**OS2000**	41°02´65´´N	03°01´22´´E	1975-1998	2	3	–	–

Despite the comprehensive sampling strategy, a full temporal and water-depth coverage was not possible owing to technical and logistical reasons (see Table 2.1 for details).

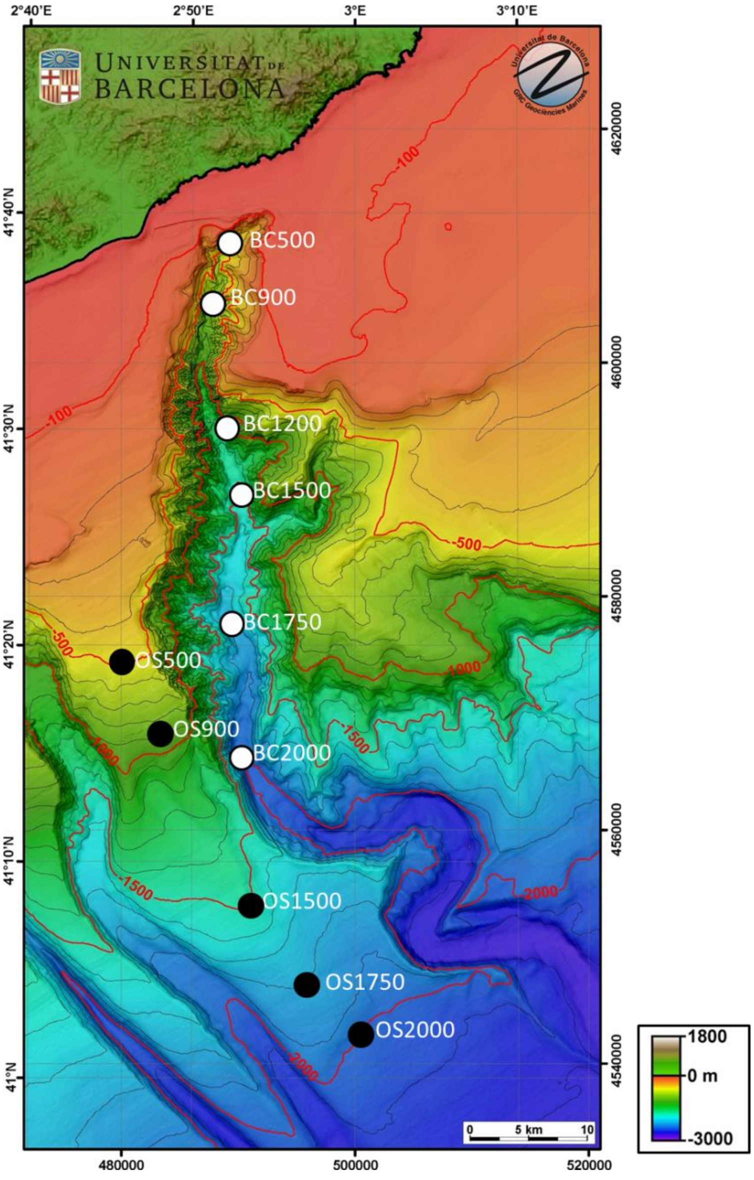


Figure 2.3. General bathymetric map of Blanes Canyon (BC) and sampling stations along the canyon, and on the adjacent western slope (OS). Contours are every 100 m (black lines and 500 m (red lines) excepts for the 100 m contour that is also red coloured. Bathymetric data from Canals et al (2004).

From each multicore deployment, one core was used for meiofaunal and three for sediment analyses. They were all carefully sub-sampled on board by means of a small PVC tube (36 mm of diameter, 5 cm sediment depth) taken from the center of the core to maintain a consistent sample surface area for all replicates (Fig. 2.4B-C). Each sub-core was successively sliced on board down to 5 cm along the sediment profile: 0-1 cm, 1-2 cm and 2-5 cm sediment layers (Fig. 2.4D), which were fixed immediately in a buffered 4% formaldehyde seawater solution for meiofauna and frozen and stored at -20°C for sediment analyses.

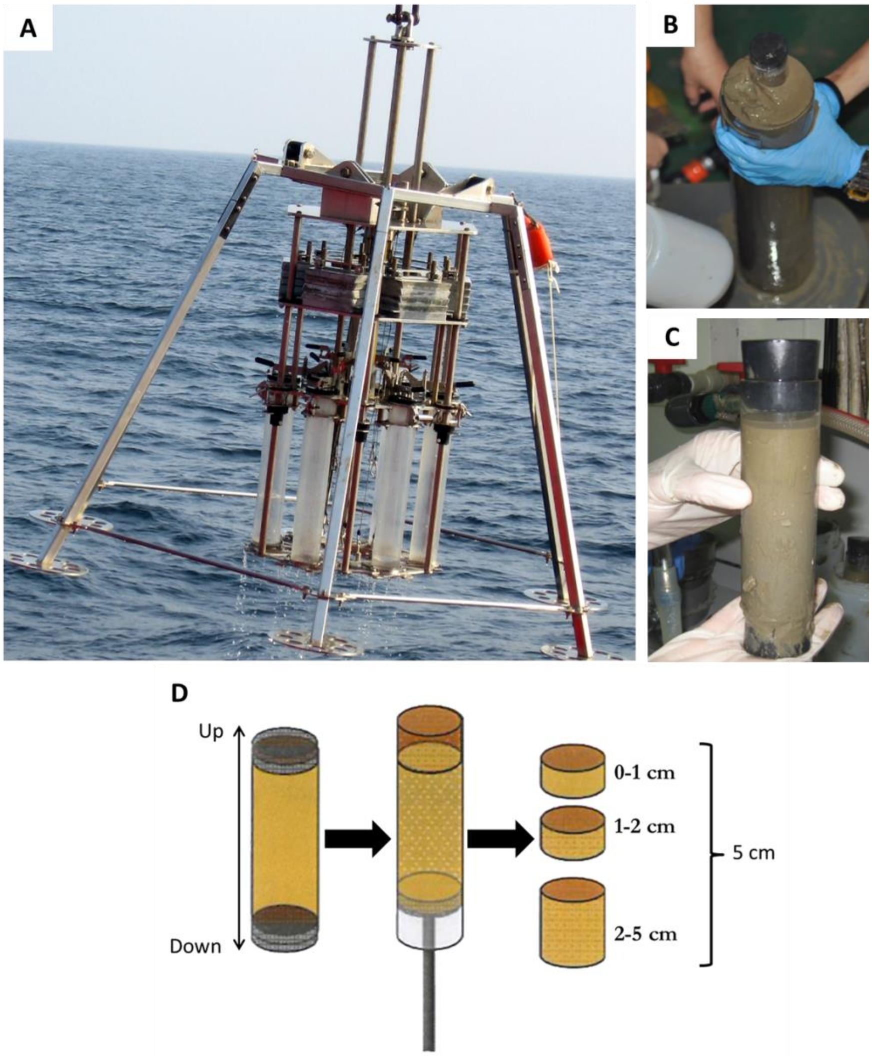


Figure 2.4. A)Multicore (6 tubes), B) Core subsampling. C) Sub-core of sediment for meiofauna and sediment samples. D) Scheme illustrating sub-core slicing.

2.3 ANALITICAL METHODS

2.3.1 Sediment variables

2.3.1.1 Grain size

Grain size was assessed using a Master Sizer 2000 laser analyser. Each sample was first defrosted and homogenized. Then, aliquots (~1 g) were treated with a 6% hydrogen peroxide solution for 48 h (to remove OM) and subsequently washed with distilled water to eliminate salts. Before each measurement 10 drops of distilled water with sodium hexametaphosphate (0.05 %) were added to disperse the sediment. Then, samples were homogenized prior to running the analysis. Sediments were grouped into grain size fractions as follows: clay (<4 μm), silt (4–63 μm) and sand (63 μm–2 mm).

2.3.1.2 Geochemical analysis

Aliquots of 0.5 to 5 g of sediment from each sample were freeze-dried to analyze total organic carbon (OC) and total nitrogen (TN) using an elemental analyzer Flash 1112 EA interfaced to a Delta C Finnigan MAT isotope ratio mass spectrometer at the "Centres Científics i Tecnològics de la Universitat de Barcelona". Samples for OC were first de-carbonated using repeated additions of 25% HCl with 60°C drying steps in between until no effervescence was observed (Nieuwenhuize et al., 1994).

2.3.1.3. Pigment content

Sediment aliquots were freeze-dried and homogenized to extract pigments. About 1 g dry weight of sediment was mixed in 4 ml 90% acetone and the obtained extract was centrifuged (4 min at 3000 rpm, 4°C) and filtered through a Whatman Anodisc 25 (0.1 μm). Pigments were analysed by Ultra Performance Liquid Chromatography. Chlorophyll *a* (Chl *a*) and chlorophyll degradation products were identified by checking the retention times and the absorption spectra against a library based on commercial standard mixtures (DHI, PPS-MiX-1) and extracts from pure cultures of algae and bacteria (protocol modified of Buchaca and Catalan, 2008). Chloroplastic Pigments Equivalents (CPE: sum of Chl *a* and its degradation products as phaeopigments) served to estimate surface-derived primary productivity at the seafloor (Thiel, 1978). The ratio Chl *a*: phaeopigments (Chl a: phaeo) was used as a proxy for OM quality at the seafloor. (Plante-Cuny and Bodoy, 1987). High pheopigments v.s Chl *a* are generally associated with detrital and degraded autotrophic matter inputs.

2.3.2 Meiofauna sampling and extraction

Each sediment layer was washed over stacked sieves with a 1000 µm mesh and a 32 µm mesh. The retained 32 µm fraction was washed and centrifuged three times using the colloidal silica polymer LUDOX HS40 (specific gravity 1.18) to achieve density gradient separation of the meiofauna (Heip et al., 1985). The supernatant of each washing cycle was again collected on a 32-µm sieve. After extraction, samples were kept in 4 % formaldehyde and stained with Rose Bengal. All metazoan meiobenthic organisms were counted under a stereomicroscope (50 x magnifications) and classified at higher taxon level following Higgins and Thiel (1988).

2.3.3 Nematodes

For nematode processing, between 100-150 individuals (all if density < 100) were randomly hand-picked out from each layer (0-1, 1-2 and 2-5 cm), gradually transferred to glycerine (De Grisse, 1969) and mounted on glass slides. Nematodes were identified under compound microscope (100 x magnification) to genus level using pictorial keys (Platt and Warwick, 1988) and the NeMys database (Guilini et al., 2016). Specimens that could not be ascribed to a genus level were grouped within the appropriate family to account for its presence in the sample.

The study of nematode community composition at genus level is broadly accepted and several studies have provided an appropriate basis for comparing communities in and between deep-sea habitats on a regional (e.g., Vanaverbeke et al., 1997; Ingels et al., 2009) and global scale (Vanreusel et al., 2010). Moreover, studies dedicated to answering ecological questions and the effect of environmental factors on nematode diversity, identification down to genus level may be sufficient as most deep-sea nematode genera have low environmental specificity (Leduc et al., 2012a).

The length (excluding filiform tails tips) and maximum body width of all nematodes identified were measured using a Motic BA210 compound microscope and TopView 3.7 imaging software. Nematode total biomass was then calculated according to Andrassy´s formula $G=W^2 \times L/1.5 \times 10^6$, where G= wet weight in µg, W= maximum body diameter (µm) and L= total length (µm) (Andrassy, 1956).

The wet weight was then converted into carbon weight assuming a 12.4% carbon ratio (Jensen, 1984). Total nematode biomass for each sediment layer, sampling station and feeding type, each average genus biomass was multiplied by the respective densities.

Trophic composition was assessed using Wieser (1953)´s feeding-types, based of buccal cavity organization (i.e., the size of buccal cavity, the present or absent of teeth): selective deposit feeders (1A), non-selective deposit feeders (1B), epistrate feeders (2A) and predators/scavengers or omnivores (2B) (Fig. 2.5). An additional feeding type (3) was incorporated for the mouthless genus *Astomonema* (Ingels et al., 2011a).

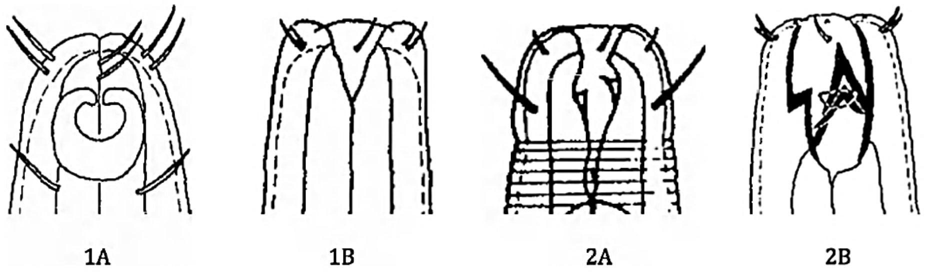


Figure 2.5. Representatives of the nematode feeding guilds according to Wieser (1953). !A: selective deposit feeder. 1B: non-selective deposit feeder. 2A: epistrate feeder and 2B: predator/scavenger.

2.3.4 Kinorhynchs

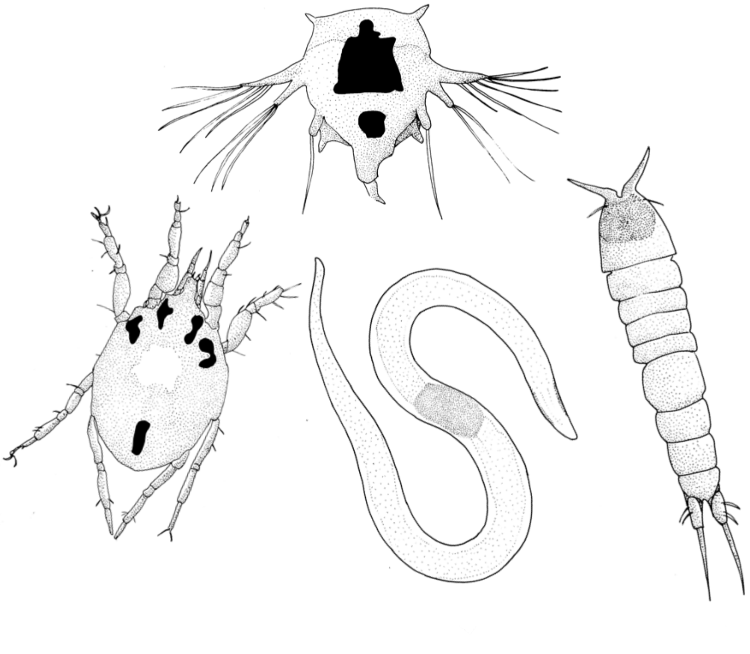
Kinorhynch specimens were sorted for the remaining meiofauna and counted under a stereomicroscope (50 x magnifications), and subsequently preserved in 70% ethanol until being identified. Specimens for light microscopy observations (LM) were dehydrated through a graded series of ethanol and transferred to glycerin prior tomounting in Fluoromount G®. They were examined and photographed using a Zeiss Axioplan 2 microscope with differential interference contrast optics (DIC) equipped with a Zeiss-Axiocam503-color camera. Specimens for scanning electron microscopy (SEM) were cleaned by exposing them to ultrasound intervals of 5–10 s, and then dehydrated through a graded series of ethanol, critical point dried, mounted on aluminium stubs, sputter coated with platinum-palladium, and imaged with a Hitachi S4700 field emission scanning electron microscope. Coating and SEM imaging were performed at the Bioimaging Facility at the University of British Columbia (UBC).

2.4 DATA ANALYSES

The specific methodologies, analytical and statistical measures and graphical representations are described in the Material and Methods section of Chapter 3 to 6. All analyses were performed using PRIMER v6 with PERMANOVA + add-on software (Clarke and Gorley, 2006; Anderson et al., 2008) and XLSTAT (Addinsoft) software.

3

Spatial and Temporal variability of Meiofaunal assemblages in the Canyon and its adjacent Open Slope



3.1 INTRODUCTION

There is increasing evidence that submarine canyons play important ecological roles in the functioning of deep-sea ecosystems (Amaro et al., 2016; Thurber et al., 2014) Submarine canyons are important routes for the transport of organic matter from surface waters and continental shelf areas to the deep sea basins (Granata et al 1999; Durrieu de Madron et al., 2000; Palanques et al., 2005; Canals et al., 2006; Pasqual et al., 2010), and they contribute significantly to regional biodiversity and secondary production along continental margins (Gili et al., 1999, 2000; Sardá et al., 2009; Ingels et al., 2009; Vetter et al., 2010; De Leo et al., 2010;). Different oceanographic and geological processes are responsible for their role as organic matter supply routes, including slope instabilities, turbidity currents events caused by river floods or episodic storms, and dense shelf water cascading (De Stigter et al., 2007; Allen and Durrieu de Madron, 2009; Puig et al., 2014). Additionally, anthropogenic activities along canyon flanks such as bottom trawling can alter seafloor community structure and biodiversity through physical habitat disturbance and the re-suspension of sediments, which ultimately accumulate at greater depths inside the canyon axis (Palanques, et al., 2006b; Martín et al., 2008, 2014; Puig et al., 2012, 2015a, b; Pusceddu et al., 2014; Wilson et al., 2015).

More than 700 large submarine canyons have been identified in the Mediterranean Sea (Harris et al., 2014) with several of them located along the Catalan margin (NW Mediterranean). Among those, Blanes Canyon has been intensively studied in the past decade (Zúñiga et al, 2009; Sánchez-Vidal et al., 2012; Canals et al., 2013; Lopez-Fernandez et al., 2013). It has been shown that the canyon axis experiences strong current intensifications coupled with high particle fluxes to the deepest parts of the canyon, which are linked to major events like storms and subsequent river discharges, as well as dense water formation during winter. These characteristics cause the off-shelf sediment transport through the Blanes Canyon axis to vary substantially over time. Moreover, the downward particle fluxes within the canyon can be affected by sediment resuspension caused by daily bottom trawling activities along the canyon flanks (Company et al., 2008, Sardà et al., 2009), with increased fluxes attributed to this process observed mainly between 900 and 1200 m depth (Lopez-Fernandez et al., 2013). In general, the particulate matter fluxes in the canyon area and at the adjacent open slope have two defined seasonal periods (Zúñiga et al., 2009; Lopez-Fernandez et al., 2013). During autumn-winter mass fluxes increase in response to river flooding coupled with major coastal storms. During the spring-summer months the biological component of the particulate fluxes becomes more important as a response to the phytoplankton bloom. However, the open slope mass flux has been reported to be three orders of magnitude lower than inside the Blanes Canyon with sinking particles containing lower amounts of lithogenics and higher levels of organic carbon (OC) (Lopez-Fernandez et al., 2013).

Most, if not all, oceanographic and geological processes that drive the ecological function and processes in submarine canyons elicit a response from the canyon fauna (Amaro et al., 2016). Elevated and depressed faunal densities have been reported in canyons, compared to slope areas (Cartes et al., 1994, 2009, 2010; Vetter and Dayton, 1998; Garcia and Thomsen, 2008; Ingels et al., 2009, 2013; De Leo et al., 2010; Tecchio et al., 2013; Romano et al., 2013a,b). Complex canyon hydrodynamics can lead to areas characterized by strong deposition next to areas typified by intense erosion processes, which are highly variable in time. These factors have been identified as driving marked temporal and spatial variability in benthic fauna standing stocks (De Bovée, 1990; Ingels et al., 2013; Romano et al., 2013b; Tecchio et al., 2013). Despite the difficulties associated with sampling these deep-sea environments, the collection of benthic biological time-series is crucial in providing the necessary information to understand ecological processes and their importance in canyon systems.

The main forcing processes affecting canyons influence benthic organisms, and the meiofauna is no exception in this context (Giere et al., 2009). Metazoan meiofauna dominate the deep-sea benthos in terms of abundance and biomass (Vincx et al., 1994; Rex et al., 2006; Giere, 2009; Pape et al., 2013a; Pape et al., 2013b). Quantitative deep-sea meiofaunal studies in the Mediterranean, have mostly focused on slope and basin environments (De Bovée et al., 1990; Soetaert et al., 1991; Tselepides and Lampadariou, 2004; Bianchelli et al., 2008; Romano et al., 2013b; Rumolo et al., 2015), while studies targeting wide bathymetric gradients along canyons axes are particularly rare (De Bovée, 1990; García et al., 2007; Bianchelli et al 2010). Meiobenthic taxa, such as Nematoda, seem particularly resilient to both natural and anthropogenic physical disturbance compared to other benthic organisms such as the macrofauna (Pusceddu et al., 2013). However, chronic deep-sea anthropogenic activities such as bottom trawling on canyon flanks can cause a reduction in meiofauna abundance and diversity through a decrease in OM content in the trawled sediments (Pusceddu et al., 2014).

The main objective of this Chapter was to study the distribution patterns of the metazoan meiofauna along the Blanes Canyon axis compared with those from the adjacent open slope. Samples were collected covering a bathymetric range from 500 to 2000 m depth in two periods (i.e. spring and autumn) and over two successive years (2012 and 2013) to identify variations in the main meiofaunal descriptors (i.e., density, community composition and richness of taxa). Subsequently, their relationship with the main driving sediment variables (e.g. grain size, organic content), including those related with food input (e.g. Chlorophyll *a* content), and environmental constraints (both natural and anthropogenic) were analyzed. Based on previous observations from submarine canyon systems we expect that Blanes Canyon will be characterized by high-density meiofauna communities, but also by high temporal community variability which is only partly explained by seasonal differences in food input.

3.2 MATERIAL AND METHODS

3.2.1 Sampling

Sediment samples were collected during the four DOS MARES cruises (DM-I: early spring 2012, DM-II: autumn 2012, DM-III: spring 2013 and DM-IV: autumn 2013), taking into account all the pool core samples collected in this study. The bathymetric range covered was from 500 to 2,000 m depth in the Blanes Canyon and on the adjacent western slope (Fig. 3.1). Despite a comprehensive sampling campaign, full temporal and water-depth coverage could not be obtained owing to technical and logistical reasons (see **Chapter 2** section **2.2** and Table 2.1 for details on cruises and sampling strategy).

3.2.2 Sediment and meiofauna analyses

In this study, the sum of the 5 cm of each sub-core collected was considered for meiofauna and environmental sediment variables (see **Chapter 2** section **2.2.1** for details on sediment cores collection). Meiofauna density was expressed as number of individuals per cm2 and communities were characterized based on the abundance of major taxa. Sediment variables considered in this studied were: grain size (clay (<4 μm), silt (4–63 μm) and sand (63 μm–2 mm)), organic carbon (OC) and total nitrogen (TN), Chlorophyll *a* (Chl *a*), phaeopigments (phaeo) and Chloroplastic Pigment Equivalents (CPE). The ratios Chlorophyll *a*: phaeopigments (Chl *a*:phaeo) and carbone:nitrogen (C:N) were also calculated. See **Chapter 2** section **2.3** for details on analytical methods for meiofauna and sediment samples.

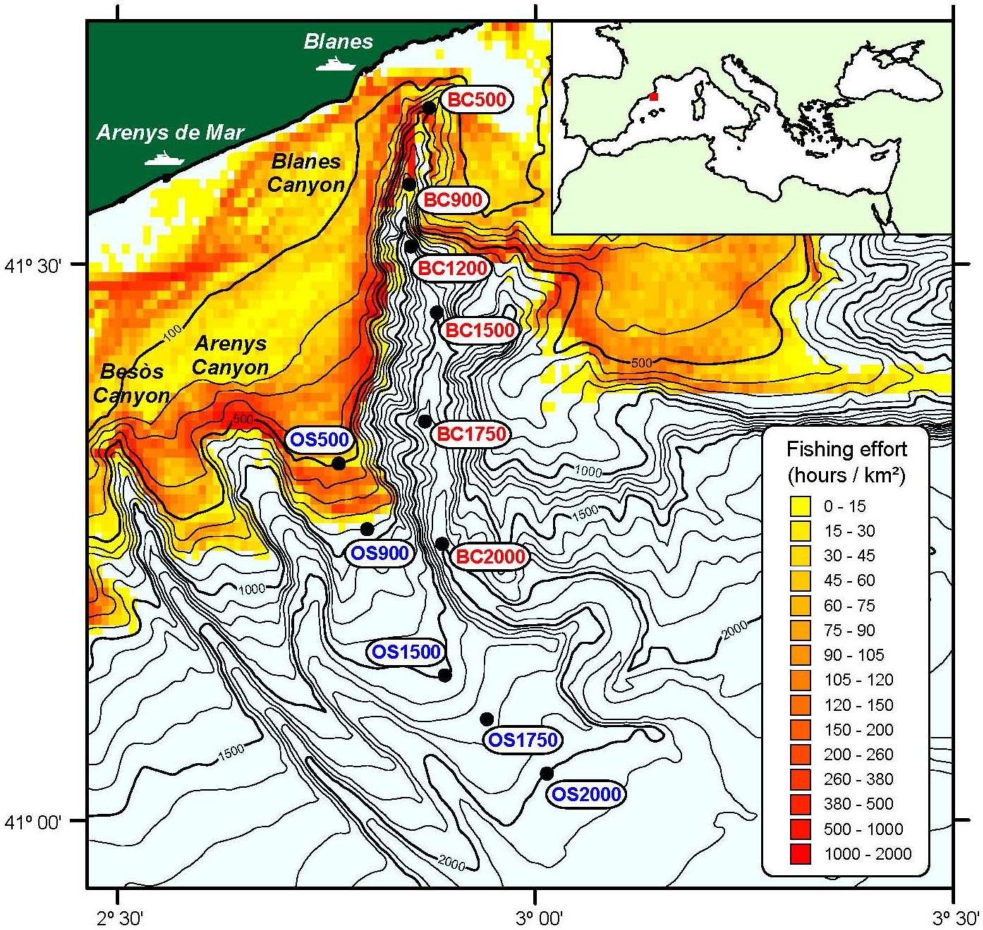


Figure 3.1. Map of the study area showing the sampling stations and the fishing effort (hours/km2). Contours are every 100 m (black lines) and 500 m (thick-black lines). Dots represent each station sampled. BC: Blanes canyon, OS: Western open slope.

3.2.3 Data analysis

Both univariate and multivariate non-parametric permutational (PERMANOVA) analyses were performed to test differences in meiofauna density and assemblage composition, as well as sediment variables between sampled periods, locations and depths. Differences between locations (i.e. canyon vs slope) and water depths were tested with two different 2-way crossed designs, using Location (Lo: fixed) and Water Depth (WD: fixed) as factors, both for uni- and multivariate analysis (PERMANOVA Anderson, 2005; Anderson et al., 2008). In the first 2-way PERMANOVA analysis to assess overall differences between locations we pooled the data from the four sampling times and considered all the depths (6 levels: 500, 900, 1200, 1500, 1750, 2000 m). Subsequently, we considered data from each campaign separately and performed a 2-way PERMANOVA analysis for each campaign including only the sampled depths that were in common between

canyon and slope (e.g. spring 2012: 1750 and 2000 m; autumn 2012: 1500, 1750, 2000 m; spring 2013: 500, 900, 1500, 1750, autumn 2013: 1750 m).
Considering that the sampling effort of this study was mainly focused on the canyon and for the slope a less complete data matrix was available, we also analyzed the data from each location separately by means of two-way crossed design using Time (Tm: fixed) and WD (fixed) as factors. For the canyon data the factor WD had 6 levels (500-2000 m, see Table 3.3) and for the slope it had 3 levels (1500-2000 m).

Additional non-metric multidimensional scaling (MDS) visualization was used to show the spatial variations of meiofauna community structure based on Bray Curtis similarity values.

Differences in sediment variables over the terms Tm and WD were assessed by univariate PERMANOVA, except for the grain size content, where clay, silt and sand percentages were considered as multivariate variables. Additionally, Principal Component Analyses (PCA) based on sediment variables were performed to assess differences in spatial distribution between the canyon and slope samples and the temporal and spatial patterns in the canyon. Prior to calculating the Euclidean distance resemblance matrix, the full set of 10 sediment variables was tested for collinearity (Draftsman plot and Spearman correlation) and variables with correlations ($R^2 > 0.95$ (redundant) were omitted from the analyses (i.e. phaeopigments). The data were then checked for uniform distribution (Chl *a*, CPE and Chl *a*: phaeo were log (0.01+X) transformed) followed by normalization (subtracting the mean and dividing by the standard deviation, for each variable) to bring them to a common unit before analysis.

The univariate meiofaunal density data were square root transformed and Euclidean distance was used to calculate the resemblance matrix. Multivariate analyses of the meiofauna assemblage composition were done on a Bray-Curtis resemblance matrix based on fourth-root transformed data.

Additionally, regression analyses were performed to reveal the form, distribution and significance (null hypothesis rejected when the significance level was > 5%) of the functional relationships between each dependent variable (sediment variables and meiofauna density, using water depth as independent variable) and assess the differences between canyon and slope samples.

Spearman correlations were used to assess the strength of the relationship between the selected sediment variables and meiofauna density (based on the complete data set, including both canyon and slope samples). Finally, the relationships between meiofauna composition and sediment variables were investigated using the

Distance based Linear Model routine (DISTLM) in PERMANOVA + (Anderson et al., 2008). The DISTLM assemblage was built using a step-wise selection procedure and adjusted R^2 was used as selection criterion.

3.2.4 Ancillary data: external forcing parameters

To assess the relative role of the various oceanographic processes contributing to the temporal variability of downward particle fluxes in the study area during the study period, surface primary production was obtained from satellite data of Chl *a* concentration at www.nasa.gov. Daily river discharge series measured at the nearest gauging station to Tordera River mouth was obtained from Agència Catalana de l´Aigua (ACA) and significant wave height was provided by Puertos del Estado (www.puertos.es).

To assess the impact of bottom trawling fisheries in the Blanes canyon, positioning of fishing vessels was obtained from the Fishing Monitoring Centre of the Spanish General Secretariat of Maritime Fishing (SEGEMAR) as Vessel Monitoring System (VMS) data, a protocol established by the Common Fisheries Policy of the European Union (2011). Each vessel equipped with VMS provide its registered harbour, heading, speed and Global Positioning System coordinates with an error margin of 100 m, and transmits this information by Inmarsat-C to the Fishing Monitoring Centre in less than 10 min at 2-hour time intervals (Gerritsen et al., 2013). VMS positioning from bottom trawlers operating in the study area during 2006-2013 was subsequently converted to fishing effort (hours/km2), computed in grid cells of 0.5´ latitude x 0.5´ longitude as shown in Figure 1, following the method and the software tools described in Hitzen et al. (2012).

3.3 RESULTS

3.3.1 Sediment characteristics

Canyon and slope sediments were predominantly muddy (2-63 μm) with high silt content (62% to 77%, respectively) over the whole bathymetric gradient (Table 3.1). Sediments were in general characterized by a high sand content in the canyon, except at BC2000 (pair-wise comparison Lo x WD, $p < 0.01$, Fig. 3.2, Table 3.2). Along the slope, sediments became progressively finer with increasing water depth ($R^2 = 0.79$ and 0.39 respectively for clay and sand, $p < 0.05$, Fig. 3.2; Table 3.1; PERMANOVA, $p < 0.05$, Table 3.3) whilst in the canyon there was no consistent bathymetric trend (Figs. 3.2, 3.3). In general (slope and canyon samples pooled), grain size decreased until BC900, peaked at BC1200 and, then decreased again with depth (Table 3.1, Fig. 3.3).

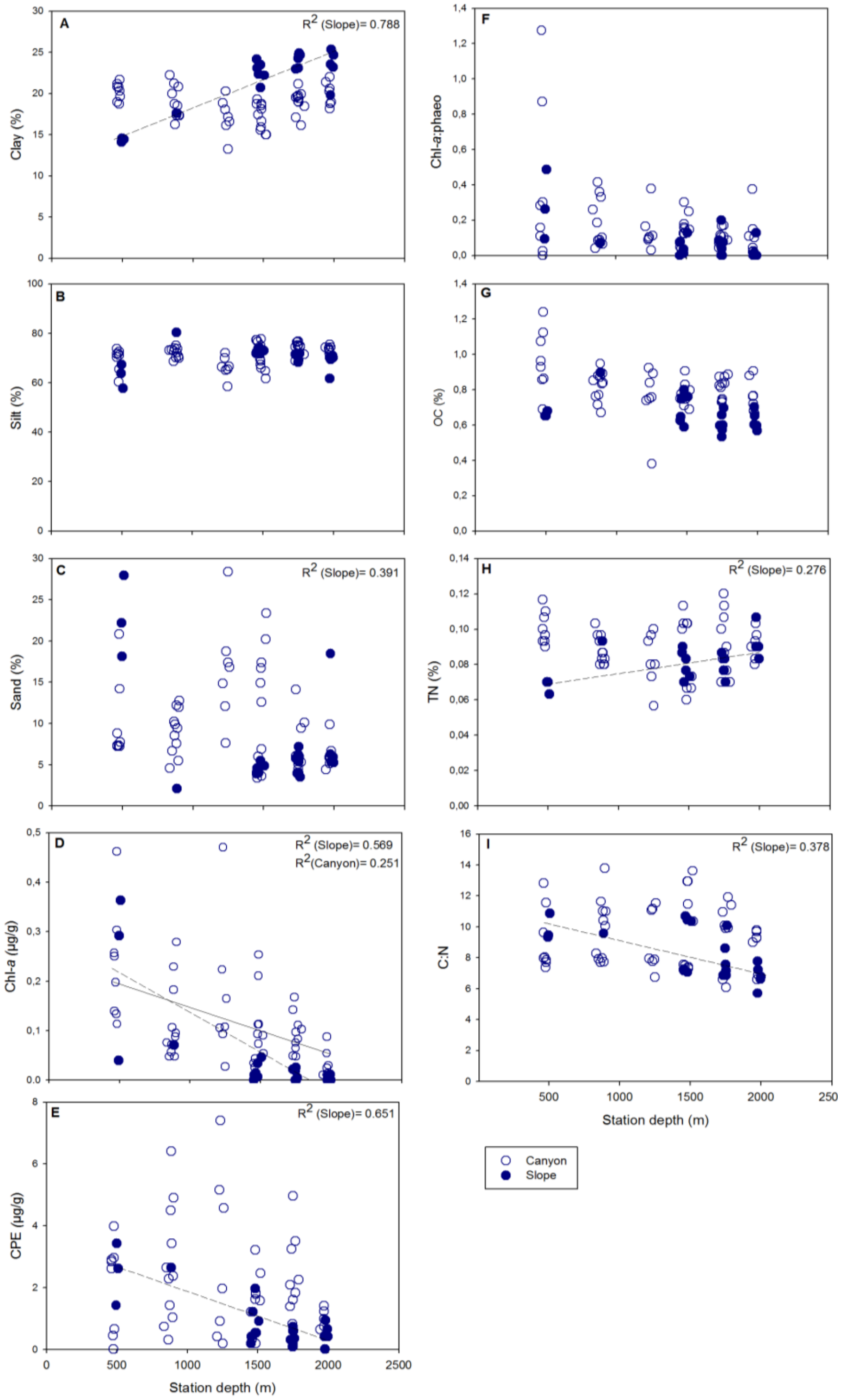


Figure 3.2. Bathymetric patterns of each environmental variable selected. Continuous and dashed lines are the significant regression ($p < 0.05$) for canyon and slope samples respectively.

Table 3.1. Mean ± standard deviation values of environmental variables for each year at each station, BC: Blanes canyon; OS: Open slope, Spr: Spring; Aut: Autumn; Clay, Silt, Sand: volume percent clay, silt and sand content; TN: total nitrogen concentration; C:N: molar carbon/nitrogen ratio; OC: organic carbon concentration; Chl *a*: chlorophyll *a*; CPE: chloroplastic pigment equivalents; Chl *a*:phaeo: chlorophyll *a* divided by its degradation products (phaeopygments) indicating "freshness" of the phytodetrital OM.

Station	Sampling period	Clay		Silt		Sand		CPE		Chl *a*		Chl *a*:phaeo		OC		TN		C:N	
		(%)	SD	(%)	SD	(%)	SD	(µg/g)	SD	(µg/g)	SD	(µg/g)	SD	(%)	SD	(%)	SD		SD
BC500	**Spr-12**	19.75	1.5	66.14	8.1	14.11	9.53	2.71	0.85	0.17	0.045	1.074	0.3	0.773	0.1	0.1	0.01	7.71	0.47
	Aut-12	19.28	0.5	73.16	0.8	7.55	0.29	4.07	1.28	0.13	0.018	0.054	0.1	0.897	0	0.113	0	7.92	0.1
	Spr-13	20.29	0	65.51	0	14.2	0	3.98	0	0.46	0	0.024	0	1.12	0	0.097	0	11.6	0
	Aut-13	21.22	0.4	71.01	0.6	7.76	0.92	2.9	0.06	0.26	0.029	0.248	0.1	1.092	0.1	0.094	0.01	11.7	2.07
BC900	**Spr-12**	21.72	0.7	70.86	3.2	7.42	3.97	2.23	1.29	0.07	0.014	0.222	0.1	0.785	0.1	0.098	0.01	7.99	0.41
	Aut-12	18.23	1.9	73.11	0.9	8.65	1.75	3.45	1.11	0.07	0.034	0.066	0	0.734	0.1	0.093	0.01	7.87	0.12
	Spr-13	17.8	0.8	71.12	1.5	11.08	2.25	3.19	1.49	0.15	0.113	0.193	0	0.871	0	0.081	0	10.7	0.57
	Aut-13	18.58	1.9	72.99	2.5	8.42	3.44	3.98	2.73	0.18	0.072	0.259	0.1	0.883	0.1	0.083	0	10.6	0.32
BC1200	**Spr-12**	16.02	3.9	62.37	5.6	21.6	9.55	1.3	1.43	0.07	0.055	0.271	0.2	0.56	0.3	0.075	0.03	7.34	0.83
	Aut-12	18.7	1.6	68.89	3.3	12.51	4.87	4.7	0.11	0.1	0.007	0.063	0	0.753	0	0.093	0.01	7.74	0.16
	Spr-13	16.9	1	67.2	2.4	15.9	3.45	5.71	1.5	0.29	0.162	0.102	0	0.886	0	0.078	0	11.4	0.2
	Aut-13	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
BC1500	**Spr-12**	17.68	1.5	71.8	8.2	10.52	9.71	3.96	1.39	0.18	0.099	0.136	0	0.755	0	0.103	0	7.36	0.08
	Aut-12	18.48	0.9	77.07	0.5	4.44	1.39	2.02	0.83	0.03	0.009	0.053	0	0.758	0	0.106	0.01	7.2	0.32
	Spr-13	16.18	2	65.09	3.6	18.73	5.53	1.94	0.47	0.12	0.082	0.191	0	0.798	0.1	0.069	0	11.6	1.74
	Aut-13	16.54	1.4	70.62	3.9	12.85	5.22	2.23	0.87	0.09	0.02	0.195	0.1	0.772	0.1	0.068	0.01	12	1.65
BC1750	**Spr-12**	20.43	1	74.99	0.3	4.58	0.73	2.16	0.46	0.07	0.008	0.084	0	0.735	0	0.107	0	6.9	0.06
	Aut-12	19.32	0.3	75.85	1.2	4.81	1.08	1.27	0.4	0.04	0.015	0.054	0	0.783	0.1	0.111	0.01	7.22	1.55
	Spr-13	17.22	1.2	71.55	2.8	11.23	2.53	2.62	0.77	0.12	0.021	0.109	0	0.861	0	0.072	0	12.1	0.72
	Aut-13	19.49	0.5	75.47	0.8	5.042	0.94	3.35	1.57	0.1	0.045	0.129	0	0.829	0	0.087	0	9.71	0.36

BC2000	**Spr-12**	18.86	0.2	74.85	0.7	6.28	0.53	0.78	0.12	0.02	0.011	0.063	0.1	0.672	0	0.1	0	6.73	0.21
	Aut-12	21.33	0.7	73.47	0.7	5.20	0.82	0.87	0.31	0.02	0.008	0.057	0	0.836	0.1	0.092	0	9.06	1.18
	Spr-13	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
	Aut-13	19.18	1.5	72.93	1.3	7.88	2.82	1.2	0.3	0.07	0.023	0.262	0.2	0.765	0	0.082	0	9.4	0.2
OS500	**Spr-13**	14.35	0	62.88	4.9	22.76	2.13	2.49	0	0.23	0.17	0.094	0.1	0.66	0	0.07	0	9.88	0.85
OS900	**Spr-13**	17.57	0	80.3	0	2.13	0	2.65	0	0.07	0	0.023	0	0.9	0	0.09	0	9.58	0
OS1500	**Aut-12**	23.57	0.3	71.96	0.3	4.46	0.16	0.38	0.18	0.01	0.005	0.01	0	0.62	0	0.09	0	7.15	0.09
	Spr-12	21.74	0.8	73.47	0.5	4.79	0.54	1.36	0.55	0.03	0.016	0.027	0	0.77	0	0.07	0	10.5	0.18
OS1750	**Spr-12**	23.82	1.4	69.78	6	6.5	0.98	0.53	0.3	0.02	0.003	0.271	0.4	0.59	0	0.09	0	6.87	0.01
	Aut-12	24.38	0.9	70.1	0.8	5.52	0.2	0.29	0.27	0	0	0	0	0.61	0	0.09	0	7.23	0.3
	Spr-13	24.66	0	71.81	0	3.53	0	0.35	0	0.01	0	0.025	0	0.7	0	0.07	0	10.1	0
	Aut-13	24.24	0	71.91	0	3.85	0	0.31	0	0.02	0	0.066	0	0.66	0	0.08	0	8.6	0
OS2000	**Spr-12**	21.65	6.9	65.95	2.3	12.4	8.62	0.28	0.3	0	0	0	0	0.653	0.1	0.09	0.01	6.73	1.46
	Aut-12	24.38	1.2	70.1	0.8	5.52	0.12	0.67	0.26	0.01	0.006	0.016	0	0.61	0.4	0.09	0	6.88	0.32

Table 3.2. Results from univariate and multivariate PERMANOVA two-way analyses for differences in sedimentary abiotic variables. Test for locations (Lo: Canyon and Slope); water depth (WD) and interaction term. MGS: Mean grain size; TN: total nitrogen concentration; C:N: molar carbon/nitrogen ratio; OC: organic carbon concentration; Chl *a*: chlorophyll *a*; CPE: chloroplastic pigment equivalents; Chl *a*:phaeo: chlorophyll a: phaeopigments ratio. Data was normalised; resemblance was calculated using Euclidean Distance. Bold values indicate significant differences at $p < 0.05$, bold italic values indicate significant differences at $p < 0.01$.

Source	df	MGS	TN	C:N	OC	CPE	Chl *a*	Chl *a*: phaeo
Lo	1	0.3014	**0.0245**	0.794	***0.0001***	***0.0022***	**0.0416**	0.7895
WD	4	***0.004***	0.4368	0.4526	***0.005***	***0.0002***	***0.0001***	0.4589
Lo x WD	4	***0.0008***	**0.0269**	0.4067	***0.0096***	**0.0347**	0.5768	0.4216
Res	66							
Total	76							

Table 3.3. Univariate and multivariate PERMANOVA two-way analyses for environmental data in the slope and canyon systems, Test for sampling periods (Tm: Spring 2012, autumn 2012, spring 2013 and autumn 2013); water depth (WD) and interaction terms. Data was normalised; resemblance was calculated using Euclidean Distance, Bold values indicate significant differences at $p < 0.05$, bold italic values indicate significant differences at $p < 0.01$.

Source	df	MGS	TN	OC	C:N	CPE	Chl *a*	Chl *a*: phaeo
Slope								
Tm	3	0.1409	**0.0146**	***0.0018***	***0.0003***	0.589	0.0645	0.7715
WD	3	**0.0149**	**0.0262**	**0.0408**	0.9999	**0.0446**	**0.0417**	0.7311
Tm x WD	2	0.1184	0.4582	0.7418	0.5727	0.2603	**0.015**	0.6779
Res	10							
Total	17							
Canyon								
Tm	3	***0.001***	***0.0001***	***0.0001***	***0.0001***	0.4549	***0.0002***	***0.0001***
WD	5	***0.0003***	***0.0028***	***0.0006***	0.2479	**0.0162**	***0.001***	***0.0001***
Tm x WD	13	0.0844	***0.0099***	0.1304	0.0938	0.1099	0.3101	***0.0001***
Res	35							
Total	56							

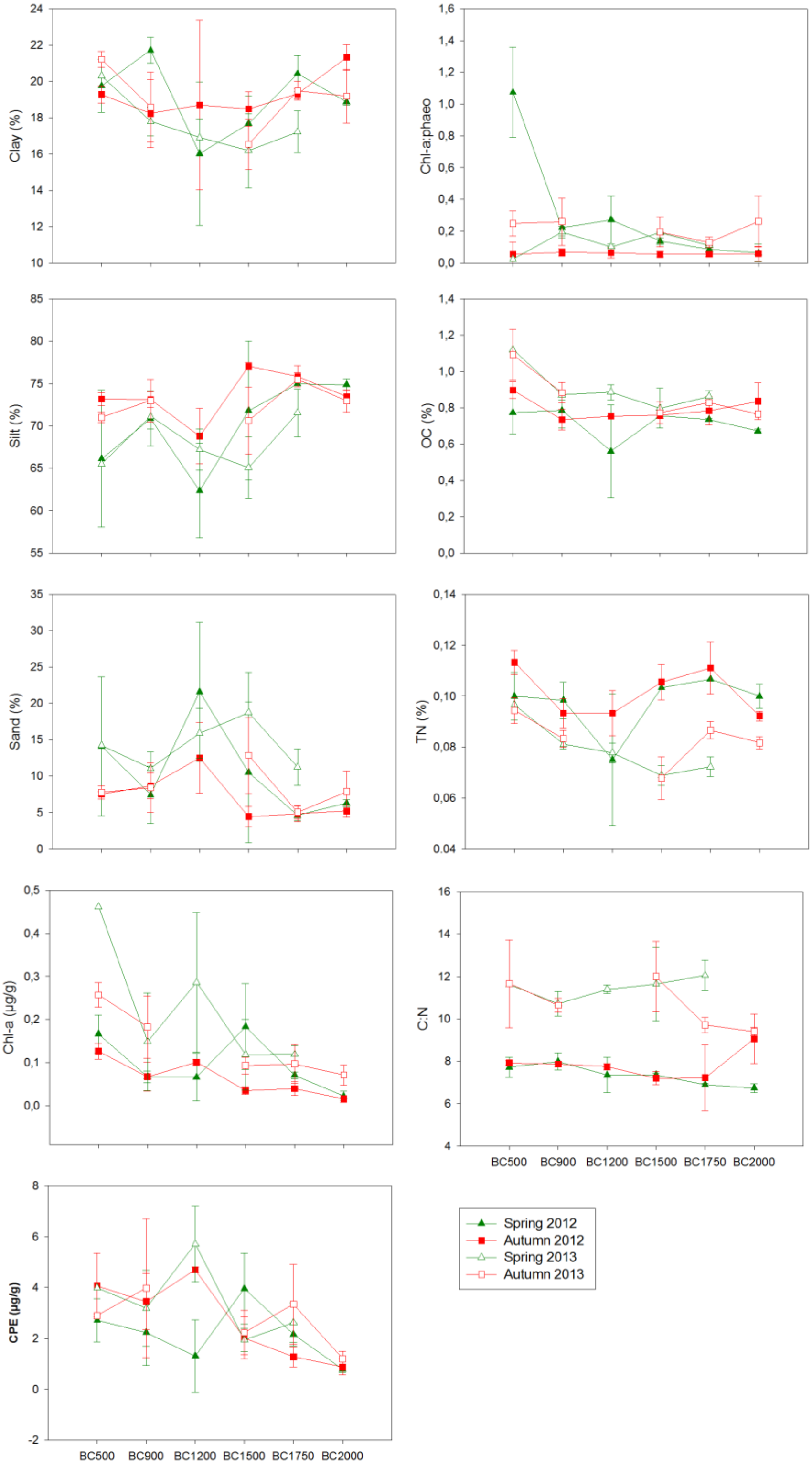


Figure 3.3. Temporal trend of mean environmental variables within the Blanes Canyon stations.

Sedimentary Chl *a* and CPE content were higher in the canyon than at the slope, except for Chl *a* at BC500 in spring 2013 (Fig. 3.2, Table 3.1, Table 3.2). Slope samples exhibited decreasing Chl *a* and CPE with increasing water depth ($R^2=0.58$ and 0.65 respectively, $p < 0.05$, Table 4; Fig. 3.2). Sedimentary Chl *a* content was different between sampling periods (Tm x WD interaction, pairwise comparisons, $p < 0.01$, Table 3.3, particularly at OS1750 between spring and autumn 2012 ($p< 0.05$, Table 3.2). CPE did not differ between sampling periods (Table 3.3). Along the canyon axis, Chl *a* decreased with increasing water depth (Fig. 3.2, $R^2=0.26$) but the high variability at BC1200 (autumn 2012 and spring 2013) and BC1500 (spring 2012) obscured this general pattern (Table 3.1, Fig. 3.3). Chl *a* differed significantly between sampling periods and water depths (PERMANOVA, $p < 0.01$, Table 3.3). As for Chl *a*, CPE decreased with increasing water depth along the canyon axis, except for the peaks observed at BC1200 (autumn 2012 and spring 2013) and BC1500 (spring 2012) (Fig. 3.3, PERMANOVA, $p < 0.01$, Table 3.3).

The "freshness" (Chl *a*: phaeo ratio) of OM did not differ significantly between the canyon and slope (PERMANOVA, $p > 0.05$, Table 3.2). At the slope, the freshness of OM did not show a clear decrease with increasing water depth (Fig. 2F) and differences between sampling periods were not significant (PERMANOVA, $p > 0.05$, Table 3.3). In the canyon, the freshness differed significantly between sampling periods and water depths, but a clear decrease with increasing depth occurred only in spring 2012 (Fig. 3.3; PERMANOVA, $p < 0.01$, Table 3.3). There were also significant differences between spring and autumn 2012 for BC500, BC900 and BC1500 and between autumn 2012 and 2013 at BC1750 and BC2000 (Tm x WD interaction, pairwise comparisons, $p < 0.01$, Table 3.3).

Sedimentary organic carbon (OC) and total nitrogen (TN) differed significantly between the canyon and slope (PERMANOVA, $p < 0.05$, Table 3.2), with the main differences occurring at 500 m depth and the 1500-2000 m stations for OC and at 500 m and 1750 m depth for TN (pair-wise comparison, $p < 0.05$, see Fig. 3.2). At the slope, OC and TN differed significantly between sampling periods (Table 3.3), particularly between autumn 2012 and spring 2013 ($p < 0.05$, see Table 2). Also TN and OC differed significantly between water depths (Table 3.3) but only TN decreased with increasing depth ($R^2= 0.27$; Fig. 3.2). Along the canyon axis, OC and TN differed significantly between sampling periods and water depths

(PERMANOVA, $p < 0.01$, Table 3.3), with a pronounced decrease with increasing water depth in spring 2013 (Table 3.1, Fig. 3.3). OC and TN were significantly higher in spring 2013 and autumn 2012 (Table 3.1, Fig. 3.3). TN also differed significantly between 2012 and 2013, particularly at BC1500 and BC1750 (Tm x WD interaction pairwise comparisons, $p < 0.01$, Table 3.4, Fig. 3.3).

Sedimentary C:N ratios did not differ significantly between canyon and slope (PERMANOVA, $p > 0.05$, Table 3.1, 3.2, Fig. 3.2). Along the canyon axis, C:N differed significantly between sampling periods (PERMANOVA, $p < 0.01$, Table 4), particularly between the years 2012 and 2013 (Table 3.1, Fig. 3.3).

Canyon and slope samples separated in the PCA (Fig. 3.4A). Furthermore, slope samples were less variable in terms of environmental variables, except for OS500. The first two PC axes explained a 69.6% of the variation. The main contributors were Chl *a* (-0.443), CPE (-0.388), Chl *a*: phaeo (-0.374) and clay (0.367) for the axis PC1 and silt (-0.562), TN (-0.468), sand (0.450) and OC (-0.396) for the axis PC2 (numbers in parenthesis represent eigenvector values).

When the Canyon axis samples were analyzed separately, the first two axes of the PCA explained 65.4% of the variation (41.5% and 23.9% for PC1 and PC2, respectively; Fig 3B). The main contributors were sand (0.427), Silt (-0.392), Chl *a* (0.379) and TN (-0.357) for PC1 and OC (0.596), Chl *a* (0.376), sand (-0.388) and CPE (0.310) for PC2 (numbers in parenthesis represent eigenvectors). Sampling periods were clearly distinguishable in the PCA plot (Fig. 3B), particularly autumn 2012 and spring 2013, in accordance with the PERMANOVA analyses (Table 3.3). In particular spring 2013 samples were characterized by having greater pigments concentrations (Table 3.1) as suggested by the PCA. No consistent bathymetric differences could be observed in the PCA plot.

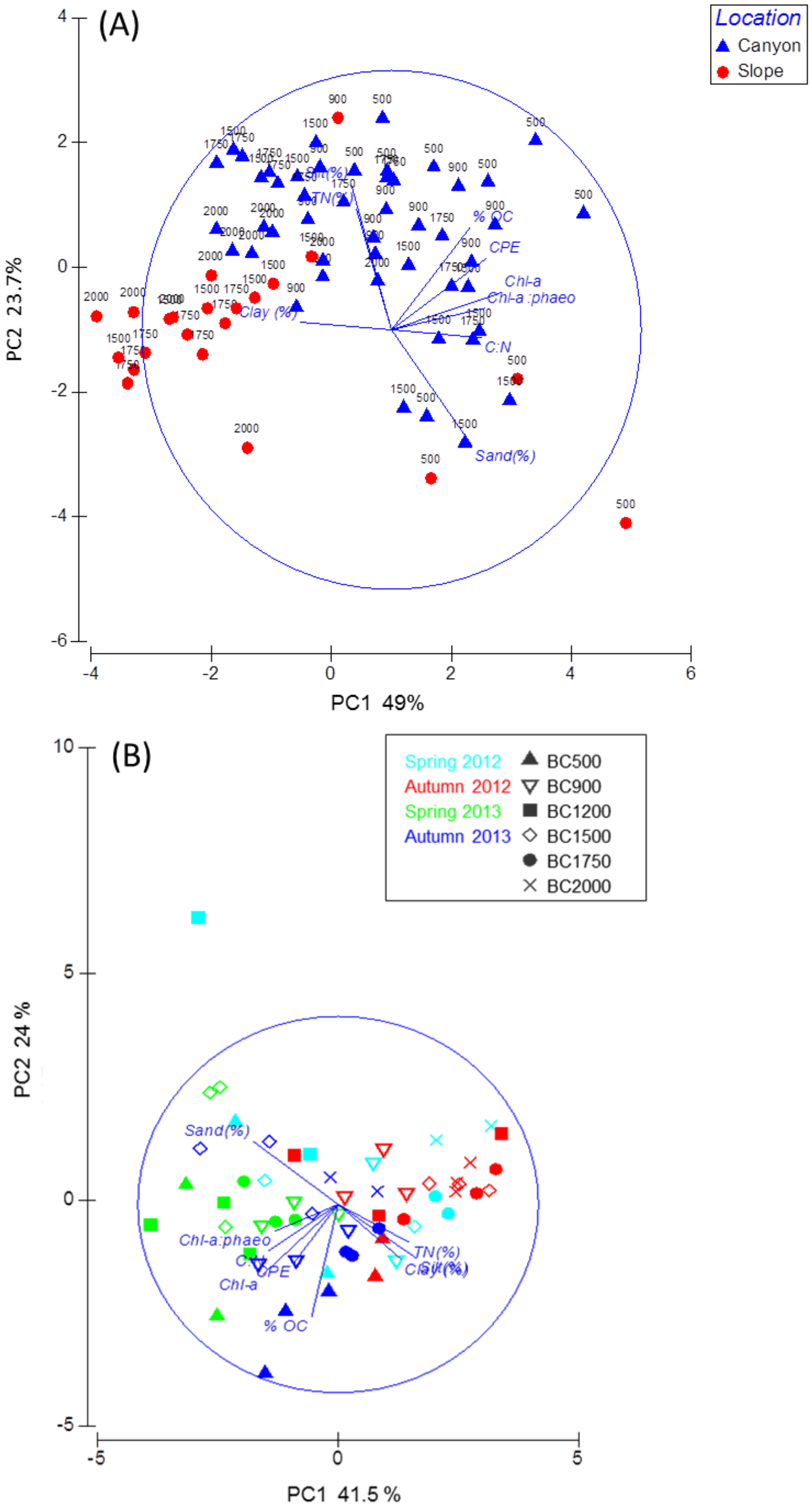


Figure 3.4. Principal component analysis (PCA) ordination based on 9 environmental variables selected. A) Canyon vs slope. B) Canyon.

3.3.2 Meiofauna density

In general, total meiofauna densities were significantly higher inside the canyon than on the slope (Table 3.4; $p < 0.01$, Table 7, Fig. 3.5), particularly during spring 2012 and autumn 2012 (Table 3.4) at 1750 and 2000 m depth (pair-wise comparison, $p < 0.05$, data not shown, see Fig. 3.5).

At the slope, there were significant differences between depths (PERMANOVA, $p < 0.01$, Table 3.5), and a density decrease with increasing water depth (R^2=0.84, p< 0.005, Fig. 3.5) was observed. There was little between-replicate variability in density at each station along the slope, in contrast to the high variability observed in the canyon (Table 3.5). The minimum slope density recorded was 209 ± 44 ind. 10 cm^{-2} at OS2000 in autumn 2012 and the maximum was 1027 ± 72 ind. 10 cm^{-2} at 500 m depth in spring 2013 (Table 3.6, Fig. 3.6B).

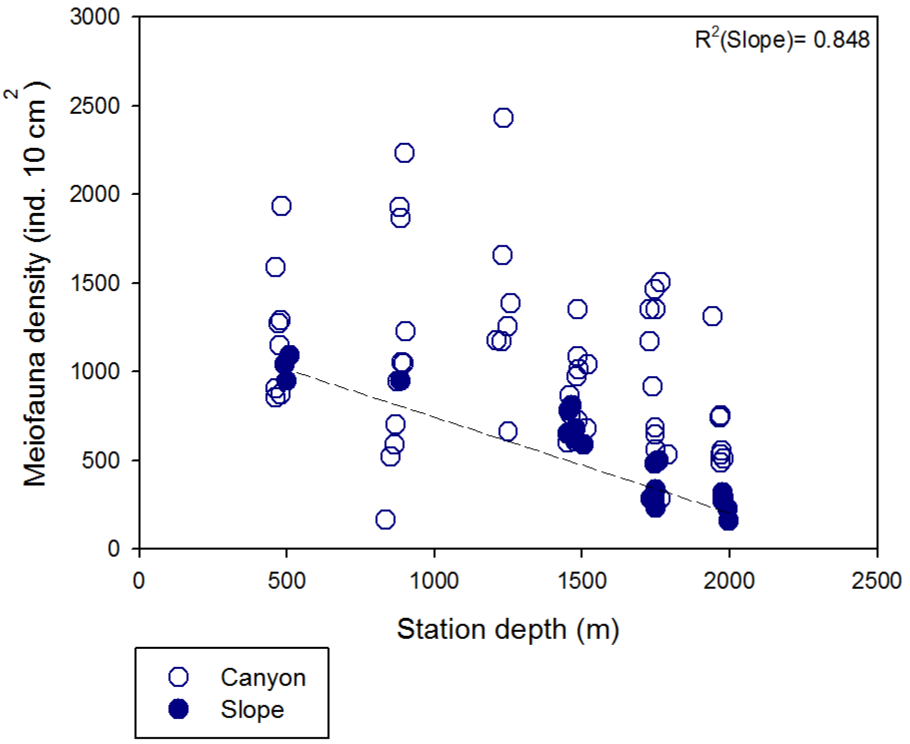


Figure 3.5. Bathymetric scatter plot of meiofaunal densities. Dashed line represent significant regression (p<0.05) for slope samples.

Table 3.4. Results of PERMANOVA two-way analyses for differences in univariate (density) and multivariate (composition) **meiofauna descriptors** among locations (Lo: canyon and slope), water depth (WD) and interaction item, ECV: estimated component of variation. Noted that the numbers of collected samples were not equal in each location, resulting different degrees of freedom (df) for interaction terms, Information about missing samples is reported in Table 1. *: Monte Carlo inferred values (PERMANOVA permutations <100).

Source	df	P(perm)	perms	ECV	P(perm)	perms	ECV
All samples		Density			Community		
Lo	1	***0.0048***	9816	14186	***0.0015***	9950	44.254
WD	5	***0.0002***	9952	19683	***0.0001***	9903	76.817
LoxWD	4	0.2167	9960	3203	**0.0263**	9902	34.775
Res	66			37.75			228.71
Total	76						
Spring 2012							
Lo	1	**0.0301**	257	30374	**0.0395**	270	203.41
WD	1	0.2523	269	0.1619	0.1971	270	22.417
LoxWD	1	**0.056**	269	3.32	0.7886	270	47.525
Res	4			0.74108			151.7
Total	7						
Autumn 2012							
Lo	1	***0.0001***	9833	51798	***0.0082***	9965	65.749
WD	2	0.0165	9964	12271	**0.0125**	9942	66.353
LoxWD	2	***0.0056***	9966	35035	0.1453	9948	52.148
Res	12			13829			224.41
Total	17						
Spring 2013							
Lo	1	0.1099	9945	13076	**0.0551**	9949	48.16
WD	3	0.476	9941	-1,0705	0.1405	9932	33.898
LoxWD	3	0.9418	9961	-45.482	0.4453	9913	35.567
Res	10			64.624			224.88
Total	17						
Autumn 2012							
Lo	1	0.722	4*	-28.614	0.6623	4*	-28.614
Res	2			44.914			44.914
Total	3						

Table 3.5. Univariate and multivariate PERMANOVA two-way analyses for meiofauna density and composition data in the slope and Blanes canyon**,** Test for sampling periods (Tm: Spring 2012, autumn 2012, spring 2013 and autumn 2013); water depth (WD) and interaction terms, Bold values indicate significant differences at p < 0.05, bold italic values indicate significant differences at p < 0.01.

Source	**df**	**P(perm)**	**perms**	**ECV**	**P(perm)**	**perms**	**ECV**
Slope		**Density**			**Community**		
Time	3	0.2364	9949	0.58963	0.0617	9919	34.852
WD	2	***0.0006***	9949	14.902	***0.003***	9937	88.6
Tm x WD	2	0.0602	9959	49.737	0.2846	9933	17.488
Res	10			40.131			
Total	17						
Canyon		**Density**			**Community**		
Time	3	**0.0234**	9957	66.955	**0.0119**	9918	18.466
WD	5	**0.0271**	9951	609712	***0.0001***	9994	77.258
Tm x WD	13	**0.0203**	9938	18.078	0.0602	9894	32.665
Res	33			31.739			192.63
Total	54						

Along the canyon axis, the densities did not show a clear bathymetric pattern, and they were characterized by high variability over the different sampling periods within and between depths (Fig. 3.5, 3.6A). The minimum density recorded was 378 ± 69 ind. 10 cm-2 at BC900 in spring 2012 and the maximum was 1763 ± 245 ind. 10 cm^{-2} at BC500 in autumn 2012 (Table 3.6, Fig. 3.6A). The highest variability was observed at BC900 and BC1200 (Fig. 3.6A). There was a significant influence of water depth at each sampling period, except in spring 2013 (PERMANOVA, p < 0.05, Table 3.5). In fact, the observed differences were caused by the densities at BC500, which were significantly higher than those at the deepest stations (BC1500, except in spring 2012, BC1750 and BC2000) (Table S1, pair-wise comparison within Tm x WD; Fig. 3.6A).

Regarding the temporal variability, at the slope stations, meiofauna densities did not exhibit significant differences at any sampling depth (PERMANOVA, p> 0.05, Table 3.5; Fig. 3.6B). In contrast, densities inside the canyon differed significantly between sampling periods (Fig. 3.6A, Table 3.5), but no clear seasonal patterns were observed. In 2012, densities were higher in autumn than in spring, (except at BC1500) while the trend was the opposite in 2013 (except at BC900) (Fig. 3.6A). Significant intra-annual differences occurred in 2012 at BC1750, with higher densities in autumn compared to spring. Significant inter-annual variability mainly occurred between autumn periods at BC500 and BC1750, where densities were higher in 2012 than 2013, and at BC900, where the opposite was observed (Table S1, pair-wise comparisons). There were no temporal differences detected at stations BC1200, BC1500 and BC2000 (Table S1, pair-wise comparison; Fig 3.6A).

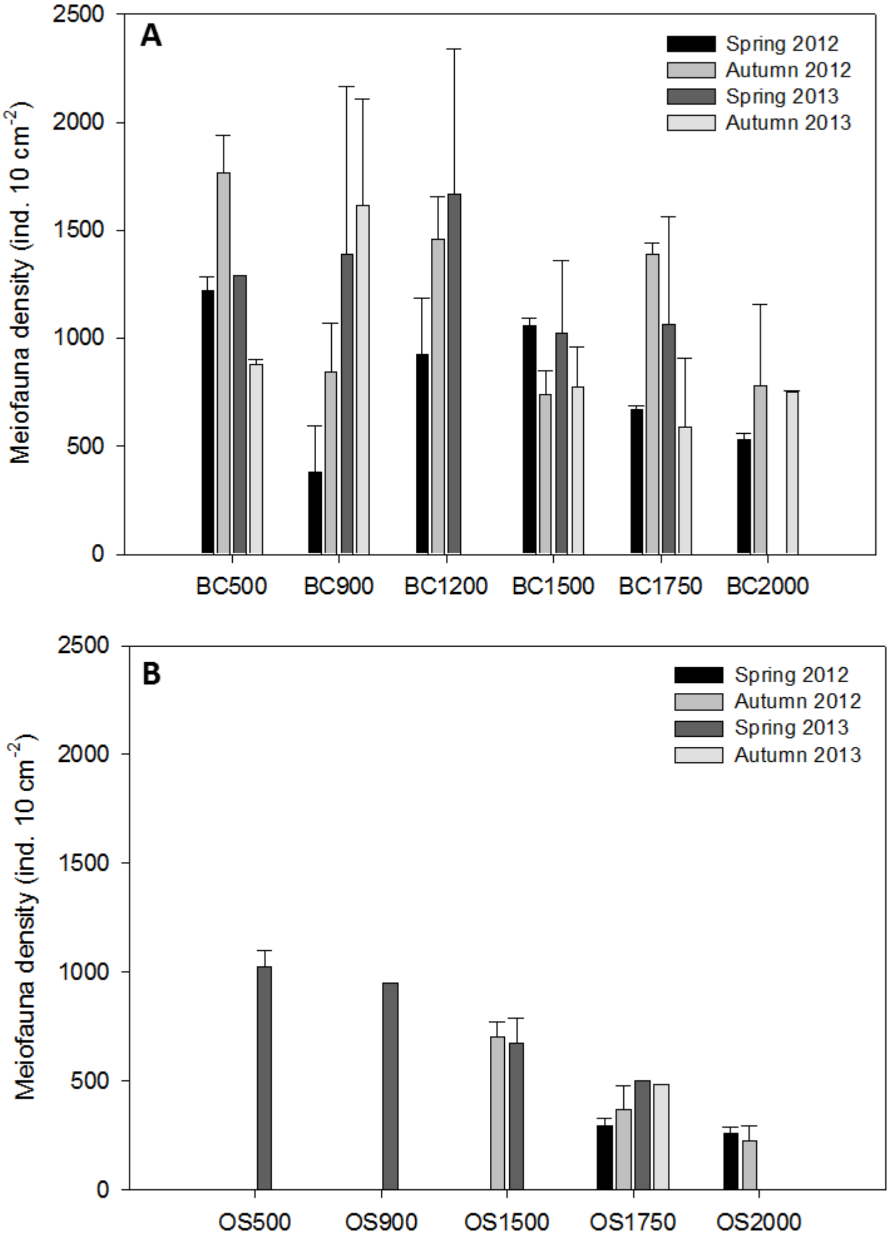


Figure 3.6. Temporal fluctuations of meiofaunal density within the A) Blanes canyon and B) Slope.

3.3.3 Meiofauna community composition

The meiofauna composition in the canyon and the slope was comparable to what is usually observed in the deep sea, with nematodes being dominant (79.2-97.6 %), followed by copepods (0.68-5.14%, mainly harpacticoids), nauplii larvae (0.65-4.9%) and polychaetes (0.51-3.12%) (Table 3.6). Other taxa (such as tardigrades and kinorhynchs) were regularly found but in low densities (less than 2%). A total of 21 major taxa were identified in the canyon, while only 16 taxa were collected at the open slope. Aplacophora, Amphipoda, Nemertea, Cumacea and Sipunculida were only present in the canyon samples (Table 3.6). Relative nematode abundance was slightly higher in all canyon samples compared to slope samples, except at the 900 m and 1750 m stations in spring 2013 and autumn 2013, respectively.

Considering all samples, meiofauna composition differed significantly between the canyon and the slope (Table 3.5, $p< 0.05$), except at the 900 m stations (Lo x WD, pair-wise comparison, $p< 0.05$). For each sampling period, differences between locations were also evident, especially in spring and autumn 2012 (Table 3.5). At the slope, meiofauna composition varied between water depths (Table 3.5). The maximum of 13 taxa was found at OS1500 in autumn 2012 and the minimum number of 5 taxa was found at OS1750 in autumn 2013, Despite of the reduced number of slope samples, the number of taxa decreased with increasing water depth in autumn 2012 and spring 2013 (Table 3.6). In the canyon, the meiofauna community structure also differed among water depths (Table 3.5), except for the BC500-BC1200, BC900-BC1200, BC1500-BC1750 and BC1750-BC2000 pairwise comparisons (Table S2). However, the MDS ordination showed that there were no clear relationships between meiofauna community structure and bathymetry, except in autumn 2013 (Fig. 3.8). The maximum number of taxa (14) was found at BC900 in autumn 2012 and spring 2013 and the minimum (6) at BC2000 in autumn 2013 (Table S1). The reduced dominance of nematodes at BC900 is countered by a relative increase of copepod and nauplii densities (Table 3.6).

The structure of the slope assemblages did not differ between sampling periods (Table 3.5), despite there was an increase in number of taxa from spring to autumn 2012 (Table 3.6). In the canyon, there were clearly significant time differences (Table 3.5), with the most significant variations occurring both in 2012 (intra-annual) and between the two autumn periods (inter-annual, 2012-2013) (Table 3.5, Table S2, pair-wise comparison). These differences were mainly owing to the increase in number of taxa from spring to autumn 2012 at all sampling stations (except at BC2000) (Table 3.6).

Table 3.6. Relative abundance of meiofaunal taxa and total density per sampling period and station. BC: Blanes canyon, OS: open slope; Std: standard deviation; S: number of taxa.

Time	**Spring 2012**							
Station	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**BC2000**	**OS1750**	**OS2000**
Relative abundance (%)								
Amphipoda	-	-	-	-	-	-	-	-
Aplacophora	-	-	-	-	-	-	-	-
Bivalvia	-	-	0.21	0.09	0.15	0.37	-	-
Cnidaria	-	-	-	-	-	-	-	-
Cumacea	-	-	-	-	-	-	-	-
Gastrotrycha	-	-	-	-	0.15	0.09	-	-
Halacaroidea	-	0.13	0.05	-	-	-	0.19	0.16
Holoturoidea	-	-	-	-	-	-	-	-
Copepoda	1.77	11.39	3.92	1.96	4.50	4.32	6.56	4.06
Isopoda	0.12	0.13	-	0.04	0.45	0.28	0.19	-
Kinorrhynca	0.50	0.79	0.21	0.14	0.22	0.18	-	-
Loricifera	-	-	-	-	-	0.01	-	-
Nauplii	1.73	4.9	2.88	1.62	2.78	3.85	5.79	3.89
Nematoda	**93.94**	**79.21**	**89.51**	**94.42**	**90.55**	**88.53**	**84.94**	**89.49**
Nemertea	-	-	-	-	-	-	-	-
Oligochaeta	-	-	-	-	-	-	-	-
Ostracoda	0.04	-	0.27	0.09	-	-	-	-
Polychaeta	1.86	3.31	2.77	1.38	1.05	1.03	1.73	1.86
Rotifera	0.04	-	-	0.23	0.15	-	0.57	0.16
Sipunculida	-	-	-	-	-	-	-	-
Tanaidiacea	-	-	-	-	-	-	-	-
Tardigrada	-	0.13	0.16	-	-	1.22	-	0.33
Total (ind. 10 cm^{-2})	**1212.5**	**377.5**	**919.5**	**1048**	**666.5**	**532**	**259**	**295**
Std	**122.4**	**69.2**	**52.1**	**111.9**	**166.4**	**104.3**	**35.3**	**32.2**
S	**8**	**8**	**9**	**9**	**9**	**10**	**7**	**7**

Table 3.6: *continued*

Time	**Autumn 2012**								
Station	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**BC2000**	**OS1500**	**OS1750**	**OS2000**
(%)									
Amphipoda	0.02	-	-	-	-	-	-	-	-
Aplacophora	-	-	-	-	-	-	-	-	-
Bivalvia	0.19	0.47	0.26	0.53	0.05	0.09	0.14	0.18	0.13
Cnidaria	-	0.03	-	-	-	-	0.04	-	-
Cumacea	-	0.07	-	-	-	-	-	-	-
Gastrotrycha	-	-	-	-	-	-	-	0.08	-
Halacaroidea	-	0.03	0.03	0.09	0.05	-	0.04	-	-
Holoturoidea	-	-	-	-	-	-	-	-	-
Copepoda	1.99	4.99	2.12	1.62	2.86	2.32	3.54	5.57	3.25
Isopoda	0.05	-	-	0.04	0.09	-	0.09	0.04	-
Kinorrhynca	0.13	0.74	0.93	0.17	0.09	-	0.09	0.08	0.13
Loricifera	-	-	-	-	-	-	-	-	-
Nauplii	2.69	3.73	1.21	0.9	2.47	1.2	2.22	3.41	2.07
Nematoda	**91.55**	**87.44**	**93.03**	**94.96**	**92.49**	**95.07**	**93.49**	**89.67**	**93.22**
Nemertea	0.05	-	-	-	-	-	-	-	-
Oligochaeta		-	0.07	-	-	-	0.04	-	0.26
Ostracoda	0.08	0.07	0.07	0.04	0.09	0.09	0.28	0.08	0.88
Polychaeta	1.9	1.78	1.99	1.35	0.84	0.51	0.56	0.9	0.15
Rotifera	0.11	0.07	0.20	0.17	0.21	0.38	0.18	0.35	0.57
Sipunculida	-	0.15	-	-	-	-	-	-	-
Tanaidiacea	-	0.03	0.07	0.04	-	-	-	0.04	-
Tardigrada	-	0.30	-	0.04	0.07	0.33	0.18	0.61	0.88
Total	**1763**	**841**	**990.3**	**741.3**	**1389.3**	**777.3**	**704.6**	**371.3**	**225.3**
Std	**244.6**	**280.8**	**831.5**	**132.6**	**64.7**	**461.9**	**68.2**	**107.6**	**66.5**
S	**11**	**14**	**11**	**12**	**11**	**8**	**13**	**12**	**10**

Table 3.6: *continued*

Time	**Spring 2013**								
Station	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**OS500**	**OS900**	**OS1500**	**OS1750**
(%)									
Amphipoda	-	0.02	-	-	-	-	-	-	-
Aplacophora	-	-	-	0.02	-	-	-	-	-
Bivalvia	0.07	0.23	0.12	0.12	0.25	0.16	0.42	0.04	-
Cnidaria	-	-	-	-	-	-	-	-	-
Cumacea	-	-	-	-	0.02	-	-	-	-
Gastrotrycha	-	0.16	0.04	-	-	0.06	0.10	-	1
Halacaroidea	-	0.09	0.01	-	0.02	0.06	-	0.04	-
Holoturoidea	-	-		-	-	-	-	-	-
Copepoda	1.16	5.14	1.58	0.68	2.05	2.43	1.27	2.77	4.59
Isopoda	-	0.05		0.02	0.21	-	-	0.04	-
Kinorrhynca	0.46	0.79	0.55	0.02	0.12	0.41	0.73	0.19	0.36
Loricifera	-	-	-	-	-	-	-	-	-
Nauplii	1.09	4.73	2.46	0.78	1.59	5.48	3.79	3.36	4.39
Nematoda	**94.88**	**86.07**	**93.71**	**97.65**	**94.32**	**89.25**	**91.35**	**92.38**	**88.22**
Nemertea	-	-		-	-	-	-	-	-
Oligochaeta	-	-	0.04	-	0.02	-	-	-	-
Ostracoda	-	0.25	0.01	-	-	0.22	0.31	0.19	2
Polychaeta	2.17	1.78	1.34	0.59	0.90	1.19	0.94	0.74	0.99
Rotifera	0.15	0.07	0.01	0.02	0.09	0.19	0.21	0.10	0.79
Sipunculida	-	-	-	-	-	-	-	-	-
Tanaidiacea	-	0.02	0.04	-	-	0.12	0.31	-	-
Tardigrada	-	0.54	0.04	0.02	0.40	0.36	0.52	0.10	-
Total	**1289**	**1387.6**	**1663.3**	**1022.7**	**1069**	**1027**	**948**	**674**	**501**
Std	**-**	**785.1**	**671.9**	**335.8**	**499.1**	**72**	**-**	**117.1**	**-**
S	**7**	**14**	**13**	**10**	**12**	**12**	**11**	**11**	**8**

Table 3.6: *continued*

Time	**Autumn 2013**					
Station	**BC500**	**BC900**	**BC1500**	**BC1750**	**BC2000**	**OS1750**
(%)						
Amphipoda	-	-	-	-	-	-
Aplacophora	-	-	-	-	-	-
Bivalvia	0.22	0.22	0.26	0.17	-	-
Cnidaria	-	-	-	-	-	-
Cumacea	0.07	-	-	-	-	
Gastrotrycha	0.03	0.06	0.03	-	-	-
Halacaroidea	-	-	-	-	-	-
Holoturoidea	-	-	-	-	-	-
Copepoda	4.41	3.99	1.39	2.00	2.01	4.54
Isopoda	0.19	0.04	0.03	0.17	-	-
Kinorrhynca	0.94	0.70	0.03	0.05	-	-
Loricifera	-	-	-	-	-	-
Nauplii	0.65	3.00	0.87	1.59	1.80	2.47
Nematoda	**89.85**	**90.19**	**96.35**	**94.32**	**95.19**	**90.09**
Nemertea	-	-	-	-	-	-
Oligochaeta	-	-	-	-	-	-
Ostracoda	0.45	0.01	-	0.05	-	-
Polychaeta	3.12	1.34	0.82	0.90	0.53	1.03
Rotifera	-	-	0.03	0.11	-	-
Sipunculida	-	-	-	-	0.06	-
Tanaidiacea	-	-	-	-	-	-
Tardigrada	0.03	0.32	0.13	-	-	1.03
Total	**877**	**1613.3**	**768**	**588**	**748**	**484**
Std	**25.5**	**487.6**	**195.6**	**318**	**9.8**	**-**
S	**11**	**10**	**10**	**10**	**4**	**5**

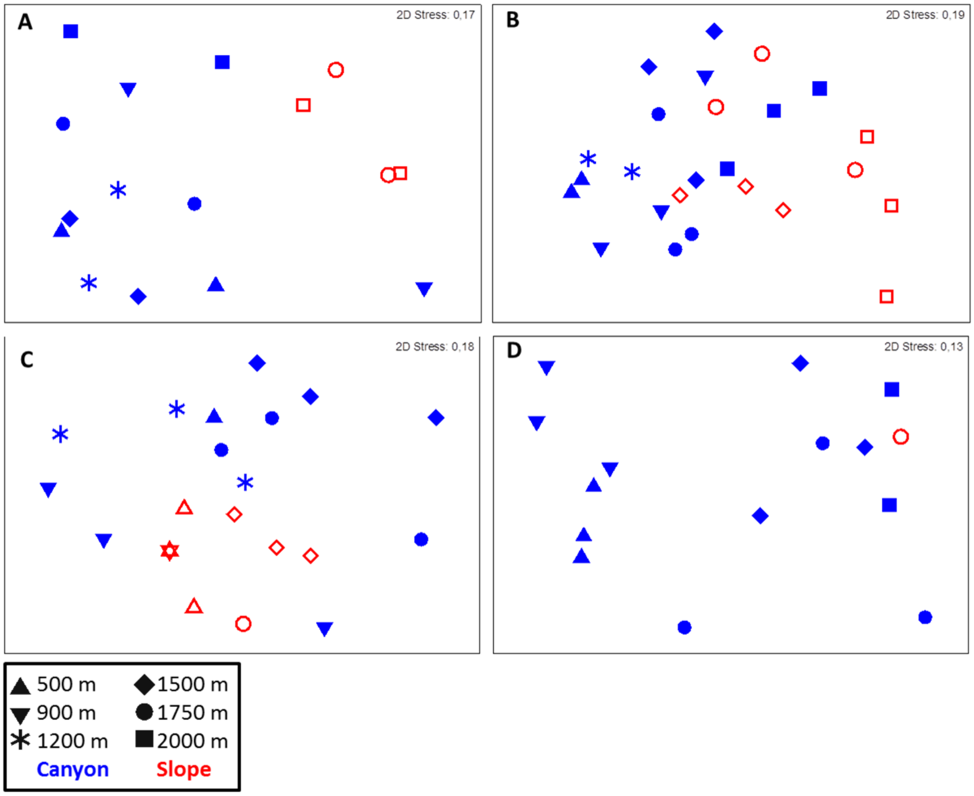


Figure 3.7. MDS based on meiofaunal community composition in the Blanes canyon and at the western slope for each sampling period (based on Bray Curtis similarity values of four-root transformed data). A) Spring 2012. B) Autumn 2012. C) Spring 2013. D) Autumn 2013

3.3.4 Relationship between meiofauna and environmental variables

Meiofaunal densities showed significant, positive correlations with the variables representing food inputs (i.e. CPE, Chl *a*, and OC) both in the canyon and at the slope, but correlations were higher for the slope than in the canyon (Spearman correlation, Table 3.7). At the slope, negative correlations between meiofauna densities and TN and clay were found, while a positive correlation with Sand occurred along the canyon axis.

Environmental variables all together explained 43% and 18% of the observed variation in meiofauna community structure in the slope and in the canyon, respectively (Table S3, S4, DISTLM), with the main contributor being Chl *a*, (24% and 9%, respectively). Other variables significantly contributing were clay, silt, OC, C:N and CPE at the slope, and clay, sand, TN, CPE and Chl *a*: phaeo in the canyon.

Table 3.7. Spearman correlation between environmental variables and meiofauna density for canyon and slope systems. Bold values indicate significant differences at $p <$ 0.05, bold italic values indicate significant differences at $p <$ 0.01.

Environmental	**Meiofauna density**	
variables	Canyon	Slope
CPE	***0.438***	***0.569***
Chl *a*	***0.474***	**0.541**
Chl *a*:phaeo	-0.059	**0.474**
OC	***0.367***	***0.591***
TN	0.013	**-0.465**
C:N	0.210	0.591
Clay	-0.253	***-0.600***
Silt	-0.201	0.147
Sand	**0.297**	-0.108

3.4 DISCUSSION

As was commented in the **Chapter 1**, the Mediterranean is considered to be an oligotrophic sea, a characteristic which is often used to explain its typically low deep-sea meiofauna densities compared to similar depths in other oceans (Soltwedel, 2010; Bianchelli et al., 2010; Pape et al., 2013b). Mediterranean meiofauna densities range between 4 and 1497 ind. 10 cm-^{2}, with a clear decrease from west to east (Soltwedel, 2000; Bianchelli et al., 2010) which has been linked to the W-E gradient of increasing oligotrophy (Danovaro et al., 1999; Gambi and Danovaro, 2006; Danovaro et al., 2008). Similar low density ranges have only been reported in the Southern Pacific Ocean, Southern Atlantic Ocean and Artic Ocean (see Bianchelli et al., 2010 for a review).

The maximum densities in the Blanes Canyon axis were higher than those observed in other Mediterranean canyons (Table 3.8). Minimum densities were equivalent to those of Cape of Creus Canyon, but about 5 to 8 times higher than in the remaining canyons, even when compared to previous data from Blanes Canyon (Romano et al., 2013b) (Table 3.8). The study by Romano et al. (2013b) included only a few canyon stations and a limited depth range, with samples from the eastern and western canyons flanks (900 m depth), from the canyon axis (1600 m depth), and from the eastern open slope (1600 m depth). In the present study, a much more comprehensive range of samples were obtained, including different seasons and years, and covering the canyon axis over a wide bathymetric gradient, whilst also including stations from the adjacent western open slope, allowing a broader temporal and spatial assessment of the meiofauna in the Blanes Canyon system.

3.4.1 Spatial and temporal distribution of the meiobenthos in the Blanes Canyon system

Contrary to the traditional perception that the whole of the deep-sea is a stable environment (e.g. Sanders, 1968), marked temporal variations of meiofaunal organisms have been observed in submarine canyons (De Boveé et al., 1990; Romano et al., 2013b; Ramahlo et al., 2014; Rumolo et al., 2015) and slope systems (e.g., Hoste et al., 2007; Romano et al., 2013b; Guidi-Guilvard et al., 2014). Such variations generally suggest contrasting scenarios between spring-summer and autumn-winter, with the former causing higher densities. Traditionally, this variability has often been associated to the seasonal trends in phytodetrital food availability, but it could also be associated with specific particulate matter transport mechanisms, particularly in submarine canyon environments (Pusceddu et al., 2013; Ramalho et al., 2014).

Previous studies in the Blanes Canyon have revealed high variability in particle fluxes, driven by a variety of causes: storms, dense water formation (by dense shelf water cascading and open sea convection), dust inputs, phytoplankton blooms, and bottom trawling (Lopez-Fernandez et al., 2013). The concatenation of such events influences sedimentary dynamics and ultimately meiofauna communities by altering sediment characteristics and food availability. Physical disturbances occurring in submarine canyons sediments are known to drive the composition of meiobenthic communities (Aller, 1997; Garcia et al., 2007; Ingels et al; 2009; Levin et al., 2012; Romano et al., 2013b).

During the study period, temporal differences in meiofauna density and community composition have been observed mainly in the Blanes Canyon axis, with slope sediments showing much lower meiofaunal variability. Despite the potential bias caused by the reduced number of slope samples in our study, our results are consistent with observations by Romano et al. (2013b). In their study, meiofaunal densities at 800 and 1600 m depth on the eastern slope reflected a less dynamic environment compared to canyon stations between spring and autumn at those depths. Additionally, in our study, meiofauna densities and composition also differed between sampling years inside the canyon (Table 3.5). These differences seemed to be related to the observed increase of primary productivity in spring 2013 (Fig. 3.8A), which occurred on the slope, but was particularly reflected in the canyon sediments, where the sedimentary Chl *a* content in 2013 was much higher than in 2012. Significant positive correlations between meiofaunal densities and Chl *a* support this observation (Table 3.7). The C:N ratios showed a similar pattern, with increased values in 2013 compared to 2012 (Table 2), which is likely related to greater terrestrial runoff from the rivers in 2013 (Fig. 3.8B) combined with the resuspension caused by the high waves registered during spring 2013 (Fig. 3.8C). However, the responses of the meiofaunal assemblages were not homogeneous along the studied bathymetric range; likely an indication of other factors that can alter food availability in the canyon-slope sediments and ultimately meiofauna densities and community composition

Table 3.8: Comparison of mean meiofaunal densities with depths in different submarine canyon areas (Iberian and Western Mediterranean coasts).

Location	**Canyon**	**Depth-range (m)**	**Density (ind.10 cm^{-2})**	**Source**
NW Mediterranean	Cassidaigne	245-810	245-801	Vivier (1978)
NW Mediterranean	Gulf of Lions (several)	672-2300	36-1005	De Bovée (1990)
NW Mediterranean	Gulf of Lions (several)	830-1380	530-1290	Grémare et al. (2002)
NW Mediterranean	Lacaze-Duthiers	600-1300	836-1050	Danovaro et al. (1999)
NW Mediterranean	Lacaze-Duthiers	434-1497	205-1391	Bianchelli et al. (2010)
NW Mediterranean	Cap de Creus	960-1874	147-597	Bianchelli et al. (2010)
NW Mediterranean	Blanes	400-1600	25-1500	Romano et al. (2013b)
NW Mediterranean	Blanes	500-2000	209-1763	**Present study**
W Mediterranean	Buscarró	600-800	40-123	Rumolo et al. (2015)
Atlantic	Cascais	445-2100	492-900	Bianchelli et al. (2010)
Atlantic	Nazaré	332-1121	9.9-236.5	Garcia et al. (2007)
Atlantic	Nazaré	458-897	747-1484	Bianchelli et al. (2010)

For instance, the food signals detected in the sediments (in terms of CPE and Chl *a*, see Table 2), together with the sand content (Table 3.1), were highly heterogeneous in the upper canyon region and tended to be higher at BC1200 (Fig. 3.3). In this area, canyon flank tributaries (see Lastras et al., 2011) connect the canyon axis with some of the most frequently trawled grounds along the canyon rims (Fig. 3.1). As previously mentioned, Blanes Canyon and its adjacent open slopes comprise fishing grounds that have been subjected to persistent bottom trawling down to 800 m depth (Company et al., 2008; Sardà et al., 2009; Ramirez-Llodra et al., 2010). The main effort concentrates along the northern open slope from late winter to early summer and over the eastern canyon rim from late summer to mid-winter (Company et al., 2008; Sardà et al., 2009; Ramirez-Llodrà et al., 2010). Data from moored sediment trap data in Blanes Canyon, recorded increase in particle fluxes (mostly lithogenic) recorded at 900 and 1200 m depth in the canyon axis (mainly in summer) that was attributed to the formation of sediment resuspension clouds by bottom trawling activities (Lopez-Fernandez et al., 2013). Our results show major differences in sediment composition (coarser) and meiofaunal densities (higher) at BC900 and, particularly, at BC1200 (Table 3.1; Figs 3.3, 3.6A), which interrupts the expected distribution patterns under normal slope conditions. Altogether, these observations indicate the possible presence of an anthropogenic depocenter (i.e. a preferential area of sediment accumulation by trawling) at these depths in the Blanes Canyon axis in a similar way to the ones previously reported in the nearby La Fonera Canyon, Arenys and Besòs submarine canyons (Martín et al., 2008; Puig et al., 2015a,b; Fig. 3.1). Recent observations on deposition rates in the Blanes Canyon have revealed high sediment deposition rates at 900 and 1,200 m depth, being ca. 2 and 1 cm·year^{-1}, i.e., triple the natural sedimentation rates (Paradis, personal communication). Therefore, we suggest that the processes involved in the formation of sedimentary deposits caused by bottom trawling activities may be an important driver for meiofaunal assemblages in canyon axes environments. Contrary to the negative direct effects of trawling over fishing grounds in canyon flanks (Pusceddu et al., 2014), the increased levels of OM around sedimentary depocenters are likely beneficial to the organisms living in the canyon axis, including the meiofauna. For instance, in La Fonera Canyon the burrowing echinoid *Brissopsis lyrifera* (Forbes, 1841) colonized and proliferated in the lower canyon axis (Mecho et al., 2014) following the formation of an anthropogenic deposit of fine-grained material (Puig et al. 2015a).

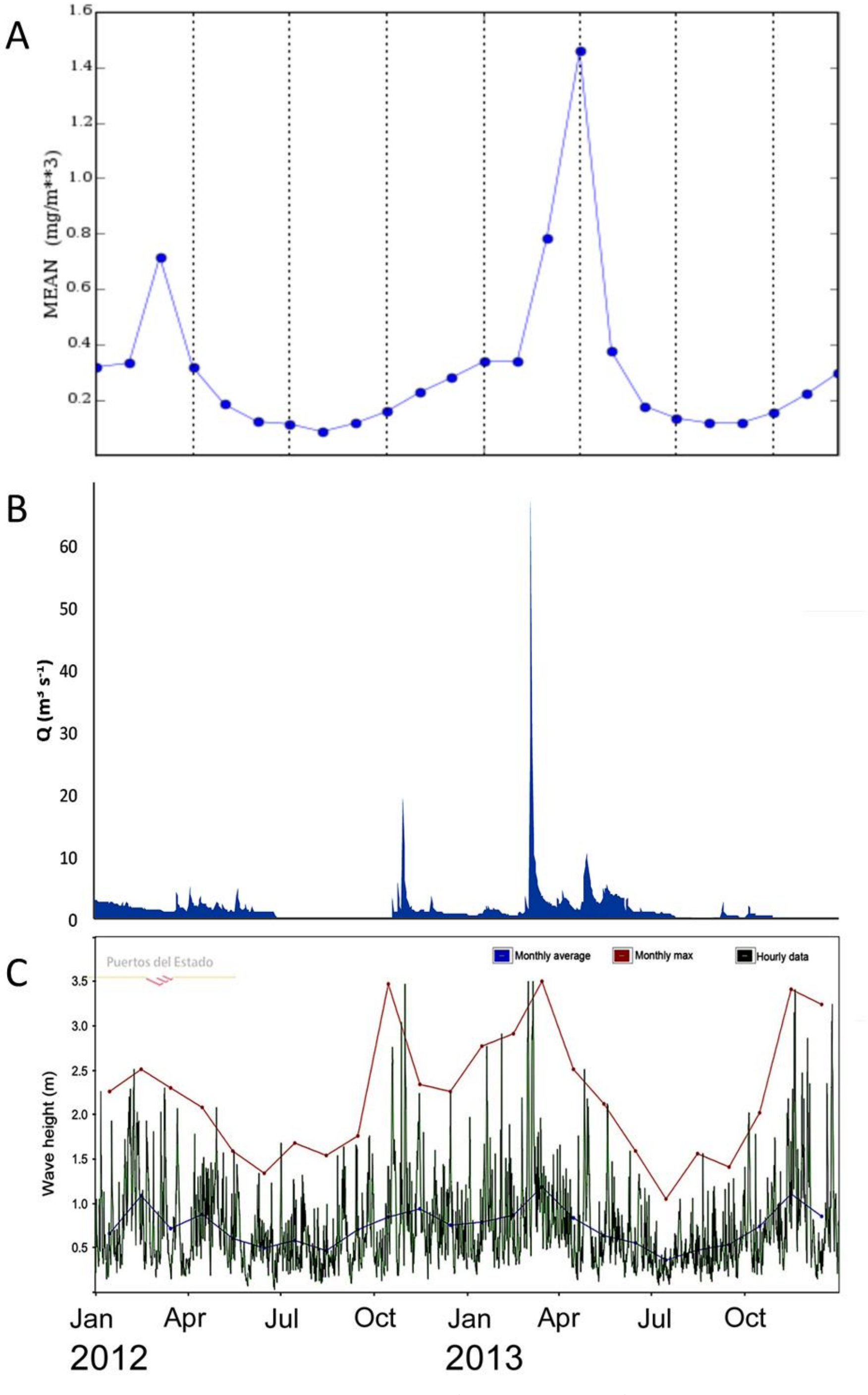


Figure 3.8. Temporal variation of ancillary data in the study area. A) Chlorophyll-a concentration /surface productivity. B) Temporal variation of high waves in the study area. Lines indicate monthly average (blue), monthly max (red) and hourly data (green). C) Monthly discharge of the Tordera river measured at the gauging nearest station to river mouth (data from Agència Catalana de l´Aigua, ACA).

The relatively low densities detected at BC500, BC900 and BC1200 in the canyon axis in spring 2012 (Fig. 3.6A), however, seem to correspond to effects of a major (i.e. deep) dense shelf water cascading event at the NW Mediterranean margin in winter 2012 (see Durrieu de Madron et al., 2013, and **Chapter 2** section **2.1.2** for further information). Such oceanographic process might have generated strong down-slope currents causing mechanical removing or burying of resident organisms. Consequently, this may lead to massive dispersal, growth inhibition, or even mortality of the meiofaunal component as has been previously reported in the Cap de Creus Canyon during the major deep cascading event in 2005 (Pusceddu et al., 2013). After the cascading event, the densities and number of taxa in autumn 2012 were much higher than in the previous spring, and also than those in autumn 2013, as dense shelf water cascading was less intense in 2013 and had almost no impact in Blanes Canyon (Anna Sanchez-Vidal, personal communication).

The meiofaunal communities in the deeper areas of the canyon, particularly at BC1500 and BC2000 seemed to be more stable over time (Fig. 3.6A) where the effects of the anthropogenic (i.e. trawling) and natural (i.e. cascading) disturbances are likely to be much reduced. Down to the deepest canyon region, further away from the sources of anthropogenic disturbance (i.e. at 2,000 m depth), the trawling influence is reduced and the actual sedimentation rates do not differ from the natural ones (Paradis, personal communication). Conversely, we could not find a straightforward explanation for the relatively high variability observed at BC1750, highlighting the complexity of the biological, chemical and physical interactions driving the spatial and temporal variability within the meiofaunal assemblages in submarine canyon environments (Ingels et al., 2013; Ramalho et al., 2014). However, it has been also reported recently that the enhanced sedimentation rates also occurred at 1,800 m depth. In fact, while the natural accumulation rates would be close to 0.18 cm·year-1 at 1800 m, the actual were measured as 0.65 cm·year-1. This observed increase is suggested to be entirely attributable to the resuspension caused by trawling activities (Paradis, personal communication) and lateral downward transport through the canyon gullies (Lastras et al., 2011). This can be related with the observed variability at BC1750.

3.4.2 Slope *vs.* Canyon

Decreasing meiobenthic densities with increasing water depth has been postulated as a basic principle in deep-sea ecology (e.g., Thiel, 1983; Tietjen, 1992), and has been linked to the bathymetric decrease in organic matter supply (POC flux) and the increasing distance from land (e.g. Soetaert et al., 1991; Danovaro et al., 1995; Gooday et al., 1996; Fabiano and Danovaro, 1999; Soltwedel, 2000; García et al.,

2007, García and Thomsen, 2008). However, as previously mentioned, such a depth-density relation may be modified by the presence of submarine canyons owing to their topographical and hydrographic heterogeneity. Canyons induce modifications in the hydrodynamic and biogeochemical processes of channeling organic matter towards the deep basin. Food availability and meiofauna density in canyons are generally higher compared to canyon interfluve areas at similar depths (De Bovée, 1990; Soetaert et al., 1991; García and Thomsen, 2008; Koho et al., 2008; Ingels et al., 2009; Gambi and Danovaro, 2016; Amaro et al., 2016; Ingels et al., 2011) and Blanes Canyon is no exception in this context.
Mediterranean canyon and slope habitats seem to lack consistent bathymetric patterns, especially in the western basin (Bianchelli et al., 2010; Pusceddu et al., 2010). In Blanes Canyon, significant decreases in meiofaunal densities with depth were reported during autumn, while the spring trends were markedly more fluctuant (Romano et al., 2013b). This bathymetric pattern, however, was not uniform as it was not clearly observed at the eastern open slope adjacent to the canyon.

Our results revealed a bathymetric decrease of meiofaunal density at the slope, but not in the canyon (Fig. 3.5). For instance, in the canyon axis, meiofauna was less abundant at BC900 than at the deeper stations (BC1500, BC1750 and BC2000) in spring 2012, but it was higher in autumn 2013 (Fig. 3.6A), likely as a result of the combined canyon heterogeneity and the oceanographic and anthropogenic drivers favoring a higher variability along the axis (see section **3.4.1**). It was clear that densities inside the canyon were higher than those at the western slope, and this was particularly evident for the deepest stations (BC1500, BC1750 and BC2000). However, the deep canyon stations exhibited high temporal meiofauna density variability, almost double the variability observed for the slope (i.e. 31% vs. 16.6% on average, respectively). These results provide further evidence that a heterogeneous canyon environment can lead to meiofaunal assemblages with highly variable distributional patterns in space and time, whilst slope environments tend to be more stable as evidenced by the uniformity of meiofauna density and community structure at the western (present study) and eastern slope (Romano et al., 2013b).

We observed a decrease in sedimentary food sources (e.g. phytopigments) with increasing depth along the slope; a pattern which agrees with previous findings (Soetaert et al., 1991; Koho et al., 2008; Ingels et al; 2009; among others) and is likely related to the dynamics of the particle fluxes in Blanes Canyon (Zúñiga et al., 2009; Lopez-Fernandez et al., 2013). Moreover, sedimentary phytopigment values were much lower at the slope than in the canyon, which supports the view of canyons playing important roles in catching and channeling organic inputs (Fig. 2). A similar depth-related pattern was observed for meiofauna density and composition (Table 3.6), which show low temporal variation and positive correlations with food sources (Chl *a*, CPE, Chl *a*: phaeo), suggesting a causal relationship.

In the canyon, the high variability within and between depths along the axis, both in terms of meiofauna density and sediment variables (Table 3.1; Fig. 3.6A) prevent us from finding a clear relationship. This is likely an indication that meiofaunal distribution can only be partly explained by the variability in food availability under canyon settings. The observed variability can, in turn, be traced back to hydrodynamic forcing, the topographic heterogeneity and/or physical anthropogenic impacts driving the canyon system (Levin et al., 2012; Gambi et al., 2010. Pusceddu et al., 2014).

Sediment grain size is known to be a key actor in driving meiofauna distributions (Giere et al., 2009). In the slope, sample sediments tend to be coarser in the shallow parts compared to the deeper parts, and meiofauna showed a strong negative correlation with clay content (Fig. 3.2). An increase in the fine sediment fractions leads to more compacted sediments, causing a reduction in the interstitial space available for meiofaunal organisms. In the canyon, sand content was positively related with meiofauna density (Table 3.7). All canyon axis samples (except the shallowest one at BC500) contained coarser sediments than those at the slope presumably caused by the local sedimentological and hydrological conditions and was reflected in the higher meiofauna densities. Grain size trends illustrate the physical dynamism and variable conditions along the axis in the Blanes Canyon, except at BC2000, where the conditions appear to be similar to those in the slope.

Slope and canyon systems clearly differ in meiofaunal composition (Table 3.4) with more rare taxa (i.e., other than nematodes, copepods, nauplii and polychaetes), and generally in greater numbers in the canyon than at the slope. This stands in contrast to the study by Gambi et al. (2010) but supports several other studies that claim submarine canyons to be hotspots of benthic biodiversity and biomass in the deep-sea at least in terms of rare meiofaunal taxa (e.g. Danovaro et al., 1999; Gili et al. 2000; De Leo et al., 2010; Ramírez- Llodra., et al., 2010; Vetter et al., 2010).

3.5 SUPPLEMENTARY DATA

Table S3.1. Results of the two-way PERMANOVA pair-wise test (Factor "Time"(Tm) with 4 levels (spring, autumn 2012, spring, autumn 2013), factor "WD ": Water Depth, with 6 levels (500, 900, 1200, 1500, 1750 and 2000 m)) for **meiofauna density** in Blanes Canyon. Data was square-root transform and resemblance was calculated using Euclidean Distance prior to analysis, Bold values indicate significant differences at $p < 0.05$, bold italic indicate significant at $p < 0.01$.

Pair-wise test		**Comparisons**	**t**	**P(perm)**	**perms**	**P (MC)**
Between Time		spring 2012 x autumn 2012	34.554	***0.006***	9956	
		spring 2012 x spring 2013	23.282	**0.036**	9967	
		spring 2012 x autumn 2013	14.303	0.185	9958	
		autumn 2012 x spring 2013	0.22509	0.818	9989	
		autumn 2012 x autumn2013	15.977	0.135	9967	
		spring 2013 x autumn 2013	12.745	0.209	9967	
Between WD		500, 900	1.92	0.098	9997	
		500, 1200	0.81684	0.456	9997	
		500, 1500	4.4243	***0.001***	9999	
		500, 1750	3.5292	***0.006***	9999	
		500, 2000	6.8515	***0.003***	9995	
		900, 1200	2.2433	**0.053**	9998	
		900, 1500	0.23221	0.833	9997	
		900, 1750	0.75359	0.479	9998	
		900, 2000	0.81258	0.451	9997	
		1200, 1500	2.0974	0.072	9995	
		1200, 1750	1.9129	0.076	9997	
		1200, 2000	2.6892	**0.04**	9998	
		1500, 1750	0.77529	0.43	9996	
		1500, 2000	1.8568	0.106	9998	
		1750, 2000	0.47584	0.675	9997	
Within WD	500	spring 2012 x autumn 2012	3.9539	0.341	3	0.088
		spring 2012 x spring 2013	0.66479	0.664	3	0.609
		spring 2012 x autumn 2013	7.4734	0.098	10	***0.002***
		autumn 2012 x spring 2013	2.2708	0.632	3	0.343
		autumn2012 x autumn 2013	10.867	0.103	10	***0.006***
		spring 2013 x autumn 2013	9.4727	0.268	4	**0.011**
	900	spring 2012 x autumn 2012	1.6721	0.293	10	0.181

		spring 2012 x spring 2013	1.8831	0.196	10	0.158
		spring 2012 x autumn 2013	3.7164	0.107	10	**0.038**
		autumn 2012 x spring 2013	1.261	0.298	10	0.277
		autumn 2012 x autumn2013	2.9272	0.11	10	**0.042**
		spring 2013 x autumn 2013	0.48187	0.684	10	0.657
	1200	spring 2012 x autumn 2012	1.3043	0.325	3	0.266
		spring 2012 x spring 2013	1.3354	0.26	10	0.211
		autumn 2012 x spring 2013	0.30886	0.887	10	0.79

Pair-wise test		Comparisons	t	P(perm)	perms	P (MC)
Within WD	1500	spring 2012 x autumn2012	4.6079	0.094	10	0.078
		spring 2012 x spring 2013	0.46649	0.724	10	0.694
		spring 2012 x autumn2013	2.3347	0.212	10	0.104
		autumn 2012 x spring2013	1.2311	0.338	10	0.299
		autumn2012 x autumn´13	0.11423	1	10	0.913
		spring2013 x autumn 2013	1.04	0.517	10	0.358
	1750	spring 2012 x autumn2012	5.2868	0.097	10	**0.001**
		spring 2012 x spring 2013	1.0636	0.395	10	0.367
		spring 2012 x autumn2013	0.41709	0.689	10	0.708
		autumn2012 x spring 2013	0.13992	0.895	10	0.899
		autumn2012 x autumn2013	2.2525	0.105	10	**0.019**
		spring 2013 x autumn2013	1.401	0.197	10	0.244
	2000	spring 2012 x autumn2012	0.7868	0.701	10	0.498
		spring 2012 x autumn2013	3.6486	0.333	3	**0.013**
		autumn 2012 x autumn2013	0.38521	0.884	10	0.748
Within Tm	Spring 2012	500, 900	2.804	0.326	3	0.113
		500, 1200	10.862	0.675	3	0.394
		500, 1500	22.942	0.349	3	0.159
		500, 1750	9.164	0.338	3	**0.013**
		500, 2000	11.324	0.361	3	**0.004**
		900, 1200	1.596	0.334	3	0.238
		900, 1500	23.997	0.325	3	0.137
		900, 1750	12.632	0.346	3	0.376
		900, 2000	0.78171	1	3	0.537

		1200, 1500	0.53989	1	3	0.631
		1200, 1750	0.96744	0.646	3	0.447
		1200, 2000	15.971	0.34	3	0.242
		1500, 1750	94.176	0.349	3	**0.015**
		1500, 2000	12.042	0.38	3	*0.008*
		1750, 2000	42.078	0.319	3	**0.043**

Table S3.2. Results from pair-wise multivariate PERMANOVA analyses for differences in meiofauna composition in Blanes Canyon. PERMANOVA pair-wise comparisons between Time and water depth (WD). Spr: spring; Aut: autumn. Data was fourth-root transform and resemblance was calculated using Bray Curtis prior to analysis. Bold values indicate significant differences at $p < 0.05$ and bold italic indicate significant at $p < 0.01$.

Pair-wise test	**Comparisons**	**t**	**P(perm)**	**perms**
Between Time	Spr.2012 x Aut. 2012	1.916	**0.0112**	9947
	Spr.2012 x Spr. 2013	1.1289	0.2843	9951
	Spr.2012 x Aut. 2013	1.5322	0.076	9959
	Aut. 2012 x Spr. 2013	0.90768	0.5252	9967
	Aut. 2012 x Aut. 2013	2.0038	***0.0082***	9950
	Spr. 2013 x Aut. 2013	1.1812	0.2435	9961
Between WD	500, 900	17.815	**0.0146**	9951
	500, 1200	13.989	0.1169	9951
	500, 1500	2.379	***0.0011***	9954
	500, 1750	20.568	***0.009***	9942
	500, 2000	40.687	***0.0012***	9955
	900, 1200	14.142	0.1109	9953
	900, 1500	24.346	***0.0011***	9951
	900, 1750	16.707	**0.0262**	9942
	900, 2000	20.524	***0.0069***	9952
	1200, 1500	21.174	***0.0067***	9947
	1200, 1750	18.127	**0.0264**	9953
	1200, 2000	25.863	***0.0095***	9954
	1500, 1750	12.921	0.1453	9947
	1500, 2000	19.719	***0.0075***	9946
	1750, 2000	15.664	0.0659	9956

Table S3.3. Distance-based linear model (DISTLM) for meiofauna assemblages and selected environmental variables for the western open slope. Variables: Selected environmental variables used to calculate the optimum model. Marginal tests: explanation of variation for each variable taken separately. Sequential tests: conditional tests of individual variables in constructing the model. Each test examines whether adding the variable contributes significantly to the explained variation. Selection procedure: step-wise, selection criterion: adjusted R^2. Prop.: % variation explained. Cumul.: cumulative variation explained. Chl *a*: chlorophyll *a*, CPE: chloroplastic pigment equivalents, Chl *a*:phaeo: chlorophyll a divided by its degradation products indicating ‘freshness’ of the phytodetrital OM, TN: total nitrogen, OC: organic carbon, C:N: molar carbon-nitrogen ratio.

MARGINAL TESTS				
Variable	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**
Clay (%)	12688	22.71	***0.0001***	0.26191
Silt(%)	2911.4	4.0922	**0.016**	6.01E-02
Sand(%)	1098.4	1.4848	0.2015	2.27E-02
TN(%)	629.51	0.8426	0.4321	1.30E-02
% OC	11701	20.382	***0.0001***	0.24154
C:N	7306.6	11.367	***0.0002***	0.15083
Log(Chl *a*+0.1)	5098.3	7.5278	***0.0008***	0.10524
Log(CPE+0.1)	11983	21.033	***0.0001***	0.24735
Log(Chl *a*: Phaeo+0.1)	273	0.3627	0.8063	5.64E-03

SEQUENTIAL TESTS							
Variable	**Adj R^2**	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**	**Cumul.**	**res.df**
+Clay (%)	1.67E-02	2813.5	3.7777	0.0149	2.27E-02	2.27E-02	163
+Silt(%)	2.52E-02	1794.8	2.431	0.0623	1.44E-02	3.71E-02	162
+Sand(%)	2.58E-02	809.8	1.0975	0.3277	6.52E-03	4.36E-02	161
+TN(%)	4.05E-02	2513.4	3.4584	0.0211	2.02E-02	6.39E-02	160
+OC(%)	4.95E-02	1817.2	2.5243	0.0487	1.46E-02	7.85E-02	159
OC(%)	4.90E-02	656.4	0.91129	0.421	5.28E-03	8.38E-02	158
+log(Chl *a*+0.1)	0.13724	11216	17.164	0.0001	9.03E-02	0.17406	157
+log(CPE+0.1)	0.14186	1199.6	1.8456	0.1206	9.66E-03	0.18372	156
+log(Chl *a*: phaeo+0.1)	0.1401	443.02	0.68023	0.592	3.57E-03	0.18729	155

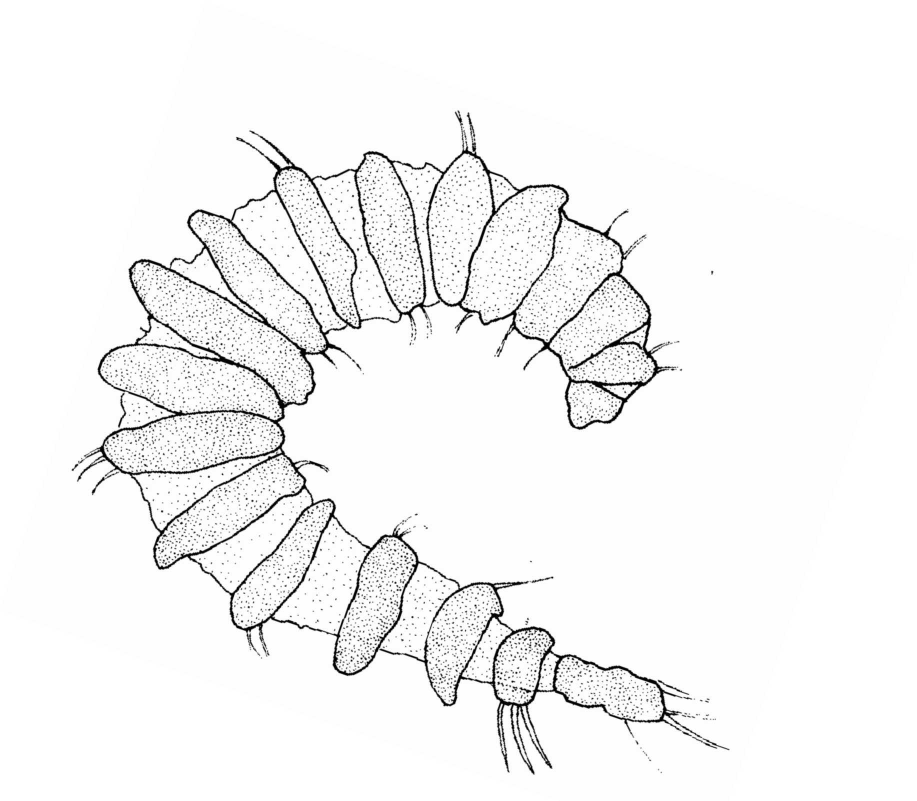
Table S3.4. Distance-based linear model (DISTLM) for meiofauna assemblages and selected environmental variables for the Blanes Canyon. Variables: Selected environmental variables used to calculate the optimum model. Marginal tests: explanation of variation for each variable taken separately. Sequential tests: conditional tests of individual variables in constructing the model. Each test examines whether adding the variable contributes significantly to the explained variation. Selection procedure: step-wise, selection criterion: adjusted R^2. Prop.: % variation explained. Cumul.: cumulative variation explained. Chl *a*: chlorophyll *a*, CPE: chloroplastic pigment equivalents, Chl *a*:phaeo: chlorophyll a divided by its degradation products indicating 'freshness' of the phytodetrital OM, TN: total nitrogen content, OC: organic carbon content, C:N: molar carbon-nitrogen ratio.

MARGINAL TESTS				
Variable	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**
Clay (%)	2813.5	3.7777	**0.0149**	2.27E-02
Silt(%)	1659.4	2.2071	0.0801	1.34E-02
Sand(%)	1884.5	2.5111	**0.0563**	1.52E-02
TN(%)	2594.2	3.4769	**0.0212**	2.09E-02
OC	1153.1	1.5274	0.1692	9.28E-03
C:N	1052.4	1.3929	0.214	8.47E-03
log(Chl-a+0.1)	11265	16.257	***0.0001***	9.07E-02
log(CPE+0.1)	4706.8	6.4198	***0.001***	3.79E-02
log(Chl a: Phaeo+0.1)	3496.9	4.7218	***0.0046***	2.82E-02

SEQUENTIAL TESTS							
Variable	**Adj R^2**	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**	**Cumul.**	**res.df**
+Clay (%)	1.67E-02	2813.5	3.7777	0.0149	2.27E-02	2.27E-02	163
+Silt(%)	2.52E-02	1794.8	2.431	0.0623	1.44E-02	3.71E-02	162
+Sand(%)	2.58E-02	809.8	1.0975	0.3277	6.52E-03	4.36E-02	161
+TN(%)	4.05E-02	2513.4	3.4584	0.0211	2.02E-02	6.39E-02	160
+OC(%)	4.95E-02	1817.2	2.5243	0.0487	1.46E-02	7.85E-02	159
OC(%)	4.90E-02	656.4	0.91129	0.421	5.28E-03	8.38E-02	158
+log(Chl-a+0.1)	0.13724	11216	17.164	0.0001	9.03E-02	0.17406	157
+log(CPE+0.1)	0.14186	1199.6	1.8456	0.1206	9.66E-03	0.18372	156
+log(Chl a: phaeo+0.1)	0.1401	443.02	0.68023	0.592	3.57E-03	0.18729	155

4

Nematode Community zonation in response to Environmental drivers along the Canyon axis



4.1 INTRODUCTION

Deep-sea ecosystems cover nearly two-thirds of the Earth's surface and represent the largest biome in the biosphere. The perception that the deep sea is relatively homogeneous is outdated, and it is now acknowledged that habitat heterogeneity and species diversity is amongst the highest on the planet.

Despite this, its spatial and temporal patterns as well as its biological and ecological diversity is amongst the least explored, whilst our knowledge on the functions and services it provides to humankind remains very poor (Rex, 1981; Danovaro et al., 2010; Vanreusel et al., 2010; Ramirez-Llodra et al., 2010b; Thurber et al., 2014). Among the various deep-sea habitats, submarine canyons have gained much increased interest from marine ecologists. Their topographical uniqueness, alongside tremendous environmental heterogeneity and hydrodynamic variability, and their prevalence along productive continental margins make them ideal subjects to study ecological patterns and faunal drivers in the deep sea.

In deep-sea sediments worldwide, nematodes are generally the most abundant and diverse metazoan component (Jensen, 1988; Tietjen, 1992; Heip et al; 1985) and this is certainly true for canyons as well (Ingels and Vanreusel, 2013; Ingels et al., 2011a; Leduc et al., 2014). Deep-sea habitat heterogeneity greatly influences the nematode diversity and assemblage structure and functional characteristics (Vanreusel et al., 2010). This includes the small-scale vertical distribution patterns along the sediment profile that are still poorly understood (Snelgrove and Smith, 2002; Gorska et al., 2014). At larger scales, the highest levels of heterogeneity in deep-sea habitats are known to occur around major geomorphological structures such as seamounts and submarine canyons, which lead to modifications in bathymetric patterns generally observed along the shelf-slope-abyss gradient. Nematode densities and biomass, along with those of meiofauna in general, typically decrease with water depth in response to food availability (Muthumbi et al., 2004; Soetaert et al., 1995; Vanaverbeke et al., 1997), but canyon meiofauna studies have shown that such a bathymetric decrease is not universal, and depends on the local topographical and related environmental conditions. Therefore, insights into nematode assemblage structure and functional characteristics have the potential to provide relevant information for understanding the environmental forces driving many aspects of the ecology of deep-sea benthos (e.g., Ingels et al., 2009; Hasemann and Soltwedel, 2011; Pusceddu et al., 2013).

Submarine canyons can modify the local oceanographic regime, by altering current directions and speed and/or by increasing the turbidity flows for instance. They also enhanced the exchange of water and sediments between the shelf and slope, acting as preferential conduits of organic matter towards the abyss and serving as deep-sea deposition areas for large quantities of sediment and particular matter (Puig et al., 2000; Pasqual et al., 2010; Heussner et al 2006; Lopez-Fernandez et al., 2013; among others). Benthic communities, including the meiobenthos, are known to reflect these unstable and organically enriched conditions in canyon systems (Aller, 1997; Garcia et al., 2007; Ingels et al; 2009; Van Gaever et al., 2009; Romano et al., 2013b; Román et al., 2016). Indeed, enhanced particle transport along canyons makes for an effective food supply to benthic organisms, resulting in higher faunal densities and biomass comparing to non-incised slopes (Rowe et al., 1982; Soltwedel et al., 2000; De Leo et al., 2010; Leduc et al., 2014; Gambi and Danovaro, 2016). High habitat heterogeneity, coupled with an enhanced food supply, results in submarine canyons being biomass hotspots that consequently boost local fisheries (e.g., Rowe et al., 1982; Vetter, 2010; Company et al., 2008; De Leo et al., 2010). As a result, canyons and adjacent areas are targeted for fish and shellfish and may subsequently suffer anthropogenic impacts. Some of them are caused by local fisheries, mainly by bottom trawling (Company et al., 2012; Pusceddu et al., 2014; Román et al., 2016), while some others are related with channelling and accumulation of pollutants such as litter (Ramirez-Llodra et al., 2013) or chemicals (Sanchez-Vidal et al., 2015). These additional disturbances certainly play an important role in the canyon system dynamics. Mediterranean margins are frequently and deeply incised by submarine canyons, and have received increased attention in recent years. For some of them, nematode diversity patterns in these ecosystems have been studied along bathymetric gradients at different spatial scales (Bianchelli et al., 2013). Identifying the importance of the factors that drive turnover diversity and would allow to predict (inter)spatial patterns and species composition in deep-sea assemblages has been highlighted as a priority to understand biodiversity dynamics in the deep sea and in submarine canyons. However, the available information on nematode community structure related to its environmental drivers in canyons is still limited (Danovaro et al., 2009; Ingels et al., 2013; Vanreusel et al., 2010). This knowledge, however, is essential to increase our understanding on the functioning of these highly heterogeneous habitats.

In this study, we provide a comprehensive analysis of the spatial patterns of nematode structural and functional diversity in Blanes Canyon, while considering their relationships with the main biogeochemical and environmental factors. Differences in assemblage composition, structure and diversity are assessed at six stations along a bathymetric gradient (500 to 2000 m depth), as well as along the vertical sediment profile (0-5 cm), to identify the main driving factors of canyon infaunal communities. The lack of clear bathymetric gradients in meiofauna canyon assemblages (Romano et al., 2013b; **Chapter 3**) is assessed further by investigating the following questions for nematodes: **1**) Are elevated densities at specific water depths in the canyon explained by the increase in particular genera or trophic groups? **2**) Are these elevated densities associated with the surface and/or subsurface sediments? **3**) Are the same environmental factors that explain density changes also associated with changes in nematode genera composition? By answering these questions we also aim at identifying spatial nematode community patterns that are related to the canyon environment.

4.2 MATERIAL AND METHODS

4.2.1 Sampling

Sediment samples were collected in October 2012 during the DM-II cruise at six stations along Blanes Canyon, from 473 m to 1964 m depth (see **Chapter 2** section **2.2**, for details on sampling strategy, Table 2.1, Fig.4.1). Between 2 and 3 multicorer deployments (replicates) were conducted at each sampling station. From each multicore deployment, one core was used for meiofaunal and three for sediment analyses, which were all carefully sub-sampled on board by means of a small PVC tube (36 mm of diameter, 5 cm sediment depth) taken from the center of the core to maintain a consistent sample surface area for all replicates. The three sediment layers from each subcore (see **Chapter 2**, section **2.2.1** for further details) were used separately in the analyses.

4.2.2 Sediment analyses

The methods to determine the quantity and quality of sediments variables are fully described in **Chapter 2** (**section 2.3.1**). Grain size (clay, silt and sand fractions), Chlorophyll a (Chl *a*, μg/g) and chlorophyll degradation products in the sediment, organic carbon concentration (OC%) and total nitrogen concentration (TN%) sediment variables were considered in the present studied. Moreover, Chloroplastic Pigments Equivalents (CPE: sum of Chl *a* and its degradation products as phaeopigments) were used to estimate surface-produced organic matter (OM). The ratio Chl *a*: phaeopigments (Chl *a*: phaeo) is used as a proxy to estimate the freshness of photosynthetically derived OM (Thiel, 1978).

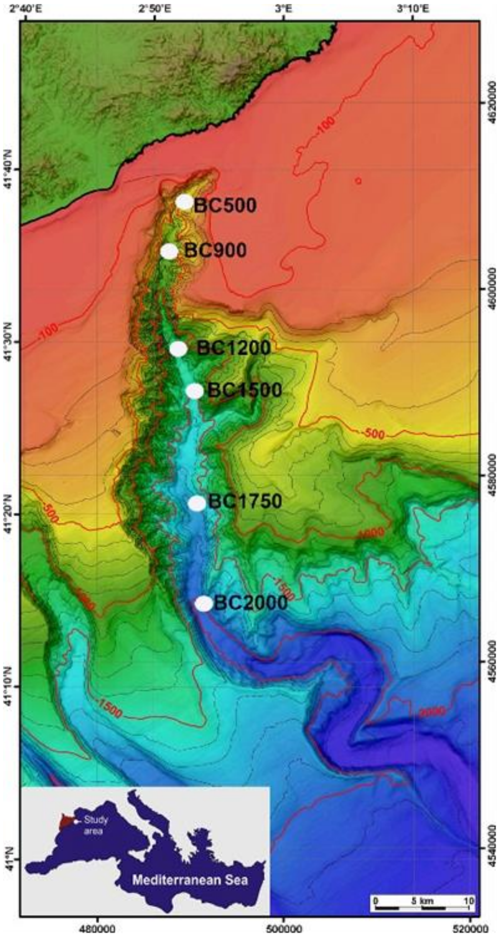


Figure 4.1. Overview (down-left) and detailed map (coluor) of the studied area. BC: Blanes Canyon. Note: only the detailed map has a scale

4.2.3 Meiofauna and nematode analyses

The procedure for meiofauna and nematode analyses are detailed in **Chapter 2**, sections **2.3.2** and **2.3.3**, respectively. Between 100-150 nematodes (all if density < 100) were picked out randomly from each layer sample and identified under compound microscope (100 x magnification) to genus level using pictorial keys (Platt and Warwick, 1988) and the taxonomic literature of the Nematode Library at Ghent University. Specimens that could not be identified to the genus level were assigned to the appropriate higher taxon level and the NeMys database (Guilini et al., 2016). Specimens that could not be ascribed to a genus level were grouped within the appropriate family to account for its presence in the sample.

The trophic composition of the nematode assemblage was assessed using the feeding-type classification of Wieser (1953): selective deposit feeders (1A), non-selective deposit feeders (1B), epistratum feeders (2A) and predators/scavengers or omnivores (2B). An additional feeding type (3) was incorporated for the mouthless genus *Astomonema* (Ingels et al., 2011a).

4.2.4 Data analyses

Nematode community descriptors analysed in this study were: density, biomass, structural and functional diversity, and structure) by means of non-parametric permutational analyses of variance (PERMANOVA) using PRIMER v6 (Anderson et al., 2005, 2008). Structural diversity were measured as genus richness, Shannon index (Shannon, 1948), the evenness index (J', Pielou, 1969) and expected number of genera (*EG*(51), using the function DIVERSE in PRIMER v6 (Clarke and Gorley, 2006). Functional diversity was calculated as index of trophic diversity as $\mathrm{TD}= 1/[\,q_1{}^2 + q_2{}^2 + q_3{}^2 \ldots + q_n{}^2]$, where q is the proportion of each feeding type and n is the number of feeding types (Heip et al, 1988).

Data on nematode densities (genus level) were standardized prior to calculating the resemblance matrix using Bray-Curtis similarity. A multivariate analysis of variance by permutation (PERMANOVA, Anderson et al., 2005; 2008) was used to assess the differences between both water depth (1-factor design; WD: fixed factor) and sediment layers (3-factor nested design; WD: fixed; sediment layers, SL: fixed; replicate, Rep: random and nested in WD). Subsequent pair-wise *t*-tests were performed to assess the differences between each pairwise combination of factor levels. When PERMANOVA permutations numbers were limited < 100; Monte Carlo values were used to infer significance. To assess the magnitude of the spatial variation at each spatial scale the Estimated Components of Variation (ECV) were used. Hierarchical Cluster analysis based on Bray-Curtis similarity matrix using the group average aggregation method was performed to assess the structure of the nematode assemblages which was superimposed on non-metrical Multi-dimensional scaling plots (MDS) to visualize the results.

The same PERMANOVA design used for multivariate community data was applied to univariate data: density and biomass, as well as to trophic and diversity indices.

A one-way Similarity Percentages analysis (SIMPER) was performed using WD as factor to reveal which genera are responsible for the multivariate assemblage patterns within stations.

Sediment variables were tested for collinearity (Draftsman plot and Spearman rank correlation matrix) and ($R^2 > 0.95$ (redundant) were omitted from the analyses (i.e., phaeo and Chl *a*: OC). Then the data were checked for uniform distribution and TN and C:N ratio were log (X + 0.1) transformed to compensate for skewness. Principal component analyses (PCA) were performed on normalized data and Euclidean resemblance matrix to assess the overall differences among stations, which were tested by separate univariate PERMANOVA tests using the same design as for community analyses.

Distance-based linear model analysis (DISTLM, Anderson et al., 2008) were performed to model the relationships between assemblages and environmental variables (clay, silt, sand, Chl *a*, Chl *a*: phaeo, CPE, OC, TN and C: N). The DISTLM was built using a step-wise selection procedure and the adjusted R^2 as selection criterion based on Euclidean distances. The results were visualized using a distance-based redundancy analysis (dbRDA) plot. To complement these analyses, non-parametric Kendal-Tau correlation were computed between selected biotic variables (density, biomass, TD, *H´*, and *EG*(51)) and abiotic variables (clay, silt, sand, OC, TN, Chl *a*, CPE, Chl *a*: phaeo and C:N). Additional Kendall-Tau correlations were performed between the abiotic variables and the most abundant nematode genera.

4.3 RESULTS

4.3.1 Sediments

The PCA plot shows a distinct separation between stations along the canyon axis (Fig. 4.2) based on sedimentary variables, forming three clear groups: upper stations (BC500, BC900 and BC1200, and showing the highest variability), middle stations (BC1500 and BC1750) and the deeper canyon (BC2000). The first two PC axes explain 67.6 % of the variation (42.2 % and 25.4 % for PC1 and PC2, respectively). The main contributors are CPE (+0.470), Chl *a* (+0.464) and sand (+0.438) for the axis PC1 and C:N (-0.597), OC (+0.525) and silt (+0.389) for the axis PC2 (numbers_in parenthesis represent eigenvectors)

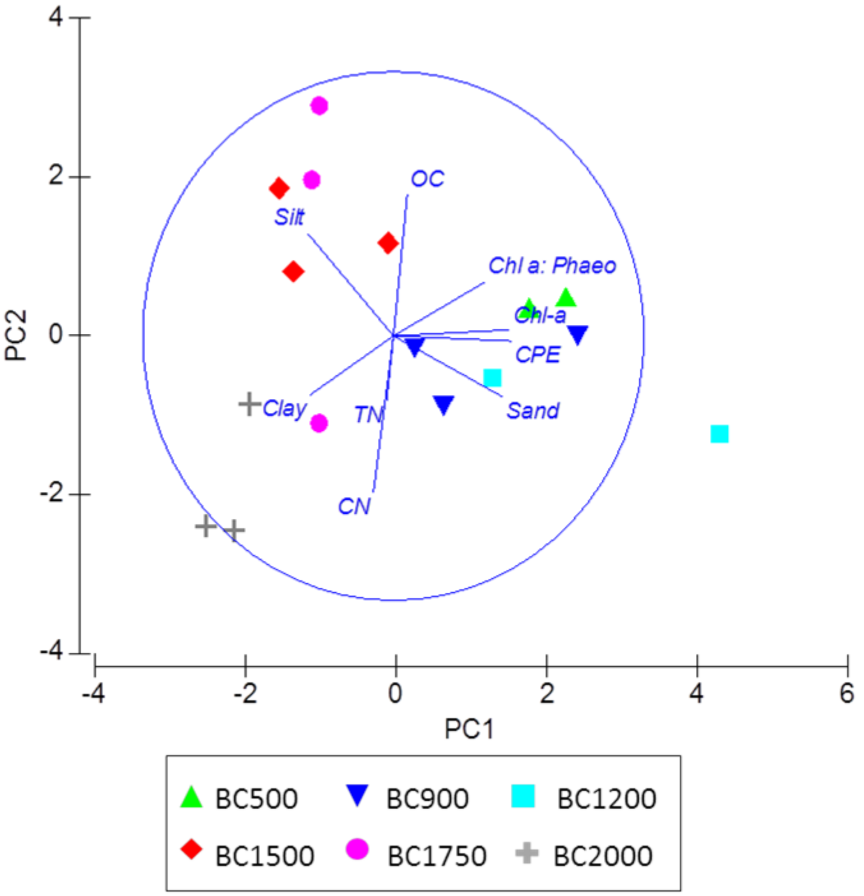


Figure 4.2. Principal Component Analysis (PCA) ordination (based on Euclidean Distance values) for environmental selected variables.

All sediment descriptors (grain size, OC, Chl *a*, CPE and Chl *a*: phaeo), except TN and C: N, showed significant differences between stations (WD), while only the grain size fractions differed between sediment layers (SL) (PERMANOVA, $p<0.01$, Table S2).

Along the canyon axis sediments were predominantly muddy with high silt content (69-77%) (Table S1). The highest sand content (12.5% ± 4) was observed at BC1200 (Table S1, Fig. 4.3). Overall, sediment became finer when deeper into sediment profile, especially at BC2000 (Table S1). Sand content was higher along the whole sediment profile at BC500, BC900 and BC1200 compared to the deeper stations (Fig.4.4). The sediment grain size composition also differed significantly for the interaction term WD x SL (Table S2). Pairwise comparisons showed that significant differences between WD mainly occurred for deeper sediment layers (2-5 cm) ($p < 0.05$, data not shown, see Table S1).

Organic carbon concentration was higher on average at BC500, followed by BC2000. TN showed the highest content at BC500 and BC1750 (Table S1). A decrease of OC along the sediment profile was only observed at BC900 and BC2000 and for TN at BC1750 and BC2000 (Table S1). The highest C: N values were observed at BC2000.

Chl *a* and CPE decreased with the increasing water depth (Table S1), except for particularly high peaks at BC1200 (0.1 ± 0.007 and 4.7 ± 0.1 respectively) (Table S1, CPE: Fig. 4.3). Pair wise comparisons for Chl *a* and CPE indicated differences were significant and greater in the upper canyon (BC500, BC900 and BC1200) compared to the deeper canyon (BC1500, BC1750 and BC2000) (pair-wise comparisons, $p < 0.05$, data not shown but see Table S1). A decreased of Chl *a* and CPE with increasing sediment depth only occurred at BC900, BC1750 and BC2000 (Table S1, CPE: Fig. 4.4).

The "freshness" (Chl *a*: phaeo) of the sedimentary OM did not show specific trends, not along the bathymetric decline, nor along the sediment profile (Table. S1).

4.3.2 Nematode assemblages

4.3.2.1 Density and biomass

Nematodes dominated the meiofaunal assemblages at all stations (87-95%), with densities ranging between 718 and 1614 ind. x 10 cm^{-2} (Fig. 4.3). Densities differed significantly between WD and SL (PERMANOVA, $p < 0.01$, Table 2). However, there was no clear decrease with increasing water depth, since the densities were significantly higher at BC500, BC1200 and BC1750 compared to BC900, BC1500 and BC2000 (pairwise comparisons, Table S3).

Nematode densities were mainly concentrated in the surface layers (0-1 cm) and decreased with increasing sediment depth (Fig. 4.4). There was a significant WD x SL interaction (Table 4.1), with the pairwise comparison indicating significant differences between sediment layers (SL) at all stations, except at BC1500 and BC2000 (Table S3, Fig. 4.4). In the surface layer, the density was significantly higher at BC1750 compared to all other stations, while densities in the sub-surface and deeper layers were highest at BC500, followed by BC1200 and BC2000 (pairwise comparisons, Table S3, Fig. 4.4).

Nematode biomass was significantly higher at BC1200, followed by BC500 (104.3 ± 20 and 98.9 ± 4 µg C 10 cm^{-2}, respectively, Fig. 4.3) and significantly lower at BC900 (29 ± 2.1 µg C 10 cm^{-2} (PERMANOVA, $p < 0.01$, Table 4.1, Table S4). Overall, larger nematodes such as *Sabatieria* dominated the deepest layers.

There were no clear overall trends in total biomass distribution along the sediment profile (Fig. 4.5, Table 4.1). However, at BC900, BC1500 and BC1750, biomass was mostly concentrated in the surface layer, while at BC500 and BC1200 biomass was highest in the deepest sediment layer.

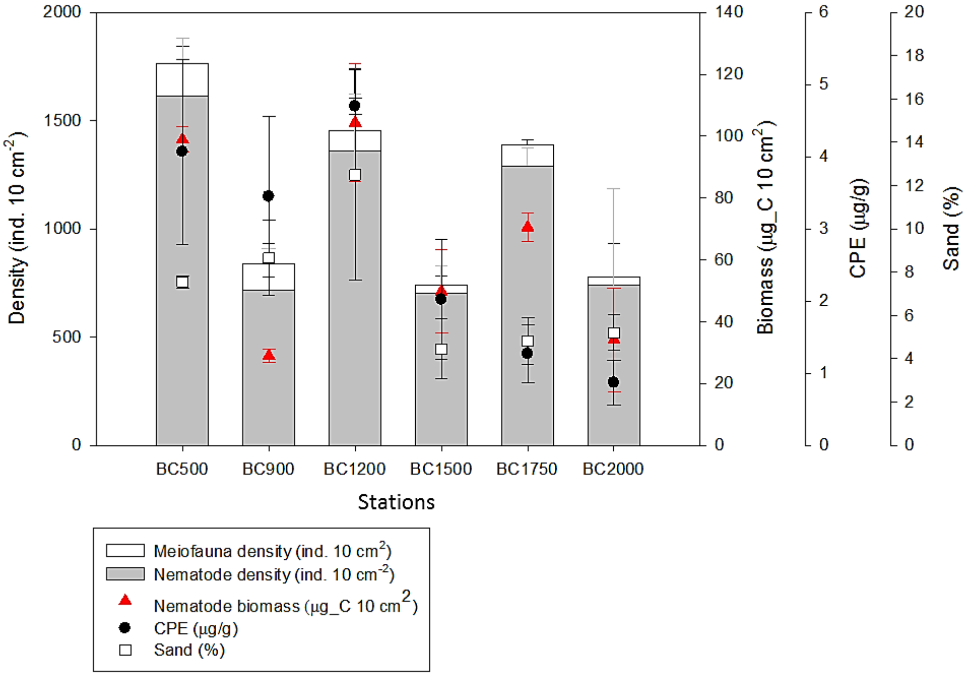


Figure 4.3. Meiofauna and nematode density (ind·10 cm^2) in the studied stations. Superimposed are: nematode biomass (µg C· 10 cm^{-2}), sand content (%) and CPE (µg/g). Scales were made uniform for better comparison. Values average over replicates, error bars denote standard deviations.

Table 4.1. Results of PERMANOVA analyses for differences in univariate (density and total biomass) and multivariate (composition) nematode characteristics using water depth (WD), sediment layer (SL) and Replicates (Rep) as factors. WD x SL: double interaction factor. *: $p < 0.05$; **: $p < 0.01$; n.s.: non-significant; df; degrees of freedom; SS: sum of squares; MS: mean squares; Pseudo-F: pseudo-*F* ratio; P(perm): permutation *P*-value; Perms: permutations; ECV: Estimated coefficient of variation.

Source	df	SS	MS	Pseudo-F	P(perm)	Perms	ECV
Density							
WD	5	438.79	87.759	86.453	**	9955	9.762
SL	2	1422.6	711.29	37.54	**	9953	44.874
Rep (WD)	10	101.51	10.151	0.53574	n.s	9951	-2.9322
WD x SL	10	529.24	52.924	2.79	*	9941	12.821
Residual	20	378.95	18.948				18.948
Total	47	3012.1					
Biomass							
WD	5	58.89	11.778	70.879	**	9961	1.3221
SL	2	0.1180	0.0599	0.0626	n.s	9941	-0.05923
Rep (WD)	10	16.62	1.662	17.897	n.s	9939	0.33029
WD x SL	10	17.897	17.897	18.989	n.s	9940	0.94247
Residual	20	17.907	0.9424				
Total	47	105.59					
Community							
WD	5	21046	4209.1	3.338	**	9849	367.24
SL	2	25889	12944	12.55	**	9924	830.88
Rep (WD)	10	12610	1261	1.225	n.s	9776	56.198
WD x SL	10	13441	1344.1	13.032	*	9809	208.62
Residual	20	20629	1031.4				1211.1
Total	47	94840					

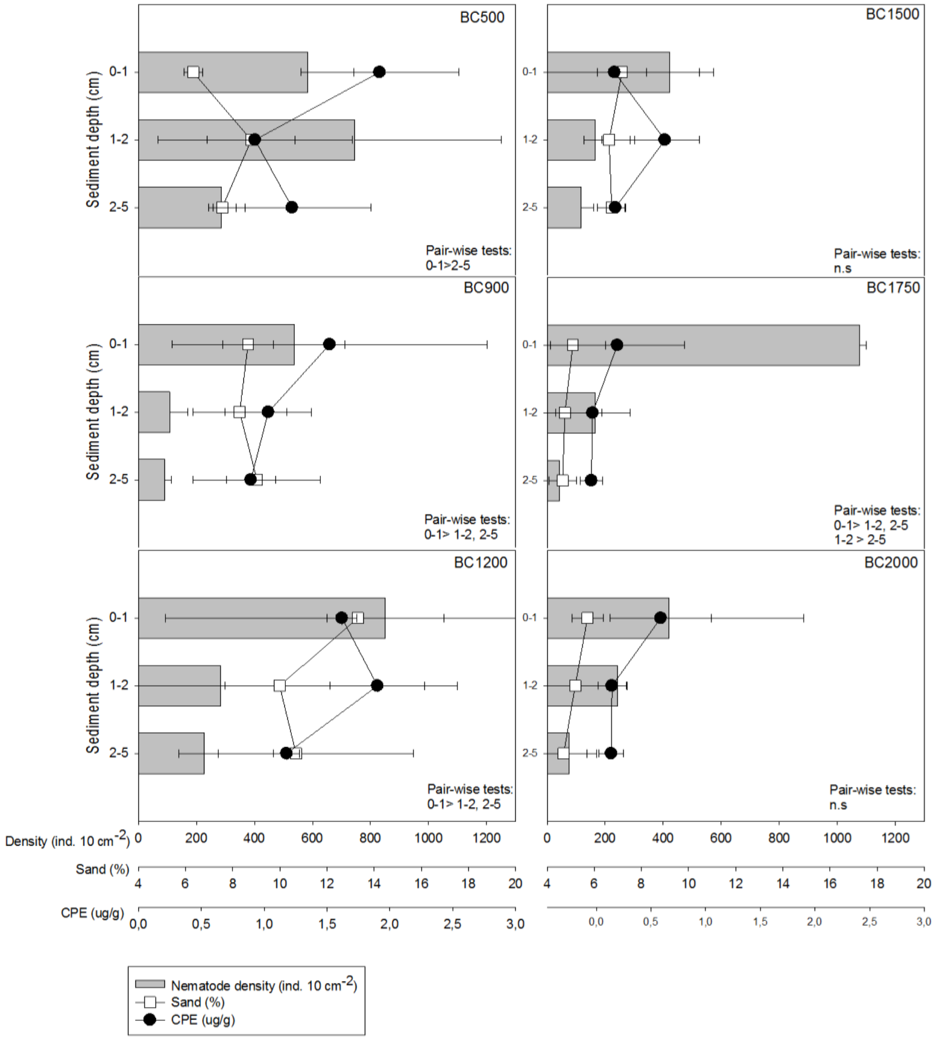


Figure 4.4. Nematode density (ind·10 cm^{2}) along the sediment profile in the studied stations. Superimposed: nematode biomass (µgC 10 cm^{-2}), sand content (%) and CPE (µg/g). Scales were made uniform for better comparison. Numbers represent the sediment layers compared in PERMANOVA pair-wise comparisons (WD x SL) in order to assess differences in nematode density among layers. Only significantly different (p<0.05) pairs are shown. Values average over replicates, error bars denote standard deviations.

4.3.2.2 Structural and functional nematode diversity

In total 3979 nematode specimens belonging to 27 families and 109 genera were identified (*S*, Table 4.2, and **Appendix 3**). Nineteen families were present in all stations. The family Ironidae was exclusively found at BC2000 whereas Oncholaimidae and Axonolaimidae were absent from the two deepest stations (BC1750 and BC2000). More genera were recovered from BC500 and BC2000 samples (67 and 68, respectively, Table 4.2) and only 29 occurred at all stations. The upper (BC500, BC900 and BC1200) and the middle canyon (BC1500 and

Table 4.2. Diversity index based on genera abundance per sediment depth layer (0-1, 1-2, 2-5) and total (0-5) per stations. *S*: richness of genera; *H´*: Shannon index; *J´*: evenness number; *EG*(51): expected number of species; TD: trophic diversity index.

Station	Sediment layer (cm)	*S*	*H´*	*J´*	*EG*(51)	TD
BC500	0-1	45	3.00	0.86	20.02	3.10
	1-2	44	3.08	0.89	20.86	3.46
	2-5	28	2.09	0.70	12.92	1.58
	0-5	**67**	**3.32**	**0.84**	**23.2**	**3.04**
BC900	0-1	50	2.91	0.84	19.46	3.36
	1-2	35	2.64	0.92	16.68	3.05
	2-5	30	2.08	0.79	12.83	2.25
	0-5	**61**	**3.16**	**0.85**	**21.8**	**3.31**
BC1200	0-1	45	3.00	0.86	20.49	3.42
	1-2	32	2.76	0.85	17.44	3.42
	2-5	30	1.77	0.60	11.21	3.37
	0-5	**61**	**3.22**	**0.82**	**22.3**	**3.30**
BC1500	0-1	47	3.08	0.92	20.61	3.83
	1-2	38	2.70	0.88	17.13	2.68
	2-5	22	1.87	0.70	11.63	2.40
	0-5	**65**	**3.24**	**0.87**	**21.8**	**3.66**
BC1750	0-1	53	3.06	0.88	20.41	2.78
	1-2	29	2.51	0.80	16.30	3.09
	2-5	19	1.51	0.72	8.56	2.98
	0-5	**64**	**3.19**	**0.86**	**21.7**	**3.33**
BC2000	0-1	46	2.81	0.88	18.20	3.54
	1-2	50	2.64	0.79	17.81	2.56
	2-5	35	1.74	0.72	8.39	1.68
	0-5	**68**	**3.08**	**0.81**	**21.4**	**3.42**

BC1750) stations shared 43 and 44 genera, respectively. Upper stations shared 31 and 35 genera with middle and deep (BC2000) stations, respectively, while middle and deep stations shared 39. In general, genera were more numerous in surface layers than in deeper sediment layers (Table 4.2). Overall, the percentage of rare genera (genera with a relative abundance of < 1% per station) averaged 13.3 %.

Average *H´* and *EG*(51) was higher at BC500 (Table 4.2). However, significant differences were only found for SL (PERMANOVA, p<0.05, Table S5), where by *H´* and *EG*(51) decreased along the sediment profile (Table 4.2). Pielou evenness did not show any clear patterns.

Average TD was higher at BC1500, although significant differences only occurred for SL (PERMANOVA, p< 0.05, Table S5). TD decreased in deeper layers (Table 4.2).

4.3.2.3 Nematode community

Overall, the most abundant families were Comesomatidae (23%), Chromadoridae (14%), Oxystominidae (12%) and Xyalidae (10%). The most abundant genera per station and per sediment profile layer are shown in Table 4.3. *Sabatieria* (7.01-20.16%) and *Halalaimus* (6.4 -13.8%) were among the most abundant genera at all stations (Table 4.3). *Cervonema* showed relatively high abundances at BC500, BC900 and BC1200 (5.2 – 8%) and *Setosabatieria* was particularly dominant at BC500 and BC1200, while *Acantholaimus* was particularly dominant at BC1750 and BC2000 and *Molgolaimus* at BC1750 (Table 4.3).

In the first two cm of sediment, *Halalaimus* was dominant high densities (>8%) at all stations. *Cervonema* was also abundant (>4%) except at BC2000, where it was absent (Table 4.3). In the deepest sediment layer (2-5 cm) *Sabatieria* (>8%) was recovered at all stations, while *Setosabatieria* was particularly abundant at BC500, BC900 and BC1200 (Table 4.3).

The high densities registered at BC500, BC1200 and BC1750 stations (cf. section 3.2.2) were caused by different genera in the subsurface (BC500) and surface layers (BC1200, BC1750) (Fig. 4.3, Table 4.3). These were: *Cervonema*, *Halalaimus* and *Sphaerolaimus* at BC500, *Halalaimus* at BC1200, and *Acantholaimus*, *Halalaimus* and *Molgolaimus* at BC1750 (Table4.3).

Table 4.3. Relative abundance of the dominant genera (≥2%) in the studied stations along the sediment profile. (−: 0%; *: >0%, <2%; **: ≥2%, <4%; ***: ≥4%, <8%; ****: ≥8%, < 16%; *****:≥16%).

	BC500				**BC900**				**BC1200**			
Sediment layer (cm)	0-1	1-2	2-5	**0-5**	0-1	1-2	2-5	**0-5**	0-1	1-2	2-5	**0-5**
Acantholaimus	**	*		*	***	*	**	***	*			*
Actinonema	**	*	*	**	***	**	*	***	***	*		***
Amphimonhystrella	*		*	*	*	**	*	*	*	**	*	*
Campylaimus	**	**		*	*	*	**	*	***	**	*	**
Cervonema	****	***	***	****	****	***	*	***	***	****	*	***
Chromadorella	**	**		***	*	*		*	*			*
Chromadorita	*			*	*	*		*	**			**
Daptonema	***	**	*	**	***	***	*	***	***	*		**
Desmoscolex	*	*		*	*	**		*				
Dichromadora	*	***		**	****	***	*	***	***			***
Diplopeltula	**	*	*	*	*	*	*	*	**	***		**
Disconema	*	**	*	**	**	*		*	*			*
Dorylaimopsis			*	*		**	**	*		***	***	**
Elzalia	**	**		**	*	*		*	**			*
Halalaimus	***	***	**	***	****	****	*	****	*****	***	*	****
Halichoanolaimus						*		*	*	*		*
Leptolaimus	*	**	**	**	*	*	*	*	*	**	*	*
Marylynnia	**	*	*	*	*	**	*	*		*	*	*
Metasphaerolaimus	*	*	***	**		*	**	*	*	*	*	*
Molgolaimus	*	*		*	*		*	*	*			*
Monhystrella	*	**	**	*	*		**	*		*	*	*
Oxystomina	*	*		*	*			*	*	*	*	*
Paramesacanthion		*		*	*			*	*			*
Pomponema	**	**		**	**			*	***		*	**
Pselionema	*	*		*	*			*	*		*	*
Sabatieria	***	**	*****	****	*	****	*****	***	*	****	*****	****
Setosabatieria	***	*	*****	***	*	**	***	*	*	*****	*****	***
Sphaerolaimus	***	***	***	***	*	***	***	**	*	***	***	**
Syringolaimus												
Tricoma	**	**		*	***	*		**	**			*
Xylalidae				*	**	***	**	**	**	**		**

Table 4.3. continued

BC1500				**BC1750**				**BC2000**			
0-1	1-2	2-5	**0-5**	0-1	1-2	2-5	**0-5**	0-1	1-2	2-5	**0-5**
***	*		**	****	*		***	****	**	*	***
***	*		**	**			**	*	*		*
**	**	*	*	***	*	***	***	**	*	**	**
*	**	*	*	*	*	*	*	*	*		*
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				*			*	**	***		**
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*			*	***			***	**	*		**
*	*		*	*			*	**	*	*	*
**	***	****	***	*	***		*	**	**	*	**
**	**	*	**	*	*	*	*			*	*
*	**	***	*		*	*	*		*	*	*
*			*	*			*	*	*	*	*
****	****	**	****	****	***		****	****	***	*	***
**	***	*	**	*			*		*	*	*
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*	*	***	*		**	*	*				
***	****	****	****	**	***	*	**	*	*	*	*
								*	**	***	**
***	*		**	***	*		***	****	**	*	***
*	*		*	*			*	**	*		*

Water depth (WD) and sediment layer (SL) community data comparisons showed significant differences, as well as for the WD x SL interaction (PERMANOVA, $p<0.01$, Table 4.1). According to the Estimated Component of Variation (ECV), variability was much higher for SL than for WD and replicate factors (Table 4.1).

Overall, there was a bathymetric division of samples in terms of nematode composition, with four distinct groups: Upper (BC500, BC900 and BC1200), Deep (BC2000) and two Middle stations separated (Mid-Upper, BC1500 and Mid-Deep, BC1750) ($p< 0.05$, pair-wise comparisons, Table S6). These groups can be distinguished at the 58 % of similarity level (Fig 4.5A). The average relative abundance of the genera contributing most to similarity within stations as identified by the SIMPER analysis is shown in Table 4.4. *Sabatieria* and *Halalaimus* were amongst the most abundant genera along the canyon (SIMPER analyses, Table 4.4). In general, genera that contributed most to the intra-station similarity at the Upper canyon were *Cervonema, Sabatieria* and *Halalaimus,* as well as *Setosabatieria* and *Sphaerolaimus* at BC500, *Daptonema* and *Acantholaimus* at BC900 and *Actinonema* and *Dichromadora* at BC1200 (Table 4.4). *Sabatieria*, *Halalaimus* and *Sphaerolaimus* contributed most to BC1500 similarity, while *Molgolaimus*, *Halalaimus*, *Acantholaimus* and *Sabatieria* contributed most to BC1750 similarity. Finally, in the Deep canyon (BC2000) the genus contributing most to similarity was *Sabatieria,* followed by *Acantholaimus* and *Halalaimus* (Table 4.4). The highest dissimilarity occurred between Upper stations and the Deep canyon station (dissimilarity SIMPER analyses, BC500, BC900, BC1200 *vs*. BC2000= 57.9, 59.2 and 56.08% respectively, data not shown).

The differences in nematode community structure between Upper and Mid-Deep/Deep canyon occurred mainly in the first 2 cm of the sediment (Table S6, pairwise comparisons within WD x SL, $p< 0.05$; Fig. 4.5B). There were no significant differences between stations for the deep 2-5 cm layer (Fig. 4.5B). The nMDS plot shows a clear bathymetric trend, with the Upper canyon, Deep canyon and the two Middle canyon groups clearly separated. Sediment layers also show a marked pattern in the nMDS plot, with the deepest sediment layers positioned away from the surface layers, with the subsurface layers between. The observed community differences between sediment layers were most pronounced at BC1750 (Table S6, pairwise comparisons within WD x SL; Fig. 4.5B).

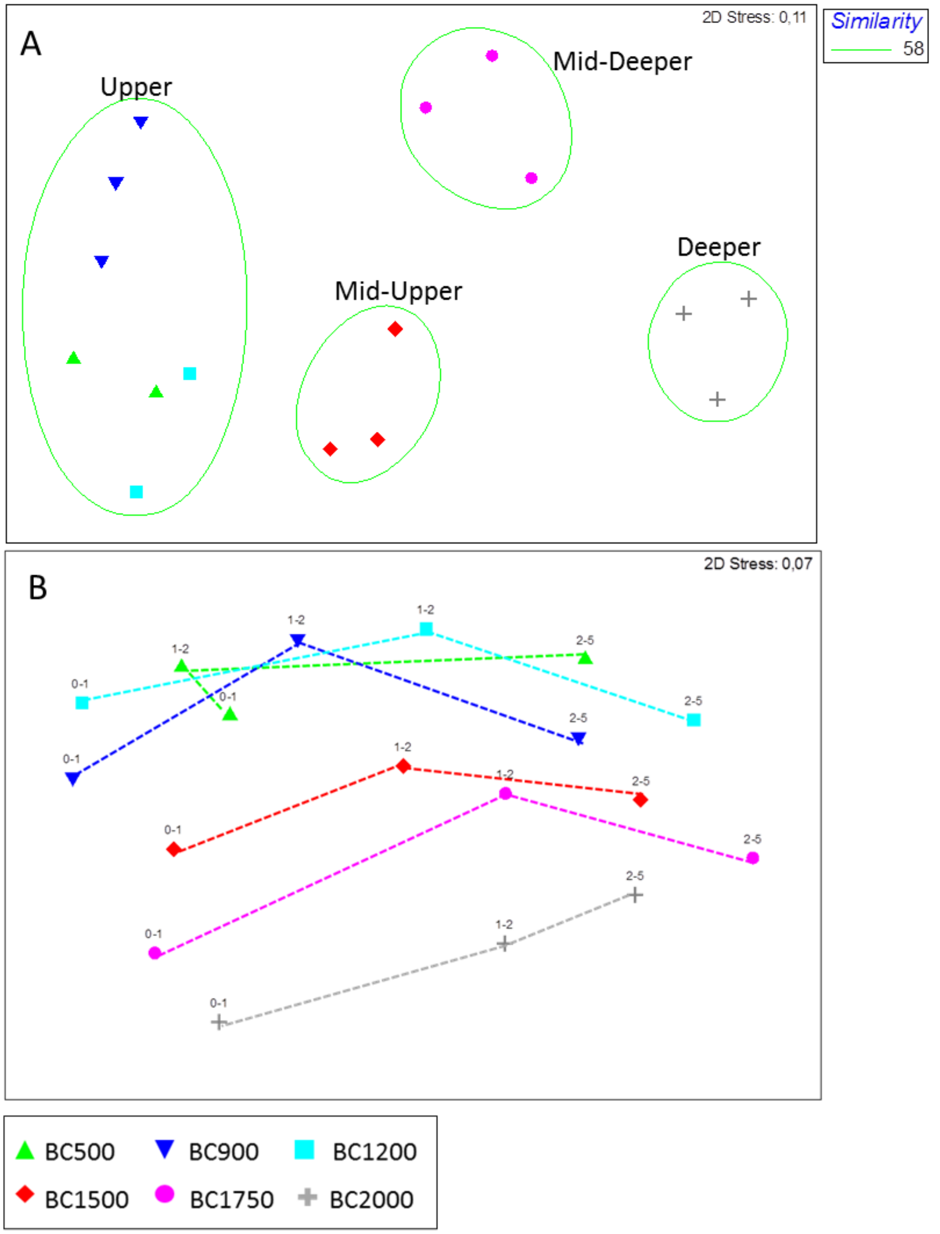


Figure 4.5. Non- metric MDS plots based on standardized nematode genera data over (**A**) water depth and (**B**) averaged over water depth and sediment layer, 0-1, 1-2 and 2-5 (in cm).

Table 4.4. Average relative abundance (%) of the genera responsible for the similarities within stations based on a SIMPER analysis (>2%).

	BC500	BC900	BC1200	BC1500	BC1750	BC2000
Cervonema	**13.20**	**8.46**	**5.6**	3.5	3.72	
Sabatieria	**10.55**	7.88	**13.35**	**15.03**	**7.72**	**21.57**
Halalaimus	**7.25**	**13.73**	**14.94**	**8.68**	**9.03**	**6.61**
Setosabatieria	**6.76**	1.87	**7.63**			
Sphaerolaimus	5.53	3.10	2.26	8.17	3.88	
Pomponema	3.90	1.88	3.68	4.84		
Chromadorella	4.40					3.85
Disconema	2.73	2.14		3.21	1.48	
Elzalia	2.60					
Paramonohystera	2.61			0.84		
Metasphaerolaimus	2.43			1.11	4.19	4.06
Daptonema	2.94	7.39	2.86	3.21	2.41	1.29
Tricoma	1.93	3.42	1.33	3.17	**6.73**	5.36
Campylaimus	2.00	1.86	3.59			
Monhystrella	1.86	1.13			2.11	2.39
Leptolaimus	2.03			1.17		1.35
Dichromadora	2.30	**8.59**	4.13			1.69
Acantholaimus	1.38	5.16		3.94	**8.39**	**9.51**
Actinonema		4.66	4.29	3.00		
Marylynnia	1.26	1.33		2.17	2.82	
Desmoscolex		1.44			4.08	
Thalassomonhystera		2.1				1.34
Amphimonhystrella		1.24	1.43	1.87	4.54	
Paramesacanthion		0.64			3.47	
Diplopeltula			3.14	4.34	1.41	2.83
Dorylaimopsis			2.90	1.75		
Oxystomina			0.98	4.23		1.94
Halichoanolaimus				2.86		
Pselionema				2.45	2.94	
Molgolaimus					**9.56**	4.80
Syringolaimus						3.34

4.3.2.4 Feeding ecology

Deposit feeders dominated at all stations (1B + 1A represented from 64%- 69%) of the communities (Table 4.5). Stations BC500, BC900 and BC1200 were dominated by 1B (44% ± 5), while at BC1500, BC1750 and BC2000 1A was dominant (37% ± 5) particularly BC1750 (42%). The trophic groups 2A and 2B were more abundant at BC900 and BC1200 and at BC1500, respectively. The gutless nematode (*Astomonema*) was found at all sampling stations except at BC2000, and mainly occurred in the deepest sediment layer (2-5 cm) (Table 4.5).

Table 4.5. Relative abundance of the nematode feeding types along the sediment profile at each studied station. 1A: selective deposit feeders, 1B: non-selective deposit feeders, 2A: epistratum feeders, 2B: predators/scavengers, 3: chemosynthetic feeder

Station	Sediment layer (cm)	1A	1B	2A	2B	3
BC500	0-1	18.92	47.49	21.52	12.07	-
	1-2	23.53	41.32	20.23	14.92	-
	2-5	8.33	78.63	6.50	5.53	1.01
	0-5	**18.78**	**48.89**	**17.90**	**14.6**	**0.17**
BC900	0-1	30.84	33.99	28.85	6.31	-
	1-2	21.35	46.06	25.51	6.79	0.28
	2-5	5.35	62.90	18.03	11.22	2.49
	0-5	**26.10**	**39.58**	**26.95**	**7.01**	**0.36**
BC1200	0-1	34.99	27.02	27.71	10.28	-
	1-2	16.85	60.28	11.92	10.00	0.94
	2-5	3.13	73.60	13.09	7.68	2.50
	0-5	**25.70**	**41.99**	**21.91**	**9.77**	**0.63**
BC1500	0-1	32.23	24.66	25.63	17.47	-
	1-2	23.82	39.27	14.17	22.29	0.45
	2-5	15.09	53.46	14.23	15.24	1.98
	0-5	**33.08**	**31.40**	**15.86**	**19.23**	**0.44**
BC1750	0-1	44.59	23.71	27.69	13.01	-
	1-2	19.34	52.27	8.64	19.75	-
	2-5	2.53	79.33	7.84	3.31	6.99
	0-5	**42.04**	**26.55**	**17.50**	**13.69**	**0.22**
BC2000	0-1	43.64	23.50	27.69	4.95	-
	1-2	22.79	46.39	21.30	9.51	-
	2-5	8.91	61.87	17.74	11.48	-
	0-5	**34.82**	**33.22**	**24.83**	**7.13**	**-**

The nematode genera with the highest nematode densities did not represent a particular feeding type: BC500: *Cervonema* (1B), *Halalaimus* (1A), *Sphaerolaimus* (2B); BC1200: *Halalaimus* (1A); and BC1750: *Acantholaimus* (2A), *Halalaimus* (1A), *Molgolaimus* (1A).

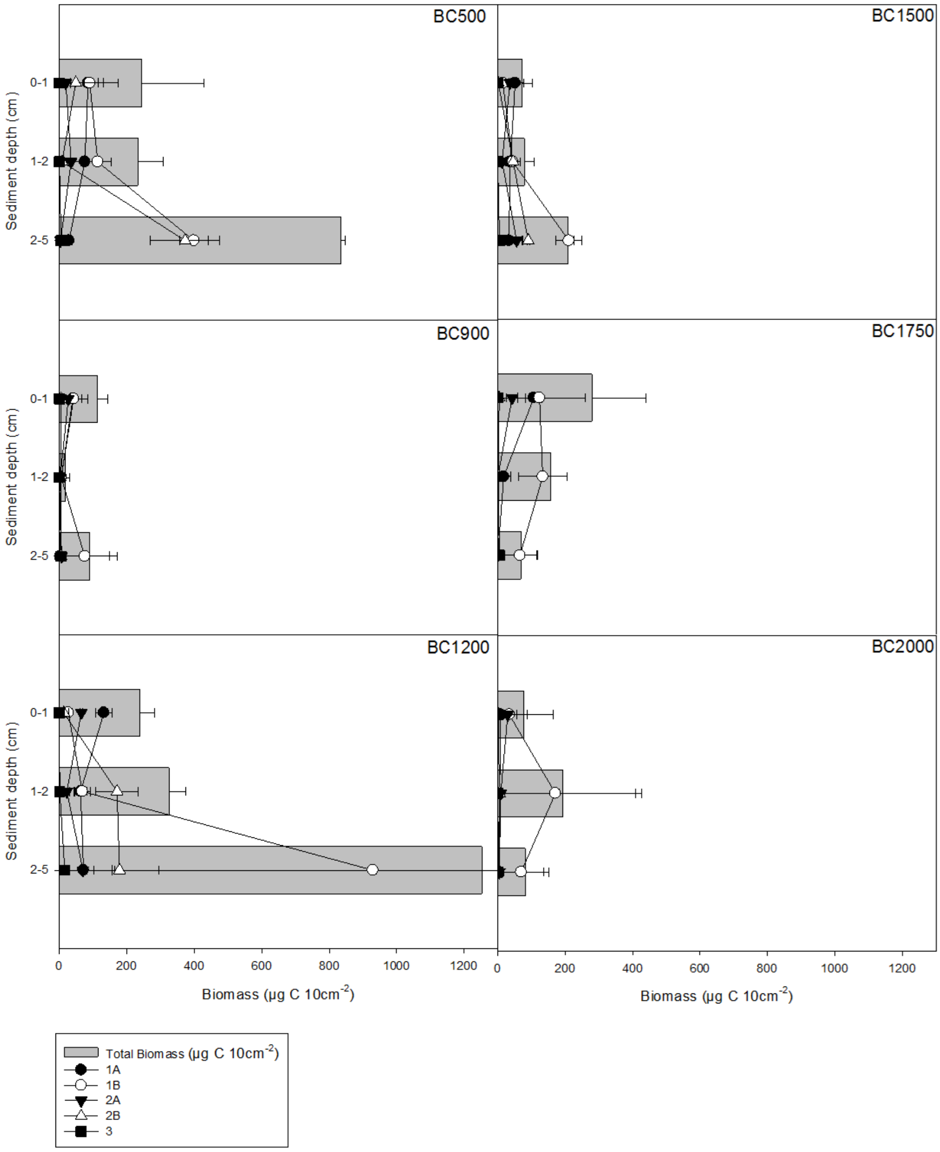


Figure 4.6. Total and trophic group biomass values for each station along the sediment profile. 1a: selective deposit feeders, 1B: non-selective deposit feeders, 2A: epistratum feeders, 2B: predators/scavengers, 3: chemosynthetic feeders.

Surface sediment layers exhibited higher trophic diversity than the deep layers (Table 4.2), mainly because of the dominance (i.e. 52 to 79%) of 1B in the deeper layer (2-5 cm) (Table 4.5). Feeding type 2A decreased in abundance along the sediment profile (p < 0.05, data not shown, see Table 4.5).

The contribution of the feeding groups to the total biomass changed along the bathymetric gradient, with 1B contributing most at all stations except at BC1500 and BC1750, where it was replaced by 2B biomass was highest (Fig. 4.6). The biomass contributions of the different feeding groups were similar at BC900 and BC2000, while group 1B dominated at BC500 and BC1200, particularly in the deep sediment layers (Fig. 4.6). At BC1750, biomass of the group 2B dominated total biomass in the surface sediment layer due to the presence of *Paramesacanthion*.

4.3.3 Relationships between environmental variables and nematode assemblages

Kendal-Tau correlations between structural community parameters and environmental variables are shown in Table 4.6. Density and biomass were positively correlated with TN and Chl *a* (Table 4.6). Density was negatively correlated with clay content, and biomass showed a positive correlation with Chl *a*: phaeo. TD and MI were positively and negatively correlated with clay and silt content respectively. *H´* and *EG*(51) were positively correlated with silt content and *H´* also showed a negative relation with clay.

Table 4.6. Kendal-Tau correlations coefficients between biotic and abiotic variables. Chl *a*: chlorophyll a, CPE: chloroplastic pigment equivalents, Chl *a*: phaeo: chlorophyll a divided by its degradation products (phaeopigments), TN: total nitrogen content, OC: organic carbon content, C:N, molar carbon-nitrogen ratio, TD: trophic diversity index, H´(Shannon index), *EG*(51): estimated number of genera. Bold: p<0.05; Bold-italics: p<0.01.

Descriptor	Chl *a*	CPE	Chl *a*: phaeo	TN	OC	C:N	Clay	Silt	Sand
Density	***0.207***	0.181	0.017	***0.257***	0.179	0.007	***-0.311***	0.130	0.185
Biomass	***0.255***	0.156	**0.127**	***0.344***	0.121	-0.143	-0.154	0.036	0.123
TD	0.042	0.010	0.148	-0.075	-0.042	-0.018	**0.210**	**-0.251**	-0.019
H´	0.104	0.123	-0.081	0.159	0.127	0.032	**-0.262**	**0.217**	0.075
EG(51)	0.091	0.090	-0.066	0.144	0.127	0.037	-0.195	**0.203**	0.038

The DISTLM analysis based on nine environmental variables explained 51.4% of the variability of the standardized (%) assemblage structure (Fig. 4.7, Table S7), with CPE explaining a 24.5% followed clay content (9%).

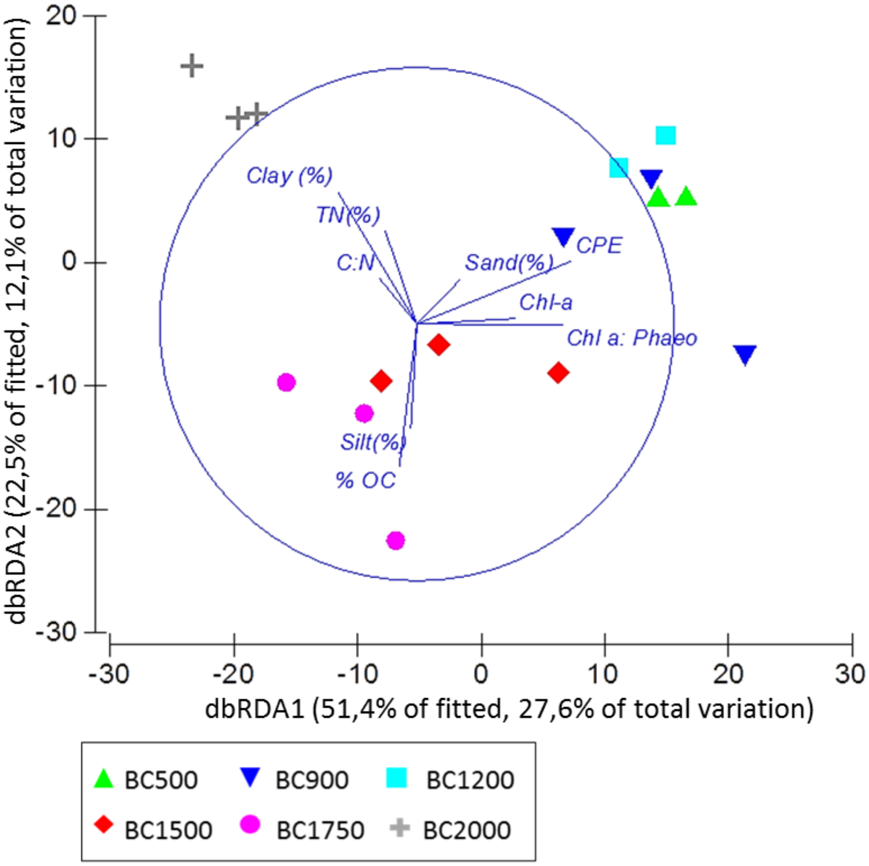


Figure 4.7. Distance-based redundancy (dbRDA) illustrating the DISTLM model based on genera assemblage data and fitted environmental variables with their vector (strength and direction of effect of the variable on the ordination plot).

Using the dominant genera, significant positive Kendal Tau correlations were found with Chl *a* and CPE respectively for *Setosabatieria* (0.684 and 0.711); *Cervonema* (0.661 and 0.594) and *Actinonema* (0.444 and 0.611); with Chl *a*: phaeo for *Cervonema* (0.594), *Daptonema* (0.417), *Halalaimus* (0.417), *Paramonohystera* (0.454), *Pomponema* (0.393) and *Setosabatieria* (0.532); with clay content in *Sabatieria* (0.467) and *Syringolaimus* (0.563); with silt content in *Marylynnia* (0.444), *Pselionema* (0.444), *Sphaerolaimus* (0.400) and *Tricoma* (0.417) and with sand content in *Cervonema* (0.393), *Dichromadora* (0.510), *Setosabatieria* (0.464). Negative correlations with Chl *a* were found for *Acantholaimus* (-0.533), *Sabatieria* (-0.383) and *Syringolaimus* (-0.479); with CPE for *Acantholaimus* (-0.567), *Molgolaimus* (-0.380) and *Syringolaimus* (-0.479); with Chl *a*: phaeo for *Acantholaimus* (-0.417), *Amphimonhystrella* (-0.417), *Sabatieria* (-0.400) and *Syringolaimus* (-0.563). Related to sediment grain size, clay content was negatively correlated with *Actinonema* (-0.427), *Halalaimus* (-0.417), *Pomponema* (-0.460) and *Sphaerolaimus* (-0.450), silt with *Dichromadora* (-0.444) and sand content with *Pselionema* (-0.460) and *Tricoma* (-0.417).

4.4 DISCUSSION

4.4.1 Nematode density and biomass indicate high environmental canyon heterogeneity

The nematode densities observed in the Blanes Canyon are among the highest reported for NE Atlantic and Mediterranean canyons at similar depths (Fig. 4.8A), and were comparable to those from shallower Iberian shelf areas (779-1802 ind. 10 cm^{-2}) (Vanreusel et al., 1992; Flach et al., 2002). Moreover, they also differ from the data previously reported by Ingels et al. (2013) on nematode standing stocks from the Blanes Canyon at similar depths (Fig. 4.8A). These differences between both Blanes studies could be attributed either temporal variability in nematode standing stocks (Ingels et al., 2013) or to the high spatial patchiness in the canyon and related to alternating erosive and deposition areas. Biomass in the Blanes Canyon showed the same patter, with higher values compared to Mediterranean canyons at similar depths (except at ca. 900 m depth). Unusual high values (about tenfold) were recorded in the upper Nazaré canyon (500-1000m) by Bianchelli et al. (2010) mainly due to the higher values in the supply of OM (2-8 times) than in Mediterranean regions. The in our study reported biomass was also higher than that previously reported from Blanes Canyon at comparable depths (Fig. 4.8B; Ingels et al., 2013).

Quality and quantity of food have been documented as key factors controlling the meiobenthos and nematodes densities (e.g., Ingels et al., 2009; Tselepides and Lampadariou, 2004; Grémare et al., 2002; Román et al., 2016), and nematode biomass (Vanreusel et al., 1995; Grémare et al., 2002; Leduc et al., 2012a) in deep-sea ecosystems. A decrease in densities and biomass with increasing water depth caused by the bathymetric decline in arriving detritus from surface waters is therefore commonly found in the deep sea (Leduc et al., 2012a; Soetaert and Heip, 1995; Soetaert et al., 1997; Vanaverbeke et al., 1997). In the Blanes Canyon the effect of both food quality and quantity on nematode density and biomass is illustrated by the positive correlations with Chl *a* with both, and Chl *a*: phaeo, only for biomass (Table 4.6), which supports findings for other canyon systems (Garcia et al. 2007; Ingels et al., 2009; Ingels et al., 2011a,b). However, this relationship between food quantity and quality and nematode density and biomass does not reflect a bathymetric trend, which is clearly disrupted at BC1200 and BC1750 (Fig. 4.3). In fact, the lack of a gradual bathymetric decrease was also observed for total meiofaunal densities at Blanes Canyon because of enhanced densities at BC1200 which remained consistent over several years (2012 and 2013) and seasons (spring and autumn) (Román et al., 2016). The elevated standing stock at BC1200 coincided with significant increases in food availability, and was reflected in other environmental variables pointing to a disturbed and depositional environment (i.e., higher CPE and sand contents). In **Chapter 3** we postulated that the sediment enrichment in terms of sand contents and food supply at 1200 m depth in the canyon was likely caused by lateral advection of particles through gullies at the northern flank of the canyon, which support the presence of an anthropogenic depocenter as a result of trawling activities and lead to increasing sediment deposition rates (see **Chapter 3**). Several studies (Rucabado et al., 1991; Sardà et al., 2004) have reported a maximum biomass around 1200 m depth in fish populations in the NW Mediterranean area, whilst peaks in biomass of benthic and suprabenthic communities of fish and decapod crustaceans have also been reported between 1150 and 1250 m depth in other deep Mediterranean areas (Stefanescu et al, 1993; Sardà et al., 1994b; Moranta et al., 1998; D'Onghia et al., 2004; Company et al., 2004). High abundance and biomass of meiofauna and nematodes have also been reported in the Nazaré (Garcia et al., 2007) and Whittard canyons (Gambi and Danovaro, 2016; Ingels et al., 2011a) around 1000-1100 m depth. Further analyses of the temporal variations would be required to assess whether these peaks are caused by comparable processes and, particularly,

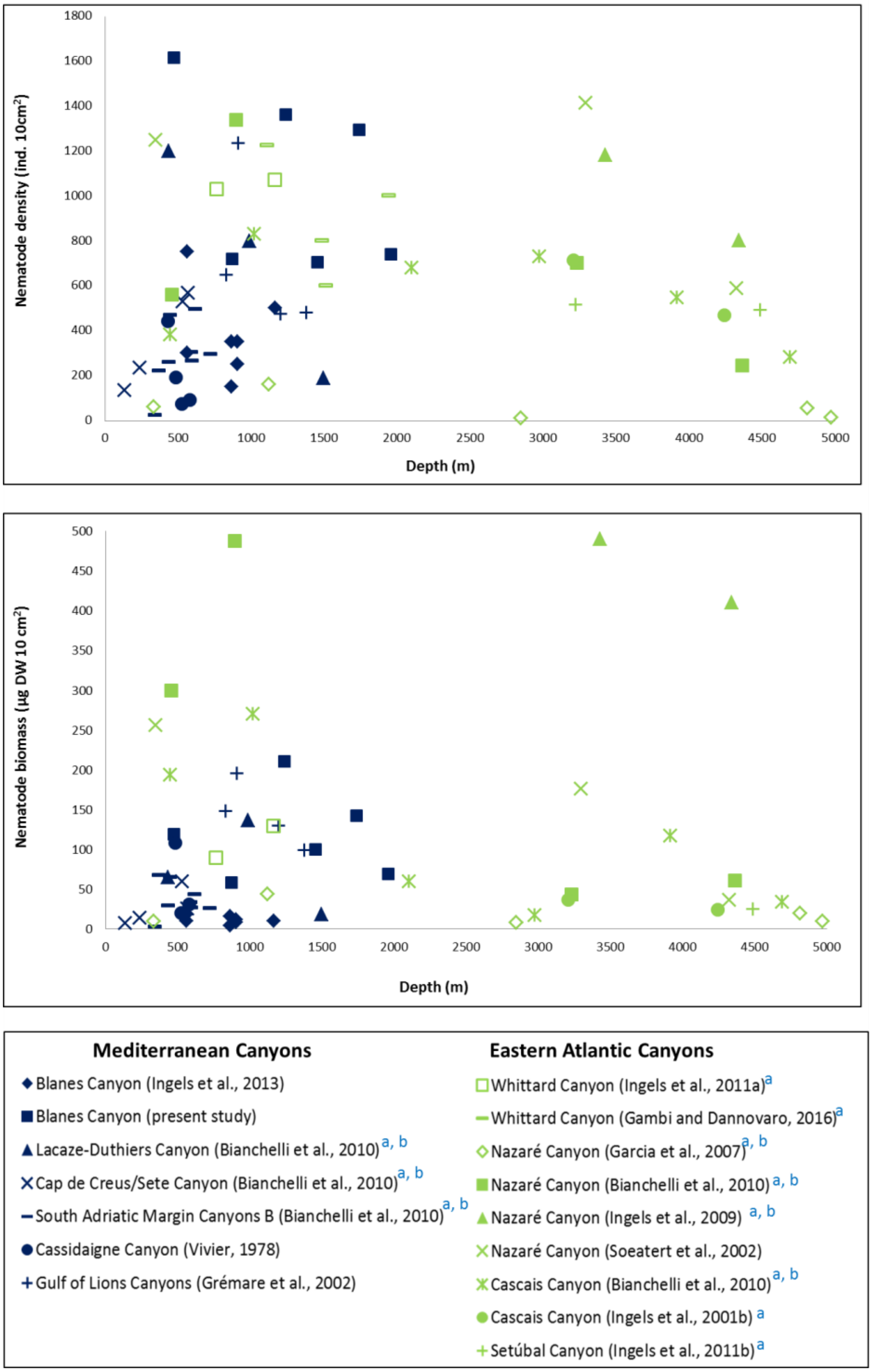


Figure 4.8. Nematode densities (A) and biomasses (B) recorded from Mediterranean and East-Atlantic canyons. [a] Estimated from figure. [b] Assuming dry weight: wet weigh ratio of 0.25.

whether these high densities are a response to the presence of an organic matter and sedimentary deposition area. Ingels et al. (2011) certainly seem to suggest this is the case for the 700m and 1000m station in the Whittard Canyon, where it was suggested that the high levels of organic matter deposition may have led to reduced conditions in the sediment which are also reflected in the nematode community compositions.

Below 1200 m depth, the amount and quality of organic matter available for direct consumption decreased with increasing water depth (Table S1, Fig. 4.3). Consequently, the unexpected high densities and biomass observed at BC1750 are likely caused by environmental conditions that are not directly related to food quality and quantity. Complexity in deep-sea environments may result from changes in the combined effects of physical processes, bioturbation and food supply along depth gradients (Vanhove et al., 2004). This is particularly valid for canyons where high currents often resuspended and transport surface sediments leading to unstable substrate conditions, whereas locally high sedimentation rates may lead to fauna being buried and being swept away from elsewhere (Garcia et al., 2007). Recent investigations have pointed out that many canyons undergo flushing events following cold water cascading episodes and gravity flows, which may cause episodic, but dramatic removals of particulate material and organisms from the upper canyon areas layers towards the adjacent bathyal and abyssal plane (De Stigter et al., 2007; Canals et al., 2006). The Blanes Canyon is one of them. It has a sinuous system and shows a high topographical heterogeneity whereby the currents are altered by the canyon morphology (Zúñiga et al., 2009). Moreover, deep-sea organisms are highly sensitive to the arrival of external inputs. Therefore, canyons channelling organic matter are often areas of increased biomass and productivity, which can exceed those of other deep-sea habitats by orders of magnitude, depending on the canyon (Tyler et al., 2009; De Leo et al., 2010; Vetter et al., 2010; Huvenne et al., 2011). At BC1750, *Molgolaimus* is one of the genera responsible for the high densities observed, mainly due to its high abundance in the surface sediment layers (Table 4.3). This genus occurs in all oceans from shallow waters to the deep sea, and it occurs in high densities in different deep-sea habitats such as soft slope sediments along the Western Indian Ocean (Muthumbi and Vincx 1996), hydrothermal vents (Vanreusel et al., 1997), the Weddell Sea (Vanhove et al., 1999; Lambshead et al., 2000), and the South and Equatorial Pacific Ocean (Gambi et al., 2003; Lambshead et al., 2003) down to 2000 m depth. Its occurrence is often associated with organically enriched and recently disturbed areas by the constant shifting of large amounts of sediment such as those resulting from iceberg scouring (Vanhove et al., 1999; Lee et al; 2001). We hypothesize that the high densities observed at BC1750 could be a response to previous deposition events in the canyon. Our sampling was carried out in autumn 2012, while in February 2012; a dense shelf cascading event was detected in the NW Mediterranean (Durrieu de Madron et al 2013). Step-by-step transport along the canyon involving the resuspension of particles previously accumulated in its shallower reaches may have contributed to the arrival of additional organic matter reaching the deepest areas of the Blanes Canyon (Pasqual et al., 2010). Alternatively, the high levels of organic carbon at 1750 m depth (higher than at BC900, BC1200 and BC1500) could be explained by sediment gravity flows channelled by the gullies that incise the Blanes Canyon walls, as has been previously reported for similar depth in La Fonera Canyon (Martín et al., 2008) which may also led to direct deposition of fauna from shallow bottoms, but also to increase the sediment deposition rates (**see Chapter 3**).

The distribution of infaunal organisms along the vertical profile in deep-sea sediments can be linked to quality and quantity of the arriving organic matter (Thiel, 1983; Lambshead et al., 1995), the oxygenation (Shirayama and Horikoshi, 1982) a combination of both (Shirayama, 1984; Vanreusel et al., 1995), as well as the degree of sediment mixing and disturbance near the surface (Carman et al., 1987). These factors are believed to drive gradients in nematode community, abundance and taxonomic composition. In the present study, nematode densities were mainly concentrated in the upper cm, as has been found in many other deep-sea sediments in slope habitats (Soetaert et al., 1991a; Vanaverbeke et al., 1997; Vanreusel et al., 1995), and submarine canyons (Ingels et al., 2009; Ingels et al., 2011a,b). However, there are differences among stations (Fig. 4.4) with for instance nematode densities decreasing gradually with increasing sediment depth at BC1500 and BC2000, but subsurface density maxima at BC500. Nematode densities in deeper sediment layers are potentially affected by oxygen limitation and levels of organic matter deposition, especially in the upper stations (BC500, BC900 and BC1200) where high sedimentation rates and burial may cause reduced conditions (Table S1, Fig. 4.4). Indeed, the dominance of *Sabatiera* in deeper layers (Table 4.3), suggests the presence sediments with reduced oxygen availability (Soetaert and Heip, 1995, Vanreusel et al., 1997; Muthumbi et al., 2004). Remarkable in this respect is also the occurrence of the "chemosynthetic" nematode *Astomonema*, in deeper layers particularly at BC1200, which indicates a reduced environment (Ingels et al., 2011b) since they are reliant on sulphide oxidising bacteria they carry within their bodies. This nematode genus has been observed before in high numbers in the Whittard Canyon and Gollum Channel sediments which were characterized by high organic carbon content (0.2-1.3%, Ingels et al. 2011a).

4.4.2 Nematode community structure, composition and function indicate bathymetric canyon zonation

Contrary to the nematode density and biomass distribution along the canyon axis, the nematode communities based on genera relative abundances revealed to be bathymetrically structured along the canyon axis (Fig. 4.5A). Although most nematode genera (109 in total) occurred at all depths, there was a shift in the nematode assemblages around BC1200 (Figs. 4.2, 4.5A, 4.7) after which food quality and grain size decreased with increasing depth (Fig. 4.3, Table S1). Despite the fact that deposit feeders were consistently dominant, as observed for deep-sea sediments worldwide (Soetaert and Heip, 1995; Gambi et al., 2003; Vanhove et al., 2004; Danovaro et al., 2008; Vanreusel et al., 2010), there was also a shift at BC1200, where non-selective deposit feeders started to decrease in relative abundance compared to selective deposit feeders. Structural and functional nematode community characteristics suggest a "bathymetric" change at BC1200, with deeper station communities being characterised by different genera and feeding type distributions. Nematode community structure suggested three distinct groups or depth categories of stations in the nMDS analyses (Fig. 4.5) and this bathymetric zonation was maintained when sedimentary variables were used for multivariate analysis (Fig. 4.7). Accordingly, the canyon axis can be subdivided into 3 areas

The **Upper canyon part (BC500, BC900 and BC1200)** is characterized by poorly sorted sediments with high food content (Chl *a* and CPE) and poorly sorted (Fig. 4.2, Fig. 4.7). The assemblages are dominated by *Setosabatieria, Sabatieria*, *Halalaimus* and *Cervonema*. *Cervonema,* and *Setosabatieria*, were correlated with food quality (Chl a, CPE). These communities are characterized by high abundance of non-selective feeders, which is related to the high organic matter content and freshness of the organic matter available in the sediments (Vanhove et al., 1999).

The **Middle canyon (BC1500, BC1750)** is characterized by high silt content, elevated OC concentrations but of a lower quality (Fig. 4.2) and the dominance of selective-deposit feeders. The two stations are not entirely similar, dominated by *Sabatieria*, *Halalaimus* and *Sphaerolaimus* (BC1500) and by *Molgolaimus*, *Halalaimus* and

Acantholaimus (BC1750). The high densities of the predator and scavenger *Sphaerolaimus* has been observed in canyons before (Danovaro et al., 2009; Gambi and Danovaro, 2016). *Molgolaimus* is mainly responsible for the elevated densities at station BC1750, while *Acantholaimus* is a genus that generally increases in relative abundances with increasing water depth (Vanreusel et al, 2010). Its negative correlation with food quality supports the assumption that this genus is a persister, which would explain its increasing success with increasing water depth.

The **Deep canyon (BC2000)** sediments are characterized higher clay content and C: N contents compared to the sediments at the other stations. A high C: N ratio may the results of preferential removal of nitrogen over time and distance from near-shore production, and serves as indicator of poor quality of OM or increased levels of terrestrial OM. There are also higher levels of OC compared to shallower stations, which may be related to high deposition rates and subsequent reduced sedimentary conditions which shape the nematode assemblages (Soetaert et al., 2002; Ingels et al., 2009), whit for instance high abundance of opportunistic genera such as *Sabatieria*. The dominant genera present in this station (*Sabatieria, Acantholaimus* and *Halalaimus*) are similar to those reported in NE Atlantic canyons such as the Setubal Canyon at 3200 m depth (Ingels et al., 2011b) and Gollum Channels at 1000 m depth (Ingels et al., 2011a).

The changes in nematode assemblages observed here along the Blanes Canyon are consistent with the patterns observed in other canyon and open slopes systems in which the genus composition gradually changes along a bathymetric range (e.g., for shelf-slope transect: Vanaverbeke et al., 1997; Vanhove et al., 1999; and canyons: Garcia et al., 2007; Gambi and Danovaro, 2016). In slope systems, the variability of nematode genera composition is usually attributed to changing food input and oxygen levels as a consequence of bathymetry (Soetaert and Heip, 1995; Soetaert et al., 1997; Vanhove et al., 1999). In submarine canyons on the other hand, food availability (Garcia et al., 2007; Ingels et al., 2011a, b), but also grain size (Ingels et al., 2011a), along with oxygen availability, sedimentation rates as well as hydrodynamic disturbance and episodic events are used explain the observed patterns and are generally related to the topographical and oceanographic conditions that typify the canyons.

Despite some canyon effects on resident nematode assemblages, general patterns of certain nematode genera responding to deep-sea environmental conditions remain strong. This can be illustrated by the genus *Acantholaimus* for instance.

Acantholaimus is a typical deep-sea genus (Platt and Warwick, 1988) that becomes more abundant with increasing water depth (Soetaert and Heip 1995; Vanaverbeke et al., 1997; Muthumbi et al 2004; Vanhove et al., 2004). It also seems to increase in abundance when food availability decreases (Vanaverbeke et al., 1997; Muthumbi et al 2004) although this observation has recently been challenged by Lins et al. (In press). In our samples, high densities of *Acantholaimus* were associated with low Chl *a* and CPE contents, in agreement with De Mesel et al. (2006) for the Antarctic shelf sediments and Lampadariou and Tselepides (2006) for the Aegean Sea. *Acantholaimus* was in fact a highly abundant genus at all water depths in the Iberian Setúbal and Cascais canyons (Ingels et al. 2011b) and it was more abundant than at the adjacent slope at similar depths in deep regions of the Nazaré Canyon (Ingels et al. 2009).

Small-scale variability (on the cm scale) and associated biogeochemical changes have been postulated as the most important spatial source of variability for the structural and functional diversity variability in deep-sea infauna communities (Ingels and Vanreusel, 2013). Our study confirms this observation with the vertical sediment profile is having a greater impact on nematode the community structure than the differences in water depths in the Blanes Canyon. Nematode diversity gradually decreased with sediment depth (Table 4.2), in accordance with several slope (e.g. Vanaverbeke et al., 1997; Soetaert et al., 1991b; Soetaert et al. 1995) and canyon (Ingels et al. 2011a) studies. Moreover, our data shows that the nematode assemblage differences between water depths are more prominent in surface layers (i.e., 0-1 and 1-2 cm) than in the deep layers (2-5 cm) (Fig. 6B), probably due to the high environmental heterogeneity that characterises the stations at the sediment-water interface. In turn, deeper layers were more similar in terms of community composition because they were dominated by *Sabatieria*, which is known to colonize anoxic to sub-oxic sediments (Soetaert and Heip, 1995, Vanreusel et al., 1997; Muthumbi et al., 2004). Conversely, other submarine canyons at the Portuguese (Nazaré, Cascais and Setúbal Canyons) (Ingels et al., 2009; 2011b) or Celtic (Whittard canyon, Ingels et al., 2011a) margins, as well as the continental slope of the Goban Spur (Vanaverbeke et al. 1997) and a Mediterranean deep-sea transect (Soetaert et al., 1995), showed nematode communities that were similar in the first cm of the sediment and then changed as one goes deeper into the sediment. The vertical structure of nematode communities in the sediment is most likely related to the differential ability of the different genera to reside in, or to penetrate into, the different sediment layers and has often been associated with the biogeochemical processes and variables such as oxygen availability in the sediments (Soetaert et al., 2002; Soetaert et al., 2009).

4.4.3 Nematode diversity

Factors such as habitat heterogeneity (Levin et al. 2001, Vanhove et al. 2004) and changes in food availability and supply (Lambshead et al. 2000, 2002) have been used to explain deep-sea biodiversity distribution, but a variety of oceanographic conditions at specific depths may interrupt and modify bathymetric horizontal diversity trends (Levin et al., 2001). In submarine canyons, because of their physically complex habitats, predictions on diversity are not straightforward since a variety of environmental and physical sedimentary characteristics (e.g. quantity and quality of OM, temperature, topography and current regime) of the sediments may also modify the biodiversity patterns (e.g., Danovaro et al., 2009; De Leo et al., 2010).

In the NE Atlantic and Mediterranean canyons, different bathymetric trends in nematode diversity patterns have been previously reported (Vivier, 1978; Garcia et al., 2007; Danovaro et al., 2009; Ingels et al., 2011a; Gambi and Danovaro, 2016). Our results showed that there is no consistent diversity gradient along the Blanes Canyon where diversity seems independent of the sedimentary variables we measured. We did, however, see a relation between diversity and clay and silt content (Table 4.6). Sandy sediments can provide a wide range of microhabitats for meiofauna organisms which may partially explain the slightly higher diversity observed in the sediment with highest sand content (BC500 and BC1200). Sediment grain size an important variable when explain meiofauna and nematode communities as it provides insight into the physical structure, porosity, and permeability and hence the living space characteristics and food availability of interstitial fauna. Accordingly, grain size has been shown to partially explain the variability in nematode diversity among canyon´s branches and open slopes at Whittard Canyon (Gambi and Danovaro, 2016).

Finally, the genus richness in Blanes Canyon is higher than was reported for the Nazaré Canyon (García et al., 2007) (55 genera), lower than at 700 – 1000 m depth (112 and 84 genera) in the Whittard Canyon (Ingels et al., 2011a) and similar to expected number of genera (25-22 respectively) in this canyon at 1483 and 1958 m depth (Gambi and Danovaro, 2016). In Mediterranean canyons, the only comparable information comes from Cassidaigne Canyon, which showed a similar richness from 430 to 580 m depth (Vivier, 1978).

4.5 SUPPLEMENTARY DATA

Table S4.1. Average values for selected environmental variables along the sediment profile and at each station. Clay, Silt, Sand: volume percent clay, silt and sand content respectively; Chl *a* (Chlorophyll *a*), CPE (Chloroplastic pigment equivalents), Chl *a*:phaeo (Chlorophyll *a*: phaeopigments), OC (Organic carbon, %), TN (Total Nitrogen %), C:N (carbone: nitrogen ratio. Values average over replicates; standard deviations are not shown.

Station	**Layer** (cm)	**Clay** (%)	**Silt** (%)	**Sand** (%)	**OC** (%)	**TN** (%)	**C:N**	**CPE** (μg/g)	**Chl *a*** (μg/g)	**Chl *a*:phaeo** (μg/g)
BC500	0-1	18.37	75.29	6.32	0.89	0.11	7.73	1.91	0.06	0.04
	1-2	19.96	71.25	8.78	0.90	0.11	8.22	0.92	0.02	0.03
	2-5	19.50	72.94	7.55	0.89	0.11	7.78	1.22	0.03	0.03
	0-5	**19.28**	**73.16**	**7. 55**	**0.9**	**0.11**	**7.92**	**4.07**	**0.13**	**0.05**
BC900	0-1	17.50	71.70	8.65	0.79	0.09	8.17	1.52	0.02	0.03
	1-2	17.91	73.79	8.29	0.71	0.09	7.92	1.03	0.02	0.02
	2-5	19.26	71.70	9.02	0.7	0.09	7.50	0.89	0.01	0.01
	0-5	**18.23**	**73.11**	**8.65**	**0,73**	**0.09**	**7.87**	**3.45**	**0.07**	**0.07**
BC1200	0-1	17.40	67.20	13.30	0.71	0.09	7.39	1.17	0.02	0.05
	1-2	19.19	70.81	9.99	0.73	0.09	7.83	1.90	0.03	0.04
	2-5	20.81	68.37	10.68	0.71	0.09	7.98	1.17	0.03	0.02
	0-5	**19.01**	**68.89**	**12.5**	**0.75**	**0.09**	**7.74**	**4.70**	**0.10**	**0.06**
BC1500	0-1	17.26	77.77	4.96	0.76	0.10	7.33	0.54	0.01	0.005
	1-2	18.18	77.70	4.10	0.75	0.10	7.26	0.93	0.02	0.01
	2-5	20.00	75.72	4.27	0.76	0.11	7.00	0.54	0.01	0.02
	0-5	**18.49**	**77.06**	**4.45**	**0.76**	**0.10**	**7.20**	**2.02**	**0.03**	**0,05**
BC1750	0-1	18.06	73.32	4.65	0.78	0.12	6.55	0.55	0.02	0.03
	1-2	17.85	77.38	4.75	0.82	0.11	7.75	0.36	0.01	0.02
	2-5	22.02	76.84	5.08	0.74	0.10	7.35	0.35	0.006	0.02
	0-5	**19.31**	**75.85**	**4.83**	**0.78**	**0.11**	**7.22**	**1.27**	**0.04**	**0.05**
BC2000	0-1	19.79	74.48	5.71	0.90	0.10	9.12	0.58	0.011	0.01
	1-2	19.86	74.95	5.18	0.80	0.09	8.92	0.14	0.001	0.004
	2-5	24.32	70.97	4.70	0.79	0.08	9.12	0.13	0.002	0.01
	0-5	**21.33**	**73.47**	**5.20**	**0.84**	**0.09**	**9.06**	**0.87**	**0.02**	**0.06**

Table S4.2. Results from the multi- (grain size) and univariate PERMANOVA three-way model for the sedimentary parameters. PERMANOVA test using water depths (WD: BC500, BC900, BC1200, BC1500, BC1750 and BC2000), sediment layers (SL: 0-1, 1-2 and 2-5 cm), both fixed and Replicates (nested in WD) as a factors. Sediment grain size, chloroplastic pigments equivalents (CPE), chlorophyll-*a* (Chl *a*), chlorophyll-*a*: phaeopigment ratio (Chl *a*: phaeo); organic carbon (OC), total nitrogen (TN) and molar carbon: nitrogen ratio (C: N). Bold values denote significant differences at $p<0.05$ and; bold italic values denote significant differences at $p<0.01$. Data was normalised (C: N and TN log (0.01+X) transformed) and Euclidean distance as measurement for resemblance.

Variable(s)	**Factors**	**df**	**SS**	**MS**	**Pseudo-F**	**P(perm)**	**Unique Perms**
Sediment grain size	Water Depth	5	67.022	13.404	4.071	***0.001***	9945
	Sediment Layer	2	12.063	60.315	16.38	***0.0001***	9945
	Replicate (WD)	10	32.92	3.292	89.403	***0.0001***	9929
	WD x SL	10	17.396	17.396	47.244	***0.0002***	9923
	Residual	20	73.644	0.368			
	Total	47	141				
CPE	Water Depth	5	23.748	47.495	10.164	***0.0018***	9959
	Sediment Layer	2	29.562	14.781	25.655	0.0992	9966
	Replicate (WD)	10	46.727	0.467	0.811	0.6337	9949
	WD x SL	10	43.017	0.4301	0.746	0.6744	9943
	Residual	20	11.523	0.5761			
	Total	47	47				
Chl*a*(µg/g)	Water Depth	5	23.483	46.967	12.191	***0.0026***	9956
	Sediment Layer	2	27.016	13.508	25.818	0.0918	9941
	Replicate (WD)	10	38.524	0.385	0.736	0.6827	9946
	WD x SL	10	67.165	0.672	12.837	0.3053	9944
	Residual	20	10.464	0.523			
	Total	47	47				
Chl*a*:phaeo	Water Depth	5	12.01	24.021	59.713	***0.0113***	9960
	Sediment Layer	2	0.144	0.0721	0.0595	0.9427	9950
	Replicate (WD)	10	40.227	0.402	0.332	0.9608	9929
	WD x SL	10	66.273	0.662	0.547	0.8359	9938
	Residual	20	24.226	12.113			
	Total	47	47				

Table S4.2. ***continued***

Variable(s)	**Factors**	**df**	**SS**	**MS**	**Pseudo-F**	**P(perm)**	**Perms**
OC (%)	Water Depth	5	12.01	24.021	59.713	***0.0113***	9960
	Replicate (WD)	10	40.227	0.402	0.332	0.9608	9929
	WD x SL	10	66.273	0.663	0.547	0.8359	9938
	Residual	20	24.226	12.113			
	Total	47	47				
TN (%)	Water Depth	5	24.029	48.058	53.799	***0.011***	9952
	Sediment Layer	2	22.632	11.316	32.171	0.076	9936
	Replicate (WD)	10	8.933	0.8933	25.396	**0.035**	9933
	WD x SL	10	43.242	0.43242	12.294	0.336	9997
	Residual	20	70.348	0.35174			
	Total	47	47				
C:N	Water Depth	5	17.296	34.591	19.239	0.1796	9963
	Sediment Layer	2	0.48393	0.24197	0.52367	0.6005	9950
	Replicate (WD)	10	17.979	17.979	38.911	***0.0037***	9945
	WD x SL	10	20.116	0.20116	0.43536	0.9189	9940
	Residual	20	92.412	0.46206			
	Total	47	47				

Table S4.3. Results from pair-wise univariate PERMANOVA analysis for differences in nematode density. PERMANOVA pair-wise comparisons within each of the WD and WD x SL levels as part of the repeated measures analysis, including Monte-Carlo permutation p values (P (MC). WD: water depth, SL: sediment layer (0-1, 1-2, 2-5 cm). Data was square root transformed; resemblance was calculated using Euclidean Distance. Bold values indicate p < 0.05 and bold italic values indicate p < 0.01.

Pair-wise test	Comparison	t	P(perm)	perms	P(MC)
Between WD					
	BC500, BC 900	4.4667	0.1038	10	**0.0218**
	BC500, BC1200	0.95656	0.6702	3	0.4415
	BC500, BC1500	53.571	0.1	10	**0.013**
	BC500, BC1750	20.888	0.1038	10	0.1257
	BC500, BC2000	24.158	0.1923	10	0.0951
	BC900, BC1200	32.043	0.0989	10	**0.0486**
	BC900, BC1500	0.10284	0.9001	10	0.9155
	BC900, BC1750	47.523	0.0959	10	**0.0089**
	BC900, BC 2000	0.075871	0.901	10	0.9456
	BC1200, BC1500	3.87	0.105	10	**0.0318**
	BC1200, BC1750	0.43654	0.6997	10	0.6835
	BC1200, BC2000	17.131	0.2871	10	0.1892
	BC1500, BC1750	66.756	0.1032	10	**0.0027**
	BC1500, BC2000	0.13024	1	10	0.9073
	BC1750, BC2000	21.028	0.1965	10	0.1019
Within SL	BC500, BC 900	0.5289	0.6925	10	0.6421
0-1	BC500, BC1200	14.692	0.331	3	0.2859
	BC500, BC1500	11.582	0.4031	10	0.3298
	BC500, BC1750	46.895	0.0978	10	**0.0151**
	BC500, BC 2000	0.6687	0.4979	10	0.5485
	BC900, BC1200	21.316	0.1007	10	0.1234
	BC900, BC1500	0.7417	0.6008	10	0.5006
	BC900, BC1750	51.857	0.1043	10	***0.0064***
	BC900, BC 2000	0.6060	0.6017	10	0.5857
	BC1200, BC1500	26.691	0.1984	10	0.0766
	BC1200, BC1750	19.232	0.1044	10	0.15
	BC1200, BC2000	12.529	0.3024	10	0.3102
	BC1500, BC1750	57.969	0.1006	10	***0.0056***
	BC1500, BC2000	0.2906	0.8046	10	0.7894
	BC1750, BC2000	22.337	0.0966	10	0.0882

Table S4.4. Results from pair-wise univariate PERMANOVA analyses for differences in nematode biomass. PERMANOVA pair-wise comparisons within each of the WD levels as part of the repeated measures analysis, including Monte-Carlo permutation p values (P (MC). Data was square root transformed; resemblance was calculated using Euclidean Distance. Bold values indicate $p < 0.05$ and bold italic values indicate $p < 0.01$.

Within WD	**t**	**P(perm)**	**perms**	**P(MC)**
BC500, BC900	64.048	0.0972	10	***0.0001***
BC500, BC1200	0.42817	1	10	0.706
BC500, BC1500	3.9416	0.1018	10	**0.0292**
BC500, BC1750	0.56144	0.6039	30	0.6315
BC500, BC2000	5.008	0.1018	10	**0.0175**
BC900, BC1200	9.8432	0.093	10	***0.0025***
BC900, BC1500	3.141	0.0974	10	0.0345
BC900, BC1750	2.5598	0.0143	60	**0.0558**
BC900, BC2000	0.37794	0.7025	10	0.73
BC1200, BC1500	3.5867	0.097	10	**0.0375**
BC1200, BC1750	0.68725	0.527	30	0.5438
BC1200, BC2000	4.6454	0.1	10	**0.0178**
BC1500, BC1750	1.213	0.341	60	0.2858
BC1500, BC2000	1.7201	0.1978	10	0.151
BC1750, BC2000	2.1274	0.0177	59	**0.0577**

Table S4.5. Results of PERMANOVA analyses for differences in univariate functional and structural index. PERMANOVA test for H´: Shannon index, *EG*(51): expected number of genera, J´: evenness and TD: trophic diversity; using water depth (WD), sediment layer (SL) and Replicates (Rep) as a factors. WD x SL: double interaction factor. *: $p < 0.05$; **: $p < 0.01$; df; degrees of freedom; SS: sum of squares; MS: mean squares; Pseudo-F: pseudo-F ratio; P(perm): permutation P-value; Perms: permutations; ECV: Estimated coefficient of variation.

Source	**df**	**SS**	**MS**	**Pseudo-F**	**P(perm)**	**Perms**	**ECV**
H´							
WD	5	79.514	15.903	34.037	*	9987	14127
SL	2	1380.6	690.29	43.927	**	9998	43.723
Rep (WD)	10	46.722	4.67	0.29732	n.s	9993	-3,681
WD x SL	10	114.42	11.442	0.72813	n.s	9994	-1.612
Residual	20	314.29	15.715				15.71
Total	47	1965					
***EG*(51)**							
WD	5	146.64	29.328	16.848	n.s	9958	1.4995
SL	2	2533.3	1266.8	30.553	**	9951	79.421
Rep (WD)	10	174.08	17.408	0,41984	n.s	9959	-8.0182
WD x SL	10	579.87	57.987	13.986	n.s	9947	6.2359
Residual	20	829.25	41.462				41.462
Total	47	4414.1					
J´							
WD	5	0.0321	0.00642	16.755	n.s	9998	0.000325
SL	2	0.277	0.1388	24.896	**	9989	0.008635
Rep (WD)	10	0.0650	0.003835	0.6878	n.s	9987	-0.00058
WD x SL	10	0.1115	0.006504	11.666	n.s	9989	0.003505
Residual	20	0.51096	0.00557				0.005578
Total	47						
TD							
WD	5	0.09642	0.01928	21.847	n.s	9957	0.001315
SL	2	0.53388	0.26694	32.268	**	9954	0.016766
Rep (WD)	10	0.08826	0.008268	1.067	n.s	9940	0.000184
WD x SL	10	0.08105	0.0081051	0.97974	n.s	9935	0.000632
Residual	20	0.1645	0.0082727				0.008272
Total	47	0.96726					

Table S4.6. Results from pair-wise multivariate PERMANOVA analyses for differences in nematode community. PERMANOVA pair-wise comparisons within each of the WD and WD x SL levels as part of the repeated measures analysis, including Monte-Carlo permutation p values (P (MC). WD: water depth, SL: sediment layer (0-1, 1-2, 2-5 cm).Data was standardized; resemblance was calculated using Bray-Curtis. Bold values indicate $p < 0.05$.

Pair-wise test	Comparisons	t	P(perm)	perms	P(MC)
Between WD					
	BC500, BC 900	1.1394	0.0986	10	0.1487
	BC500, BC1200	1.4277	0.3414	3	0.1404
	BC500, BC1500	2.2004	0.0961	10	0.0496
	BC500, BC1750	2.1743	0.1027	10	0.0465
	BC500, BC2000	2.111	0.1017	10	0.0242
	BC900, BC1200	1.212	0.0991	10	0.2103
	BC900, BC1500	1.8323	0.0973	10	0.0355
	BC900, BC1750	1.8401	0.0956	10	0.0502
	BC900, BC 2000	1.7901	0.1064	10	0.0148
	BC1200, BC1500	1.7737	0.0996	10	0.0854
	BC1200, BC1750	1.9995	0.1003	10	0.0418
	BC1200, BC2000	1.9372	0.0997	10	0.0347
	BC1500, BC1750	1.9355	0.0961	10	0.0501
	BC1500, BC2000	2.1769	0.1038	10	0.0252
	BC1750, BC2000	1.7878	0.1051	10	0.0498
Within SL	BC500, BC 900	1.2436	0.0946	10	0.1958
0-1	BC500, BC1200	1.2367	0.3316	3	0.2505
	BC500, BC1500	1.308	0.1022	10	0.1728
	BC500, BC1750	1.6047	0.1058	10	0.0918
	BC500, BC 2000	1.5983	0.0984	10	**0.059**
	BC900, BC1200	1.1424	0.4075	10	0.356
	BC900, BC1500	1.5273	0.1039	10	0.0749
	BC900, BC1750	1.6326	0.1043	10	**0.0458**
	BC900, BC 2000	1.677	0.1014	10	**0.0319**
	BC1200, BC1500	1.4666	0.0948	10	0.1256
	BC1200, BC1750	1.7276	0.1029	10	**0.0576**
	BC1200, BC2000	1.5435	0.0992	10	**0.0599**
	BC1500, BC1750	1.2861	0.0985	10	0.1382
	BC1500, BC2000	1.5288	0.0994	10	0.0692
	BC1750, BC2000	1.3406	0.0964	10	0.2149

Table S4.7. Distance-based linear models (DISTLM) – Marginal test and sequential test for best fitting model for nematode community and selected environmental variables. Selection procedure: step-wise, selection criterion: adjusted R^2. Prop.: % variation explained. Cuml.: cumulative variation explained. Chl *a*: Chlorophyll *a*; Chl*a*: phaeo: chlorophyll *a:* phaeopigments; CPE: chloroplastic pigment equivalents; % TN: total nitrogen content; OC:organic carbon content; C:N:carbone nitrogen ratio.

MARGINAL TEST

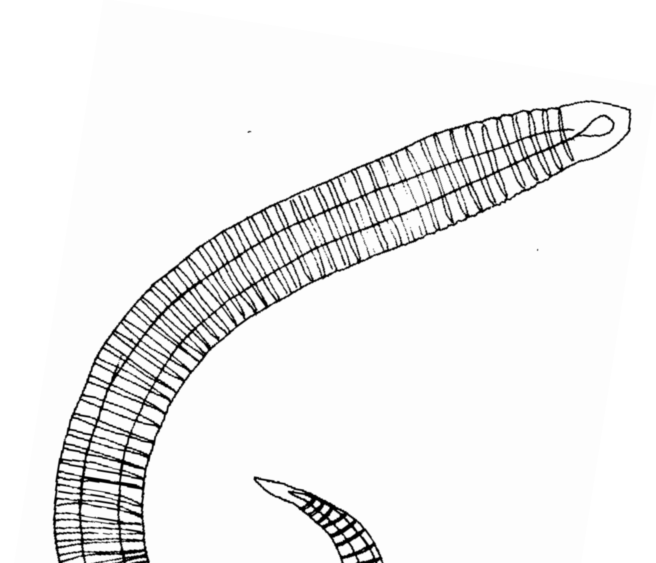
Variable	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**
Clay (%)	1880.6	2.2581	0.008	0.13889
Silt (%)	1317	1.5084	0.0801	9.7263E-2
Sand (%)	1465.7	1.6994	0.0369	0.10825
TN (%)	863.9	0.9541	0.4882	6.3802E-2
OC (%)	1081.2	1.2149	0.2226	7.9851E-2
C:N	1399.6	1.6139	0.0632	0.10336
Chl *a*	2418.2	3.044	0.0003	0.1786
CPE	2755	3.5762	0.0001	0.20347
Chl-a:phaeo	2319.5	2.894	0.0005	0.1713

SEQUENTIAL TEST

Variable	**Adj R^2**	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**	**Cumul.**	**res.df**
+CPE	0,19145	4296.2	4.5517	0.0003	0.24535	0,24535	14
+Clay (%)	0.23515	1607.1	1.8	0.0208	9.18E-02	0,33713	13
+Chl *a*: Phaeo	0.25178	1125.7	1.2888	0.1886	6.43E-02	0,40142	12
+% OC	0,26875	1091.5	1.2786	0.2152	6.23E-02	0.46375	11
+TN(%)	0.27378	912.3	1.0761	0.373	5.21E-02	0.51585	10
+C:N	0.30509	1176.7	1.4506	0.1594	6,72E-02	0.58306	9
+Chl *a*	0.30694	789.68	0.97347	0.477	4,51E-02	0.53796	10

5

Role of spatial scale and environmental drivers in shaping Nematode communities in the Canyon and the adjacent Open Slope



5.1 INTRODUCTION

Continental margins comprise the most geologically diverse components of the deep-ocean floor. They show high topographic heterogeneity (Levin and Dayton, 2009; Levin et al., 2010; Levin and Sibuet, 2012) and have been considered major reservoirs of marine biodiversity and productivity (Levin et al., 2001; Levin and Sibuet, 2012). Submarine canyons are among the major sources of this habitat heterogeneity (Vetter and Dayton, 1999; Levin et al., 2001). They provide a major transport pathway between the shelf and the deep-ocean environments by trapping, accumulating and funnelling sediments, organic matter and nutrients (Puig et al., 2014) together with pollutants and litter (Palanques et al., 2008; Tubau et al., 2015).

Active canyons are very unstable environments that are subject to constraints such as tidal currents, episodic slumps, sediment gravity flows, turbidity flows and periodic flushing (Canals et al., 2006; De Stigter et al., 2007; Palanques et al., 2006a; Puig et al., 2012). Furthermore, anthropogenic activities such as commercial fish trawling are prevalent around them, with its heavy doors causing significant furrows in the seafloor but also leading to the formation of turbid clouds of resuspended sediments, resulting in a persistent bottom intermediate nepheloid layers. These process affect the present seafloor community structure and biodiversity, especially at greater depths inside the canyon axis where transported material may accumulate (Palanques et al., 2006b, Martín et al., 2008, Martín et al., 2014b, Puig et al., 2012, Puig et al., 2015a, Puig et al., 2015b, Pusceddu et al., 2014; Wilson et al., 2015).

As a consequence, canyons axes constitute a complex of different habitats with specific hydrographic, sedimentological and geochemical characteristics (Flexas et al., 2008; López-Fernández et al., 2013; Amaro et al., 2016) that may influence the benthic community structure, diversity and abundance (Schlacher et al., 2010, Ingels et al., 2009; Garcia et al., 2007; De Leo et al., 2014; Romano et al., 2013b; Román et al., 2016). Previous studies reported that canyon inhabitants behave differently from the respective adjacent slopes located at similar depths, exhibiting differences in community composition and functioning (Vetter and Dayton, 1998; Duineveld et al., 2001; Garcia et al., 2007; Ingels et al., 2009; Gunton et al., 2015, Rosli et al., 2016; Román et al., 2016). Furthermore, habitat heterogeneity and organic matter accumulation support higher density and biomass in canyons compared to the adjacent open slope (eg. Curdia et al., 2004;

Ingels et al., 2009; De Leo et al., 2010; Huvenne et al., 2011; Romano et al., 2013a; Leduc et al., 2014). Nevertheless, other studies also have shown no "canyon effects" (e.g., Soltwedel et al., 2005; Bianchelli et al., 2010; Vetter et al., 2010) and even lower densities and biomasses in canyons compared to the slopes have been observed (Flach, 2002; Garcia et al., 2007; Maurer et al., 1994; Van Gaever et al., 2009). However, an overall generalisation for submarine canyons cannot be done because they are widely distributed along the world oceans (Harris and Whiteway, 2011), and each canyon possesses unique characteristics. In addition, benthic canyon communities, and in particular the meiobenthos, have only been investigated in a few canyon systems (De Leo et al., 2010; Amaro et al., 2016). Understanding the role of specific environmental conditions and/or anthropogenic threats to submarine canyons in determining biodiversity and ecosystem functioning at different spatial scales in submarine canyons is becoming urgent and will contribute to their effective management and conservation (Fernandez–Arcaya et al., 2017).

Nematodes are typically the dominant meiobenthic metazoans taxon in deep-sea environments (Giere, 2009) and the Blanes Canyon system is no an exception, with nematodes accounting for 90% of the total meiofauna (Romano et al., 2013 b; Román et al., 2016). Having fast turnover rates and likely suffering lower mortality rates after physical disturbances than larger benthic organisms (Schratzberger and Jennings, 2002), nematodes appear to be key taxa to investigate the structural and functional differences between an active canyon and its adjacent open slopes (Ingels et al., 2013).

The aim of the present study was therefore to compare the main descriptors (i.e., density, biomass, genus diversity, and community structure) of the nematode assemblages living in the middle and deeper regions the Blanes Canyon axis and at the same depths in the adjacent western slope, framed by the main sediment and environmental driving variables. Accordingly, the following questions were addressed: **1**) Are there similar nematode assemblage and standing stock bathymetric trends in Blanes Canyon and its adjacent slope? **2**) Which spatial scale from regional to local (water depth to sediment layer) is more important in shaping canyon and slope nematode assemblages and standing stocks? **3**) Which environmental variables are responsible for the spatial differences observed between canyon and slope?

5.2 MATERIAL AND METHODS

5.2.1 Sampling strategy

The samples studied in this study were those obtained during the DOS MARES cruise that took place in autumn 2012 (see **Chapter 2** for further sampling details in Table 2.1). In the present study, three stations were studied at ca. 1,500, 1,750 and 2,000 m water depth in the canyon (named BC1500, BC1750, and BC2000, respectively) at the western adjacent open slope (named OS1500, OS1750, and OS2000) (Fig. 5.1). Three multicorer deployments (replicates) were conducted at each sampling station. From each multicore deployment, one core was used for meiofaunal and three for sediment analyses, which were all carefully sub-sampled on board by means of a small PVC tube (36 mm of diameter, 5 cm sediment depth) taken from the center of the core to maintain a consistent sample surface area for all replicates. The three sediment layers from each subcore (see **Chapter 2**, section **2.2.1** for further details) were used separately in the analyses.

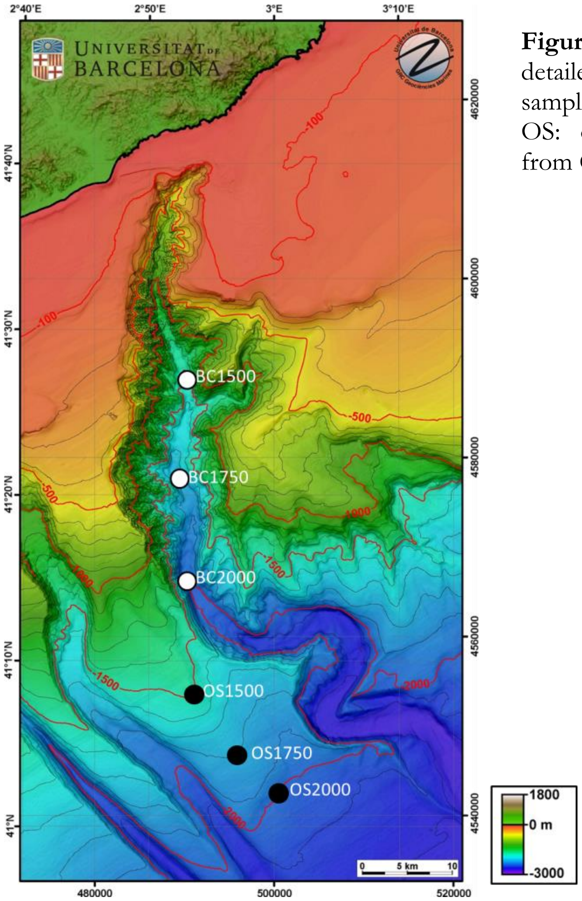


Figure 5.1. Overview (up right) and detailed map of the location of sampling stations. BC: Blanes Canyon, OS: open slope. Bathymetric data from Canals et al. (2004).

5.2.2 Environmental data

The methods to determine the quantity and quality of sediments variables are fully described in **Chapter 2** section **2.2.1**. Grain size (clay, silt and sand fractions), Chlorophyll a (Chl *a*, μg/g) and chlorophyll degradation products in the sediment, organic carbon concentration (OC%) and total nitrogen concentration (TN%) sediment variables were considered in the present studied. Additionally, Chloroplastic Pigments Equivalents (CPE: sum of Chl *a* and its degradation products as phaeopigments) were used to estimate surface-produced organic matter (OM). The ratio Chl *a*: phaeopigments (Chl *a*: phaeo) is used as a proxy to estimate the freshness of photosynthetically derived OM (Thiel, 1978).

5.2.3 Meiofauna and nematodes

The techniques to analyse meiofauna and for nematode processing are fully detailed in **Chapter 2 section 2.3.2**. Nematodes were identified under compound microscope (100 x magnification) to genus level using pictorial keys (Platt and Warwick, 1988) and the NeMys database (Guilini et al., 2016). Both density and biomass (see **Chapter 2** section **2.3.3**) were used in this Chapter.

5.2.4 Data analyses

Nematode community descriptors (density, biomass, structural diversity, and structure) were analysed by means of non-parametric permutational analyses of variance (PERMANOVA) using PRIMER v6 (Anderson et al., 2005, 2008). Structural diversity were measured as genus richness, Shannon index (Shannon, 1948), and expected number of genera (*EG*(51), using the function DIVERSE in PRIMER v6 (Clarke and Gorley, 2006).

The bathymetric trends of canyon and slope nematode assemblages and standing stocks (as sum of the 0-1, 1-2 and 2-5 cm sediment layers) were compared using a two-factor PERMANOVA design using Location (Lo: canyon and slope) and Water depth (WD: 1,500, 1,750 and 2,000 m) as fixed factors. Similarity matrices for the univariate variables (i.e., density, biomass and structural diversity) were based on Euclidean similarity. Community structure matrices were built using Bray-Curtis similarity (Clarke and Gorley, 2006) based on the genus relative abundances. To determine spatial patterns between samples based on the community structure, a principal coordinate analysis (PCO) plot computed. When the number of unique permutations was lower than one hundred, the Monte Carlo (P(MC)) *p* values were used.

The relevance of spatial scales (i.e., regional *vs.* local) in shaping canyon and slope assemblages and standing stocks was assessed by measuring the magnitude of the spatial variation through the Estimates of Components of Variation (ECV). The ECV was calculated as the percentage of total variation obtained by: a fully crossed 3-factor PERMANOVA design with Water depth (WD), and sediment layer (SL: 0-1, 1-2, 2-5 cm) as fixed factors, and Core (Co) as random factor nested in WD, performed for canyon and slope independently. The design was used to analyse the effects of WD and SL within the canyon and slope independently. Variance components were set to zero when negative values were encountered, under the postulation that they were sampled underestimates of small zero variances (Benedetti-Cecchi, 2001). The standing stocks (total density and total biomass) were normalized and Euclidean distance was used to construct the resemblance matrix. The same data treatment was applied to the set of structural diversity descriptors (i.e. genus richness, *EG*(51) and Shannon index). Nematode relative abundance data was standardised for sample size and Bray-Curtis was used as a similarity measure. Following the PERMANOVA analyses, SIMPER routines on relative abundances were performed to identify which genera were responsible for regional and local differences (with a cut-off of 90% for low contributions).

Since PERMANOVA is not able to distinguish between factor effects or data dispersion, homogeneity of multivariate dispersion was tested using PERMDISP, with distances calculated among centroids. PERMDISP analyses were performed within Lo, WD and SL and nematode communities were not significant indicating that the effects observed in the PERMANOVA analyses were due to location differences (factor effects). Nematode density and biomass were significant for the univariate variables and were thus square-root transformed prior the analyses.

To assess which environmental factors were responsible for the observed local and regional differences between canyon and slope, partial Spearman Rank correlations between the selected environmental variables (clay, silt, sand, OC, TN, Chl *a*, CPE, and Chl *a*: phaeo) and univariate descriptors (density, biomass, genus richness, *EG*(51), and Shannon index) were performed at regional and local scales by means of the XLSTAT (Addinsoft) software. Furthermore, RELATE and DISTLM (distance-based linear model) routines based on the normalized environmental data were performed (Anderson et al., 2008; Clarke and Gorley, 2006) to analyse and model the relationship between community structure and the environmental variables both for canyon and slope separately and for regional and local scales.

The DISTLM was built using a "step-wise" selection procedure and adjusted R^2 was chosen as the selection criterion (Anderson et al., 2008). The results were visualized using dbRDA (distance-based redundancy analysis) plots.

Between location differences in selected sediment environmental variables were assessed using the same PERMANOVA design as described for nematode assemblages (over the terms Lo and WD) using the three-factor design (WD, SL and Co as a factors) to assess differences between different spatial scales within each location. Sediment variable matrices were based on Euclidean similarity matrices. Additionally, Principal Component Analyses (PCA) based on environmental variables were performed to assess differences in spatial distribution between canyon and slope samples (as sum of the sediment layers 0-1, 1-2 and 2-5 cm sediment layers). Significant differences were considered when p value < 0.05.

5.3 RESULTS

5.3.1 Canyon *vs.* Open slope

Nematode density was significantly different for the interaction term Lo x WD (Table 5.1, Fig. 5.2). While a significant decrease with the increasing water depth was observed on the slope, no bathymetric gradient was detected in the canyon, where the highest densities occurred at 1750 m depth (Fig. 5.2, Table S1-A). Pair wise comparisons between canyon and slope for the same water depth, only showed significant differences at 1,750 m depth (Table S1-A, Fig. 5.2).

Similar to densities, biomass decreased with the increasing water depth in the slope, while no bathymetric trend was observed in the canyon (Fig. 5.2). Nevertheless, significant differences were observed between Lo, where canyon stations exhibited higher biomasses when compared to slope stations (Table 5.1, Fig. 5.2).

Table 5.1. Results of two-way PERMANOVA analyses for differences in nematode descriptors using Location (Lo) and water depth (WD) as factors. Bold: $p < 0.05$; bold italic: $p < 0.01$.

Source	df	SS	MS	Pseudo-F	P(perms)	Perms
Density						
Lo	1	482.03	482.03	35.823	***0.0001***	9827
WD	2	153.54	76.77	57.053	**0.0194**	9954
Lo x WD	2	224.33	112.17	83.359	***0.0073***	9959
Res	12	161.47	13.456			
Total	17	1021.4				
Biomass						
Lo	1	19.757	19.757	11.448	***0.0085***	9847
WD	2	45.913	22.956	13.302	0.3055	9949
Lo x WD	2	44.388	22.194	1.286	0.334	9957
Res	12	15.533	17.258			
Total	17	41.973				
Genus richness						
Lo	1	206.72	206.72	27.36	***0.0003***	9730
WD	2	103.44	51.722	68.456	**0.0113**	9943
Lo x WD	2	290.11	145.06	19.199	***0.0004***	9957
Res	12	90.667	75.556			
Total	17	690.94				
***EG*(51)**						
Lo	1	22.809	22.809	69.543	**0.022**	9989
WD	2	16.24	81.199	24.757	0.12	9979
Lo x WD	2	25.694	12.847	39.169	0.063	9958
Res	12	39.359	32.799			
Total	17	104.1				
Shannon index						
Lo	1	0.27411	0.27411	26.81	***0.0009***	9834
WD	2	0.13399	6.70E-02	65.523	**0.0121**	9945
Lo x WD	2	1.37E-02	6.84E-03	0.66856	0.5293	9949
Res	12	0.12269	1.02E-02			
Total	17	0.54446				
Community structure						
Lo	1	2950.8	2950.8	44.459	***0.0006***	9942
WD	2	2637.1	1318.6	19.866	***0.0062***	9910
Lo x WD	2	3756.2	1878.1	28.296	***0.0002***	9911
Res	12	7964.7	663.72			
Total	17	17309				

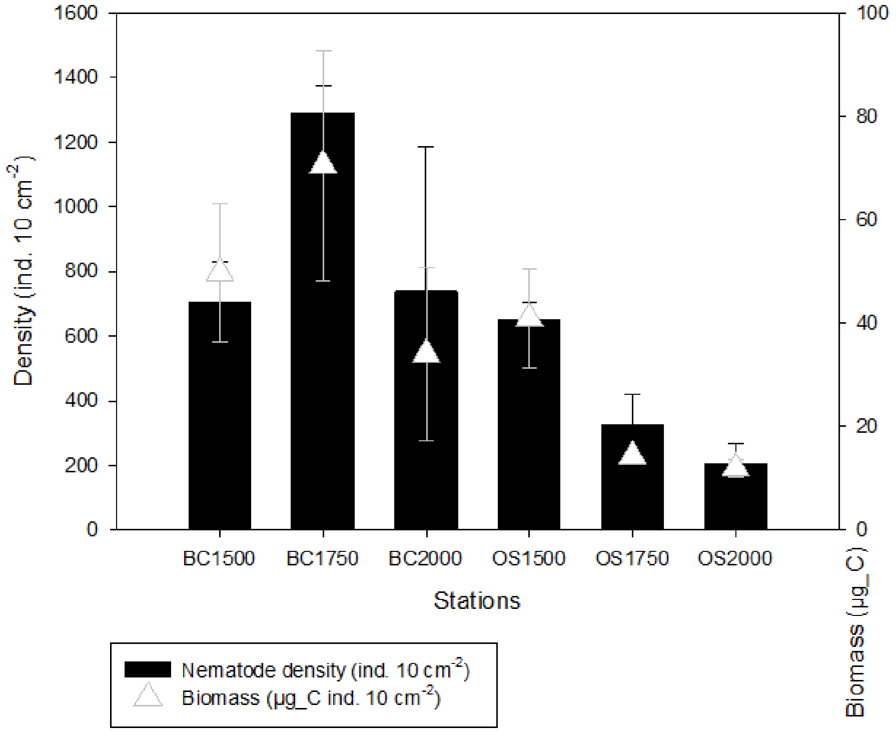


Figure 5.2. Average nematode densities (ind ·10cm^2) in the studied stations. Superimposed are: average nematode biomass (µgC·10 cm^{-2}). Scales were made uniform for better comparison. Error bars are standard deviations.

One hundred and thirteen genera were identified among the 3,479 sorted specimens (see **Appendix 4** for further details). In general, genus richness was significantly higher in the canyon (93 genera) than on the slope (80 genera). Moreover, significant differences were found for the interaction term Lo x WD (Table 5.1). Similar to density and biomass, genus richness decreased with increasing water depth in the slope, but no trend was observed for the canyon sites (Table 5.2). Pair wise comparisons only revealed significant differences at 2,000 m between Lo, with 68 and 40 genera in the canyon and in the slope, respectively (Table S1-B). Additionally, 60 genera were shared between locations, while 33 were exclusive from the canyon and 16 from the slope. Furthermore, 38 genera were shared among canyon stations over all WD *vs.* 30 among slope ones.

The expected number of genera (*EG*(51)) only differed significantly between Lo while no significant bathymetric trend was observed (Table 5.1). Shannon index differed significantly for the single factors Lo and WD (Table 5.1). No significant differences were observed for Lo x WD. Shannon index was significantly higher in the canyon than on the slope (Tables 5.1, 5.2) and generally decreased with the increasing water depth along the canyon, while no gradient was observed in the slope (Table 5.2, Table S1-C).

Table 5.2. Structural diversity indices at each study station.

Station	Genus richness		E(51)		Shannon Index	
BC1500	65	± 4.0	25.03	± 1.9	3.24	± 0.0
BC1750	64	± 1.0	25.95	± 1.8	3.20	± 0.1
BC2000	68	± 2.1	25.51	± 0.9	3.08	± 0.0
OS1500	60	± 3.0	22.18	± 3.0	2.96	± 0.1
OS1750	59	± 3.5	23.83	± 1.5	3.03	± 0.1
OS2000	40	± 1.5	19.85	± 0.7	2.75	± 0.1

Sabatieria dominated in the slope followed by *Acantholaimus* and *Halalaimus,* and by *Tricoma* specifically at OS1500 (Table 5.3). In the canyon, they were also dominant, but differed in relative abundance among water depths. Despite occurring both in canyon and slope stations, *Sphaerolaimus* and *Molgolaimus* were more abundant at BC1500 (8%) and BC1750 (9.6%), respectively (Table 5.3). On the slope, however, *Molgolaimus* showed abundances lower than 2% and was absent at OS2000 (Table 4). There were significant differences in community structure for the interaction term Lo x WD (PERMANOVA, $p < 0.05$, Table 5.1). Overall, slope communities were more similar, while there was a clear separation among water depths in the canyon (Fig. 5.3). Moreover, the community composition at BC2000 was similar to some shallower slope, being dominated by *Sabatieria, Acantholaimus* and *Tricoma* (Table 5.3). Pair wise comparisons confirmed this trend, with significant differences only being observed between canyon and slope at 1,500 and 1,750 m depth (Table S1-D).

Table 5.3. Relative abundance of the dominant (> 2%) nematode genera per station (0-5 cm, all replicates includes).

BC1500	%	**BC1750**	%	**BC2000**	%	**OS1500**	%	**OS1750**	%	**OS2000**	%
Sabatieria	14.6	*Molgolaimus*	9.6	*Sabatieria*	20.2	*Sabatieria*	22.2	*Sabatieria*	18.5	*Sabatieria*	22.9
Halalaimus	8.6	*Halalaimus*	9.1	*Acantholaimus*	9.7	*Acantholaimus*	10.4	*Acantholaimus*	9.7	*Acantholaimus*	8.3
Sphaerolaimus	8.0	*Acantholaimus*	8.4	*Tricoma*	6.5	*Tricoma*	6.7	*Halalaimus*	6.9	*Halalaimus*	7.8
Pomponema	4.6	*Sabatieria*	7.6	*Halalaimus*	6.4	*Halalaimus*	5.7	*Monhystrella*	5.2	*Diplopeltula*	5.4
Diplopeltula	4.4	*Tricoma*	6.6	*Molgolaimus*	4.2	*Amphimonhystrella*	3.9	*Chromadorella*	4.3	*Metasphaerolaimus*	4.6
Oxystomina	4.1	*Amphimonhystrella*	4.5	*Chromadorella*	3.5	*Daptonema*	3.8	*Tricoma*	4.2	*Sphaerolaimus*	4.0
Acantholaimus	3.9	*Metasphaerolaimus*	4.0	*Metasphaerolaimus*	3.0	*Pselionema*	3.1	*Syringolaimus*	4.0	*Pselionema*	3.3
Cervonema	3.6	*Sphaerolaimus*	3.7	*Diplopeltula*	2.7	*Monhystrella*	2.6	*Desmoscolex*	2.8	*Amphimonhystrella*	3.0
Daptonema	3.3	*Cervonema*	3.6	*Oxystomina*	2.6	*Chromadorella*	2.0	*Amphimonhystrella*	2.7	*Syringolaimus*	2.9
Tricoma	3.0	*Pselionema*	2.9	*Monhystrella*	2.1			*Pselionema*	2.6	*Monhystrella*	2.8
Disconema	3.0	*Marylynnia*	2.8	*Syringolaimus*	2.1			*Sphaerolaimus*	2.3	*Vasostoma*	2.7
Actinonema	2.9	*Daptonema*	2.4	*Amphimonhystrella*	2.0					*Oxystomina*	2.5
Halichoanolaimus	2.9	*Monhystrella*	2.1							*Symplocostoma*	2.0
Pselionema	2.3									*Thalassomonhystera*	2.0
Molgolaimus	2.2										
Marylynnia	2.0										

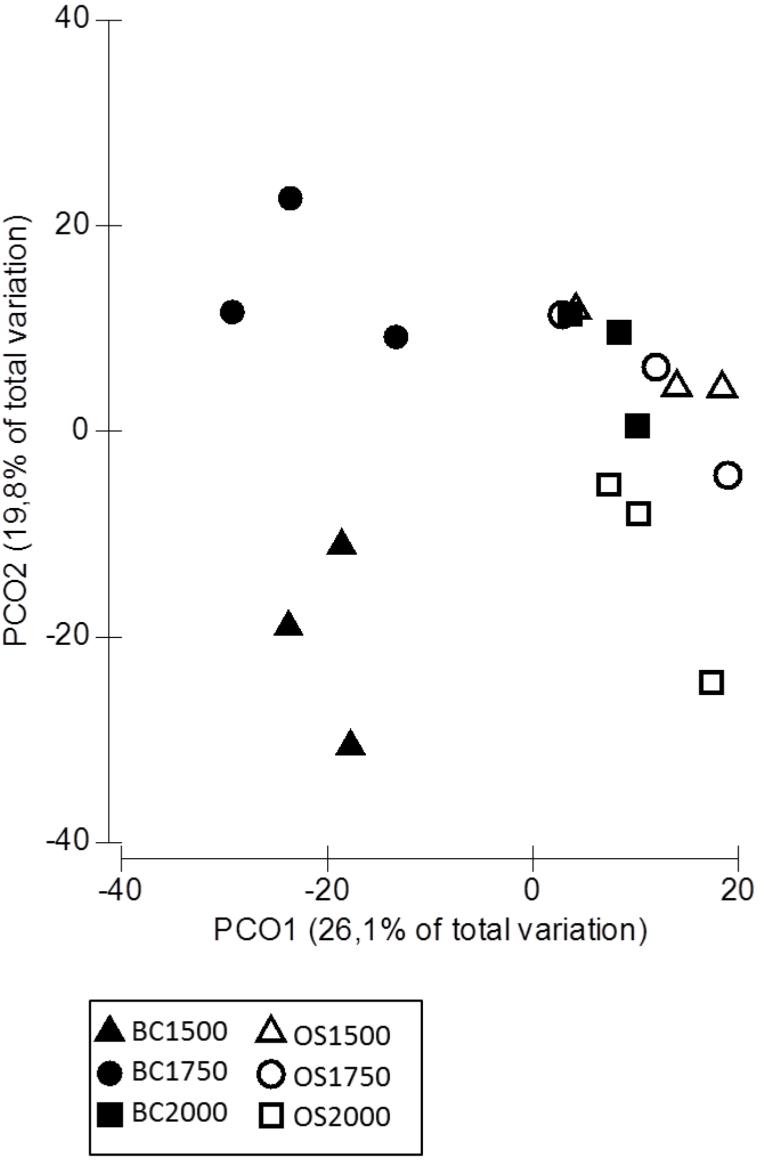


Figure 5.3. Principal coordinates analysis (PCO) plot on standardized nematode genera relative abundance data and Bray-Curtis similarity measurement at the study stations. BC: Blanes canyon; OS: open slope. Fill symbols represent canyon samples.

5.3.2 Spatial scales variability

The analyses of Estimated Components of Variation (ECV) showed that the local scale (i.e. sediment layers) was the most important and a significant factor affecting the standing stocks (total density, total biomass) in the canyon (Fig. 5.4A). Water depth (WD) and the WD x SL interaction term also contributed significantly to the variability observed. Moreover, there were significant differences between all sediment layers at BC1750 and also between the 0-1 and 1-2 cm ones at BC1500 (Fig. 5.5). At the slope, there was no significant difference in standing stocks between SL at each station (Figs. 5.4A, 5.5).

Sediment layer was also the most explanatory factor for structural diversity and community structure, and both in the canyon and on the slope, but the ECV percentages were much higher in the canyon (PERMANOVA, $p < 0.05$; Fig. 5.4B-C). WD also contributed significantly to the variability observed for all univariate variables (PERMANOVA, $p < 0.05$; Fig. 5.4B-C) on the slope, but only for community structure in the canyon (Fig. 4C).

Furthermore, SIMPER analysis showed that the dissimilarities between WD were higher in the canyon than on the slope (Table 5.4). *Sabatieria* most contributed to explain the dissimilarities in the canyon, followed by *Sphaerolaimus* and *Molgolaimus* (more abundant at BC1500 and BC1750, respectively) (Table 5.4).

The significant and high SL variability found both in the canyon and on the slope were caused by the greater dissimilarity between surface (0-1 cm) and deeper (2-5 cm) SL (Table 5.5, Fig. 5.6). *Sabatieria* was also the main contributor to community dissimilarity between SL, being relatively more abundant in the deepest one. *Acantholaimus* was the second most important contributor, more abundant in the surface SL, except when 1-2 *vs.* 2-5 cm SL were compared in the canyon, in which *Sphaerolaimus* was also abundant. The canyon *vs.* slope dissimilarity for each SL was 55.5% on average. *Molgolaimus*, *Acantholaimus* and *Tricoma* were the main responsible for the dissimilarities between 0-1 cm layers, while those between canyon and slope for the 1-2 and 2-5 cm layers were respectively caused by *Sabatieria*, *Sphaerolaimus* (higher in the canyon, 5.2 %), and by *Hopperia* and *Syringolaimus* (higher on the slope, 5.6% and 5.3%, respectively) (Table 5.5).

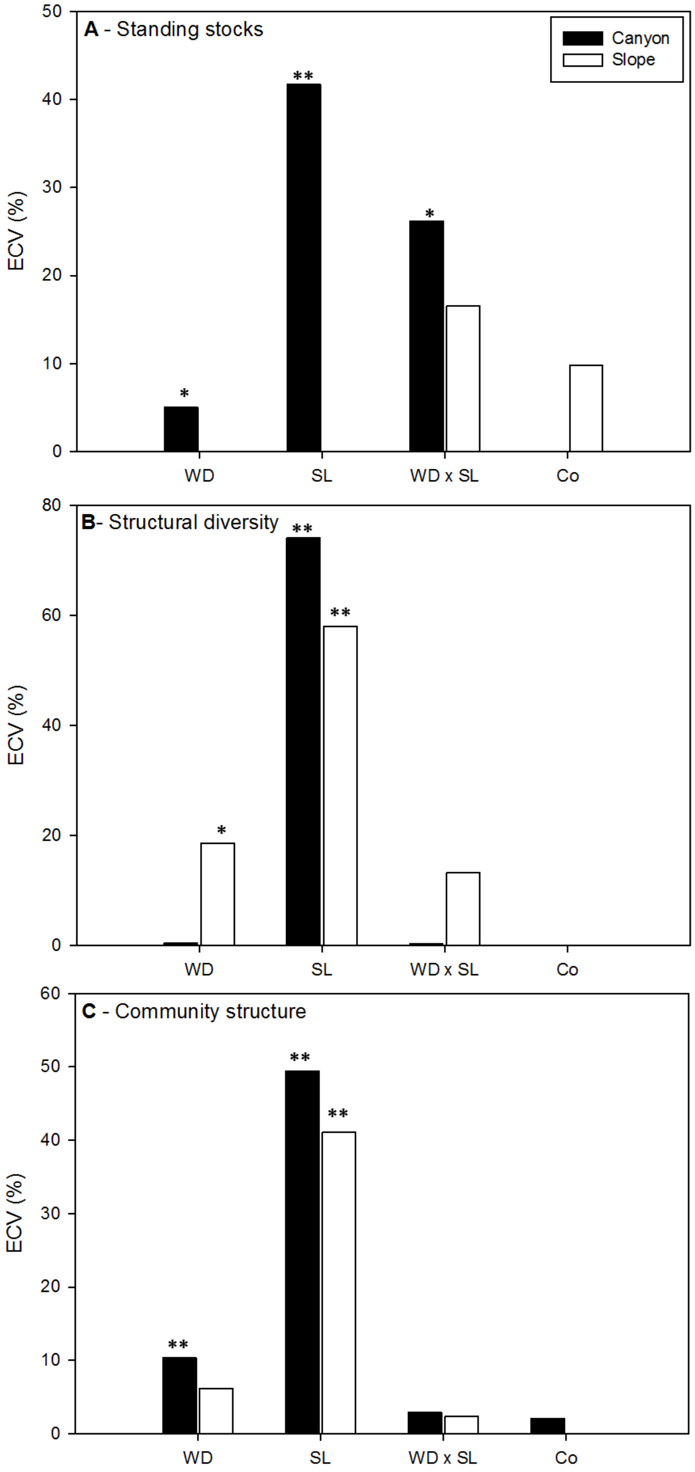


Figure 5.4. Percentages of Estimates Components of Variation from the PERMANOVA test for different biodiversity descriptors sets. BC: Blanes canyon and OS: open slope; WD: water depth; SL: sediment layer; Co: core. * indicates significance of the PERMANOVA test at $p < 0.05$; ** indicates significance at $p < 0.01$.

Table 5.4. SIMPER analyses results showing nematode genera accounting for community dissimilarity at **regional scale** (i.e., WD) in the canyon and slope locations (cut-off applied at 90% contribution). > and < symbols indicate the direction of the higher Average abundance of each genus between water depth comparisons.

Dissimilarity within locations (4> %)

1500 vs. 1750						**1500 vs. 2000**						**1750 vs. 2000**					
Canyon	**49.04**		**Slope**	**30.8**		**Canyon**	**52.29**		**Slope**	**39.06**		**Canyon**	**47.98**		**Slope**	**40.05**	
Molgolaimus	7.83	<	*Sabatieria*	9.08	>	*Sabatieria*	7.09	<	*Sabatieria*	8.59	<	*Sabatieria*	14.3	<	*Sabatieria*	9.76	<
Sabatieria	6.42	>	*Chromadorella*	5.49	<	*Sphaerolaimus*	6.35	>	*Diplopeltula*	6.58	<	*Molgolaimus*	5.21	>	*Diplopeltula*	4.77	<
Tricoma	5.41	<	*Syringolaimus*	5.2	<	*Acantholaimus*	5.28	<	*Tricoma*	5.18	>	*Tricoma*	4.0	>	*Metasphaerolaimus*	4.3	<
Acantholaimus	5.19	<	*Tricoma*	5.02	>	*Tricoma*	4.06	<	*Acantholaimus*	4.59	>				*Acantholaimus*	4.12	>
									Desmoscolex	4.04	>				*Tricoma*	4.07	>
															Chromadorella	4.01	>

Dissimilarity between locations (4> %)

BC1500 vs. OS1500	**52.1**		**BC1750 vs. OS1750**	**48.09**		**BC2000 vs. OS2000**	**45.01**	
Sabatieria	9.44	<	*Sabatieria*	14	<	*Sabatieria*	7.6	<
Acantholaimus	7.52	<	*Molgolaimus*	8.19	>	*Molgolaimus*	5.3	>
Sphaerolaimus	5.17	>	*Syringolaimus*	5.23	<	*Metasphaerolaimus*	4.49	<
			Tricoma	4.04	>	*Tricoma*	4.4	>
						Sphaerolaimus	4.13	<

Table 5.5 SIMPER analyses results showing nematode genera accounting for community dissimilarity at **local scale** (i.e., in the canyon and slope locations (cut-off applied at 90% contribution). **>** and **<** symbols indicate the direction of the higher Average abundance of each genus between sediment layer comparisons.

Dissimilarity within locations (4 > %)

0-1 vs. 1-2 cm						0-1 vs. 2-5 cm						1-2 vs. 2-5 cm					
Canyon	**69.33**		**Slope**	**66.75**		**Canyon**	**86.05**		**Slope**	**78.14**		**Canyon**	**57.78**		**Slope**	**54.96**	
Sabatieria	18.04	<	*Sabatieria*	27.84	<	*Sabatieria*	28.01	<	*Sabatieria*	18.71	<	*Sabatieria*	21.14	<	*Sabatieria*	15.42	>
Acantholaimus	6.67	>	*Acantholaimus*	6.1	>	*Acantholaimus*	6.34	>	*Acantholaimus*	8.27	>	*Sphaerolaimus*	5.29	>	*Acantholaimus*	5.83	>
Molgolaimus	4.9	>	*Tricoma*	5.13	>	*Halalaimus*	4.99	>	*Tricoma*	4.88	>	*Cervonema*	4.65	>	*Hopperia*	5.55	<
Tricoma	4.52	>				*Molgolaimus*	4.21	>	*Halalaimus*	4.37	>	*Halalaimus*	4.31	>	*Halalaimus*	4.72	>
Sphaerolaimus	4.16	<				*Tricoma*	4.06	>	*Hopperia*	4.3	<				*Syringolaimus*	4.68	<
															Chromadorella	4.42	>
															Monhystrella	4.34	<
															Oxystomina	4.28	<

Dissimilarity between locations (4 > %)

0-1 vs. 0-1 cm	**53.67**		**1-2 vs. 1-2 cm**	**57.09**		**2-5 vs. 2-5 cm**	**55.91**	
Molgolaimus	6.38	>	Sabatieria	20.1	<	*Sabatieria*	14.5	>
Acantholaimus	5.83	<	*Sphaerolaimus*	5.21	>	*Hopperia*	5.61	<
Tricoma	5.32	>	*Acantholaimus*	5.15	<	*Syringolaimus*	5.31	<
Sabatieria	5.3	<	*Cervonema*	4.82	>	*Amphimonhystrella*	4.57	>
						Monhystrella	4.48	<
						Paramphimonhystrella	4.41	>
						Diplopeltula	4.4	<

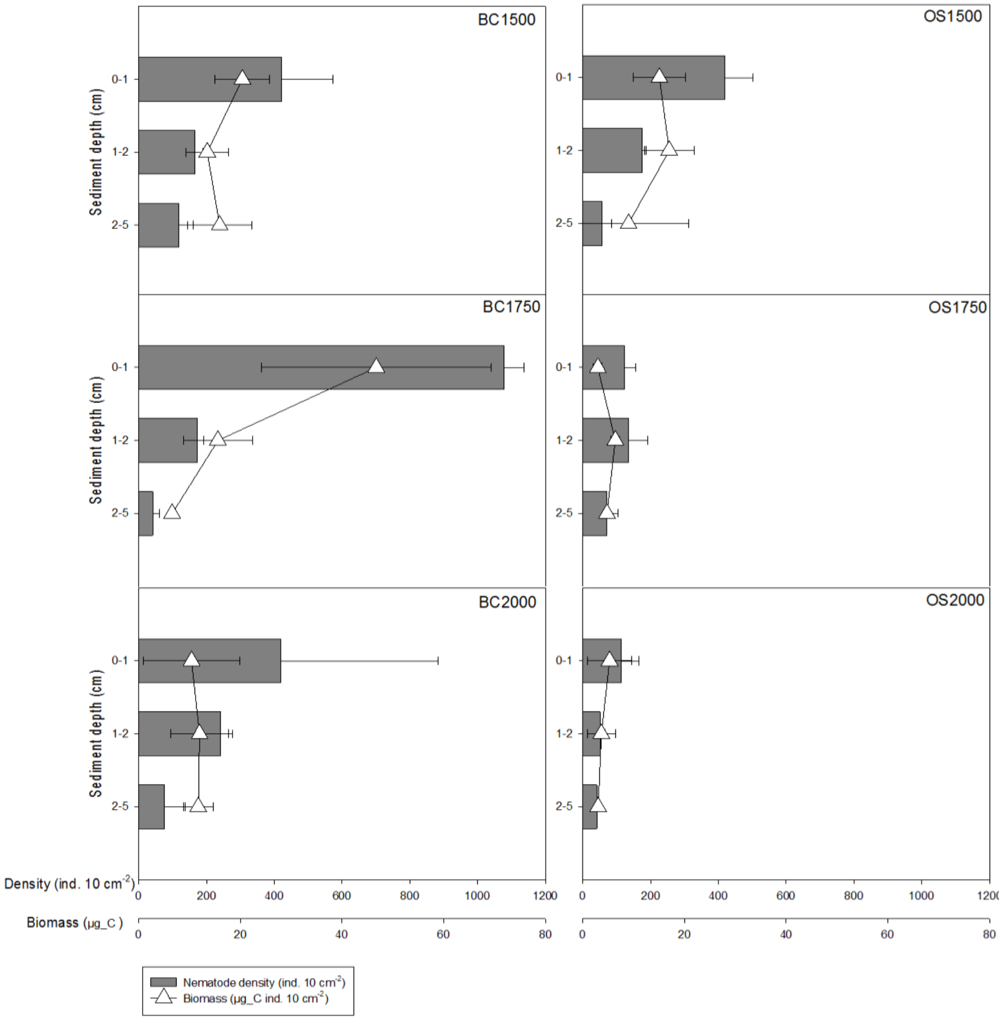


Figure 5.5. Average nematode densities (ind·10cm2) along the sediment profile in the studied stations. Superimposed are: average nematode biomass (µgC 10 cm-2). Scales were made uniform for better comparison. Error bars denote standard deviations.

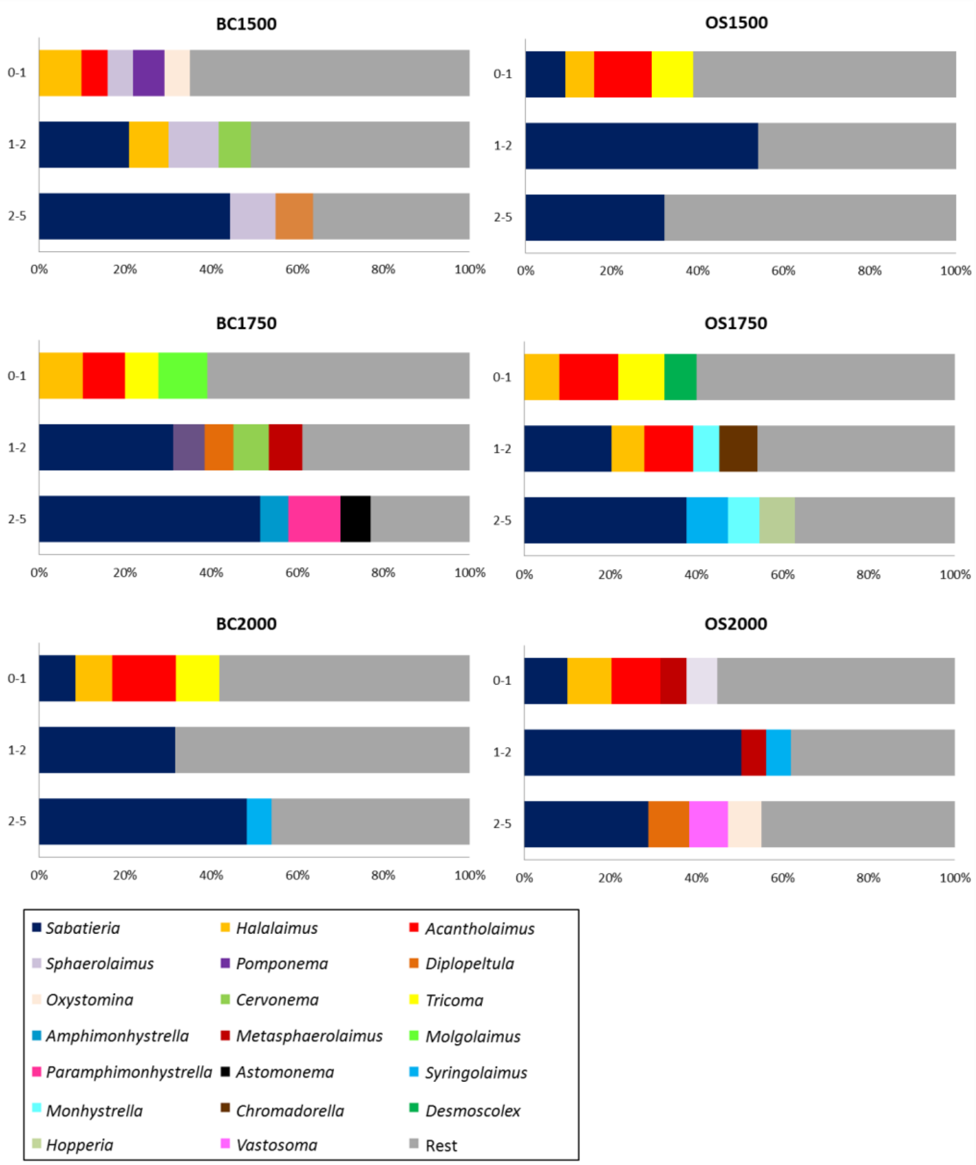


Figure 5.6. Vertical profile of nematode relative abundances for each station. Only genera with abundance > 6% in one of the layers were included, all others were grouped as "rest".

5.3.3 Environmental variables

Canyon and slope sediments were dominated by silt (70.1-77%), followed by clay (18.4-24.3%) and sand (4.4- 5.8%) (Fig. 5.7). Sediments were characterized by high silt and clay contents in the canyon and in the slope, respectively (Table 5.6, Fig. 5.7). There were significant differences for clay and silt between Lo at all WD (pair wise comparisons, $p < 0.05$, see Fig. 5.7). In the canyon, clay and silt contents increased and decreased, respectively, with water depth (pair-wise comparisons, $p < 0.05$, see Fig. 5.7), while they did not vary significantly in the slope (pair-wise comparison, $p > 0.05$, see Fig. 5.7). All show sediments that became finer along the vertical sediment profile, but this was particularly evident at the slope (PERMANOVA, $p < 0.05$, Fig. 5.8).

Organic carbon concentration (OC) was significantly higher in the canyon (0.76-0.83%) compared to the slope (0.61-0.62%) (Table 5.6, Fig. 5.7), did not show significant bathymetric trends and decreased along the sediment profile only on the slope (PERMANOVA, $p < 0.05$, Fig. 5.8).

Table 5.6. Results from univariate PERMANOVA two-way analyses for differences in sedimentary variables. Test for locations (Lo: Canyon and Slope); water depth (WD) and interaction term. Clay, Silt, Sand: volume percent clay, silt, sand content; TN: total nitrogen concentration; OC: organic carbon concentration; Chl-*a*: chlorophyll *a*; CPE: chloroplastic pigment equivalents; Chl-*a*:phaeo: chlorophyll a: phaeopigments ratio. Data was normalised; resemblance was calculated using Euclidean Distance. Bold values indicate significant differences at p<0.05, bold italic values indicate significant differences at p<0.01.

Source	df	Clay	Silt	Sand	OC	TN	Chl *a*	CPE	Chl *a*: phaeo
Lo	1	***0.0001***	***0.0002***	0.2679	***0.0003***	***0.0003***	***0.0003***	***0.0008***	0.3966
WD	2	0.1173	0.1672	0.5685	0.2882	0.1189	0.1221	**0.0269**	0.5843
Lo x WD	2	***0.0066***	***0.0001***	0.1425	0.5809	***0.0058***	**0.0383**	0.2567	0.3217
Res	12								
Total	17								

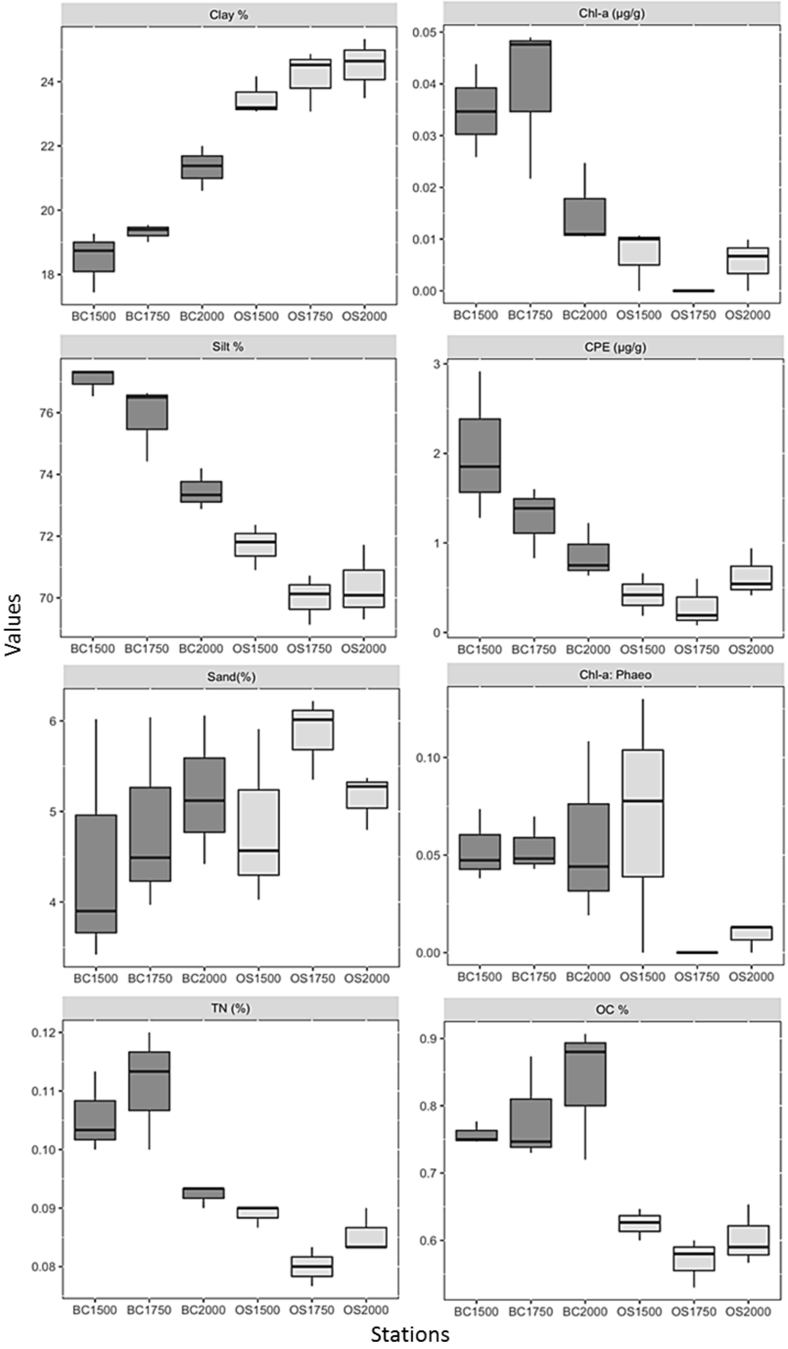


Figure 5.7. Environmental variables used in this study for canyon (BC) and slope (OS) stations: TN (total nitrogen), Chl *a* (Chlorophyll *a*), CPE (Chloroplastic Pigment Equivalents), Chl *a*: phaeo (Chlorophyll a: phaeopigments) and OC (organic carbon). Black lines represent the median and lower box indicates the first quartile and upper box the third quartile. Upper line on the boxes shows the maximum value and lower line the minimum value.

Total nitrogen concentration (TN) was significantly higher in the canyon (0.9-0.11%) compared to the slope (0.9%) (Table 5.6, Fig. 5.7), significantly differed between Lo at 1,500 and 1,750 m depth (pair wise comparisons, $p < 0.05$, see Fig. 5.7), and did not show clear gradients along the sediment profile both in the canyon and on the slope. However, there were significant differences between SL at the slope (PERMANOVA, $p < 0.05$, Fig. 5.8).

Chlorophyll *a* (Chl *a*) was significantly higher in the canyon (0.02-0.04 μg/g) than on the slope (0.0-0.007 μg/g) (Table 5.6, Fig. 5.7), differed significantly between Lo at 1,500 and 1,750 m depth, and did not show a clear bathymetric trend for both in the canyon and on the slope (pair wise comparisons, $p < 0.05$, see Fig. 5.7). However, at BC1500 sediment, Chl *a* was significantly higher than at BC2000 (pair-wise comparisons, $p < 0.05$, see Fig.5.7). Also, Chl *a* decreased with increasing sediment depth at BC1750 and BC2000, but there were no overall significant differences for SL in the canyon, nor in the slope (Fig. 5.8).

Chlorophyll pigments equivalents (CPE) was higher in the canyon (0.8-2.0 μg/g) than in the slope (0.2-0.6 μg/g) (Table 5.6, Fig. 5.7). On the slope, there was no clear bathymetric trend, while a decrease with increasing water depth was observed in the canyon. There were no significant differences between SL for both locations, although a decrease along the vertical sediment profile at 1,750 and 2,000 m depth both in the canyon and in the slope was observed (Fig. 5.8).

Chlorophyll *a*: phaeopigments ratio (Chl *a*: phaeo) did not show significant differences between canyon (0.01-0.07) and slope (0.00-0.04) (Table 5.6, Fig. 5.7), neither between SL within canyon and slope (PERMANOVA, $p > 0.05$, Fig. S2).

Canyon and slope stations appeared clearly separated in the PCA plot (Fig.5.9), with the latter being less variable in terms of environmental variables than the former. Sediment characteristics at BC2000 showed greater resemblance to slope samples compared to the other canyon stations in having a greater clay content (Fig. 5.7). The first two PC axes explained 76.3% of the variation (Fig. 5.9). The main contributors were clay (0.426), silt (−0.426), Chl *a* (−0.404) and TN (-0.403) for PC1, and sand (−0.858) and CPE (−0.344) for PC2 (numbers in parenthesis represent eigenvector).

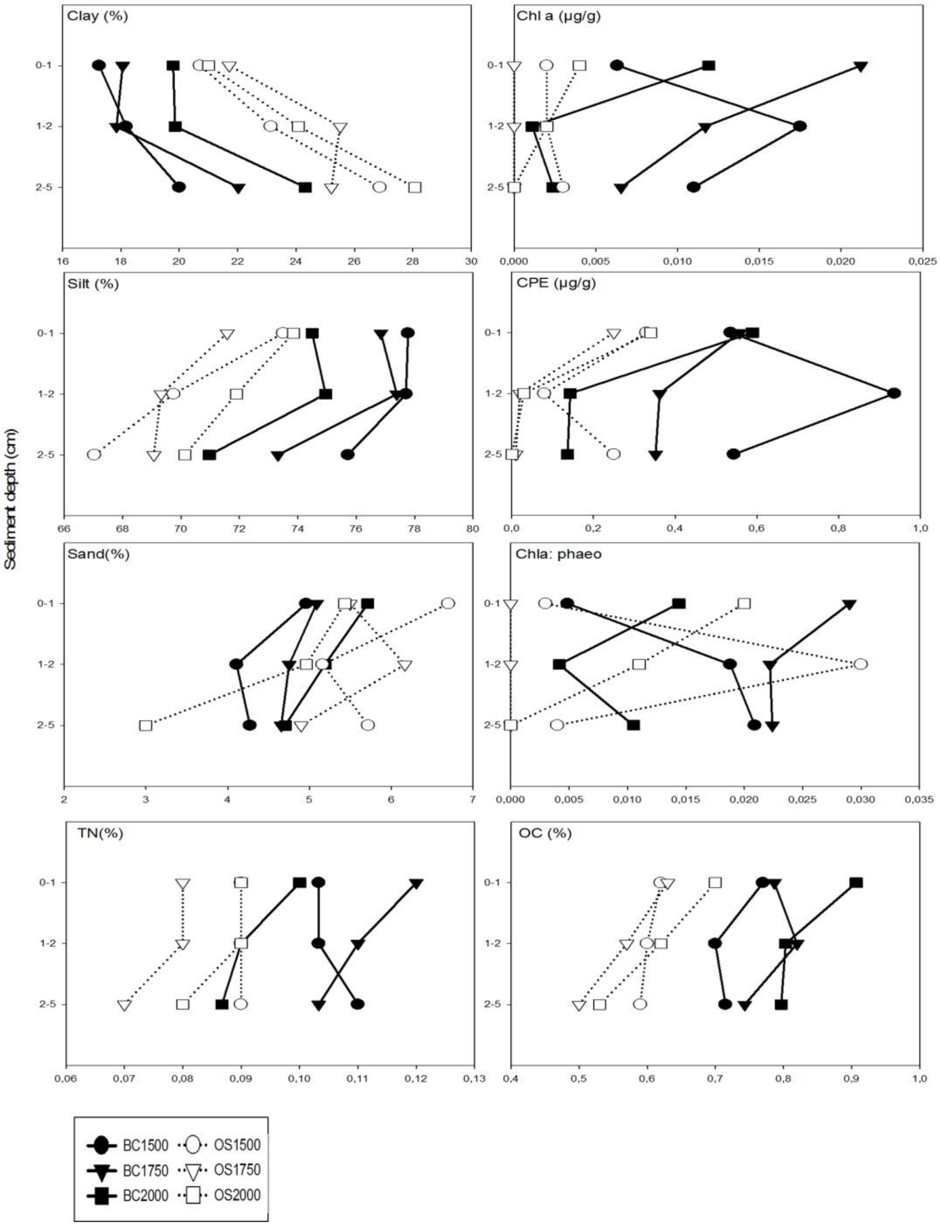


Figure 5.8. Average values for selected sedimentary parameters along the vertical sediment profile. Chl *a*: chlorophyll a; CPE: chloroplastic pigment equivalents; Chl-a:phaeo: chlorophyll a divided by its degradation products (phaeophytines) indicating 'freshness' of the phytodetrital OM; OC: organic carbon concentration; TN: total nitrogen concentration; Clay, Silt, Sand: volume percent clay, silt, sand content. BC= Blanes Canyon; OS=open slope. Values averaged over replicates.

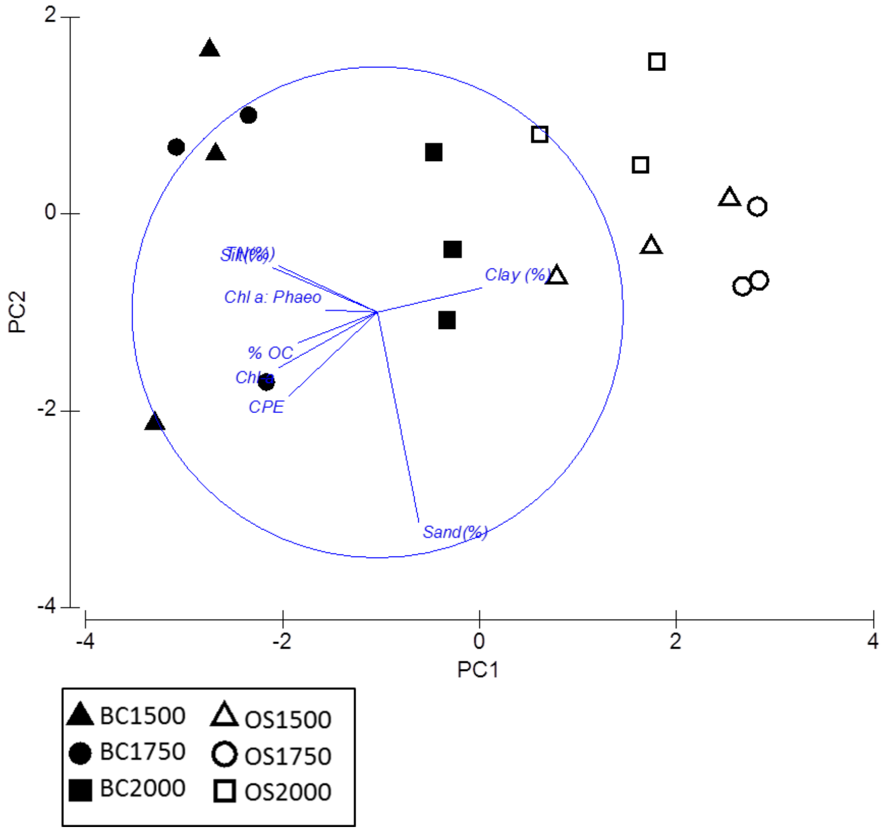


Figure 5.9. Principal component analysis (PCA) ordination based on selected environmental variables at the studied stations. Fill symbols represent canyon samples.

5.3.4 Relationship between nematode descriptors and environmental variables

At regional scale, environmental variables and univariate descriptors were only significantly correlated in the canyon (Spearman rank correlations, Table 5.7). Genus richness was negatively correlated with Chl *a* (-0.695) and CPE (-0.812), while Shannon index was positively correlated with CPE (0.750) and silt (0.900).

At local scale, the univariate descriptors in the canyon did not show significant correlations with any food-related variables (i.e. phytopigments and OC) and were positively and negatively correlated with silt and clay, respectively, while biomass was positively correlated with TN (Table 5.7). Contrastingly, on the slope, density was positively correlated with both CPE (0.413) and sand (0.421) and negatively correlated with the clay (-0.440). Genus richness was negatively correlated with Chl *a*, Chl *a*: phaeo, TN and OC, and *EG*(51) and Shannon index were negatively correlated with clay and OC, respectively.

Table 5.7. Spearman correlation coefficients between univariate nematode descriptors and sediment environmental variables at regional and local scale in the canyon and on the slope. Bold values indicate significant differences at $p < 0.05$.

Canyon	**Chl *a***	**CPE**	**Chl*a*: phaeo**	**OC**	**TN**	**Clay**	**Silt**	**Sand**
Density								
regional	0.133	-0.133	0.400	0.067	0.447	-0.183	0.183	-0.117
local	-0.036	-0.119	-0.324	0.011	0.205	**-0.721**	**0.592**	0.155
Biomass								
regional	0.533	0.350	0.000	-0.017	0.464	-0.450	0.250	0.167
local	0.083	-0.018	-0.075	-0.079	**0.408**	**-0.626**	**0.596**	-0.119
Genus richness								
regional	**-0.695**	**-0.812**	0.117	0.160	-0.258	0.653	-0.460	-0.167
local	-0.042	-0.127	-0.290	0.016	0.121	**-0.561**	**0.537**	0.032
EG (51)								
regional	0.200	0.183	0.217	0.184	-0.371	0.250	0.083	-0.100
local	0.075	0.002	-0.246	-0.015	0.145	**-0.665**	**0.584**	0.120
Shannon index								
regional	0.633	**0.750**	0.300	0.084	0.473	-0.683	**0.900**	-0.483
local	0.083	0.020	-0.233	-0.027	0.206	**-0.719**	**0.641**	0.175

Slope	**Chl-a**	**CPE**	**Chl*a*: phaeo**	**OC**	**TN**	**Clay**	**Silt**	**Sand**
Density								
regional	0.338	-0.200	0.338	0.429	0.353	-0.714	0.600	-0.429
local	0.070	**0.413**	0.082	0.217	0.182	**-0.440**	0.259	**0.421**
Biomass								
regional	0.101	-0.543	0.101	0.314	0.177	-0.771	0.714	-0.600
local	-0.361	-0.189	-0.354	-0.363	-0.225	-0.077	-0.018	0.090
Genus richness								
regional	-0.304	-0.257	-0.304	-0.086	-0.177	-0.371	0.086	-0.086
local	**-0.599**	-0.276	**-0.577**	**-0.538**	**-0.674**	0.015	-0.148	0.235
EG (51)								
regional	0.507	0.543	0.507	0.200	0.441	-0.086	-0.029	0.086
local	-0.335	0.307	-0.322	0.010	-0.152	**-0,52**	0.348	0.327
Shannon index								
regional	0.270	0.429	0.270	0.257	0.000	-0.029	-0.143	0.486
local	-0.448	-0.315	-0.410	**-0.473**	**-0,6**	0.054	-0.100	0.113

The spatial pattern based on environmental variables was only significantly related to the community structure inherent at the canyon (RELATE, regional: $\varrho=0.40$, $p<0.01$; local: $\varrho=0.17$, $p<0.01$). At regional scale, the best fitting model based on the environmental variables showed that TN explains nearly 22% of the variation observed, followed by silt (10.5%) (DISTLM, Table S2, Fig. 5.10A). At local scale, clay explains nearly 21 % and Chl *a* 6% of total variation of the community structure (DISTLM, Table S3, Fig. 5.10B).

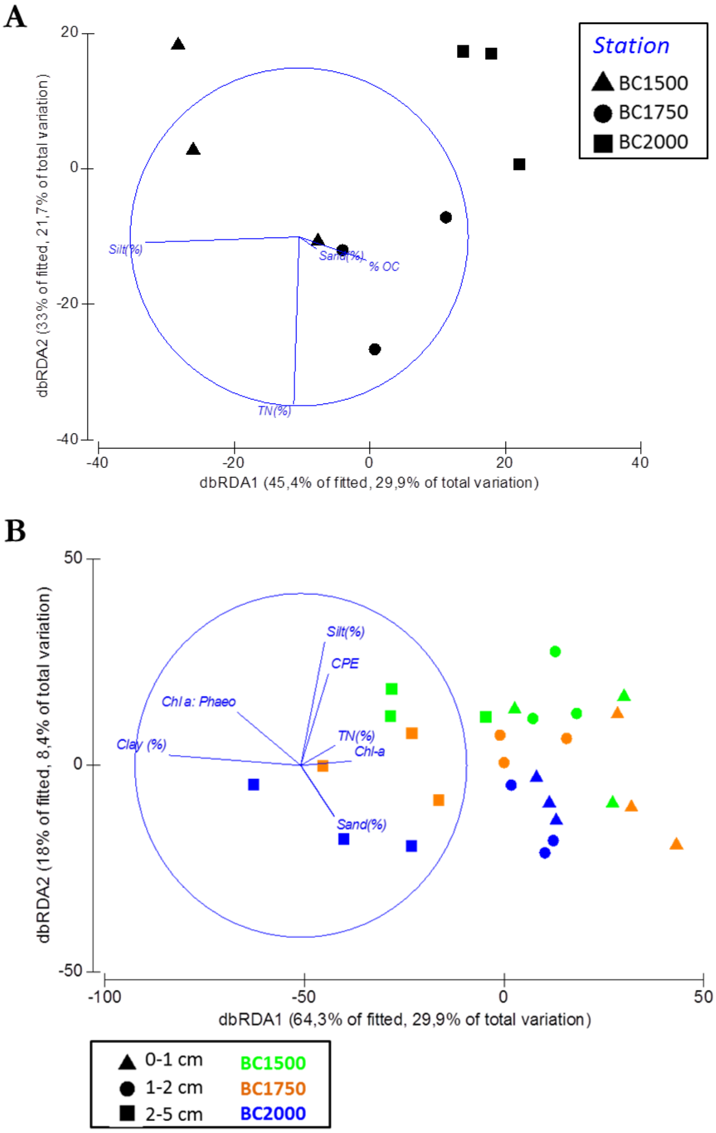


Figure 5.10. Distance-based redundancy (dBRDA) plots illustrating the DISTLM model based on the nematode genera assemblages and the fitted environmental variables as vectors for the canyon at **A)** at regional and **B**) at local scale.

5.4 DISCUSSION

5.4.1 Distribution of nematode community descriptors in the Blanes Canyon and its adjacent western slope

The number of benthic studies carried out on submarine canyons strongly increased during the last decades. However, much remains to be investigated since several studies report different results. Indeed biodiversity, as well as abundance and biomass patterns may contrast when canyon and slopes habitats are compared. Canyons are often characterized by higher faunal abundance and biomass, but lower diversity, compared to the adjacent slopes (Gambi and Danovaro, 2016; Danovaro et al., 2009; Vetter and Dayton, 1998; Curdia et al., 2004; Romano et al., 2013; Leduc et al., 2014; Gunton et al., 2015; Ingels et al., 2009).

Our results showed that all nematode community descriptors differed significantly between canyon and slope (Table 5.1). Density was higher in the canyon than on the slope, a difference that may be attributable to the 1,750 m depth assemblages, in accordance with previous studies (e.g., Ingels et al., 2009; Romano et al., 2013; Leduc et al., 2014; Gambi and Danovaro, 2016; Rosli et al., 2016). Similar differences in density between Blanes Canyon and its adjacent slope occurred for the meiofauna along a broad bathymetric gradient, from 500 to 2,000 m depth and for different seasons (Román et al.; 2016). Indeed in **Chapter 3** already concluded that the "classical" deep-sea principle of decreasing densities with the increasing water depth (e.g. Thiel 1983 and Tietjen, 1992; Soltwedel, 2000) only occurred on the slope. The existence of a "canyon effect" is widely accepted, as submarine canyons induce modifications in this bathymetric pattern of declining density due to their complex topography and heterogeneous hydrography (e.g. Canals et al., 2006; Levin and Sibuet, 2012). However, canyon patterns are not always consistent, as reported for several Mediterranean and NE Atlantic canyon/slope systems (Bianchelli et al., 2010; Gambi and Danovaro, 2016).

Nematode biomass was significantly higher in the canyon than on the slope, but there were no clear bathymetric differences in both habitats (Table 5.1, Fig. 5.2). Differenced in canyon *vs.* slope food availability often explain the density and biomass patterns for benthic organisms (Ingels et al., 2009; Soetaert et al., 1991a; Leduc et al., 2014; De Leo et al., 2010; Rosli et al., 2016). Accordingly, the available food (i.e., Chl *a*, CPE or OC) was higher in the Blanes Canyon than on its

western slope, resulting in a higher nematode biomass as previously reported for other canyons (Ingels et al., 2009; Leduc et al; 2014).
The canyon effect was also acting for the nematode diversity, mainly due to those genera present in the canyon and absent on the slope. However, most of them are rare genera so that we cannot discard under sampling as an important cause for the observed differences. In the canyon, diversity indices (except the Shannon index) did not display clear bathymetric pattern, while a decreasing trend with the increasing depth was observed on the slope, especially for genus richness. Such a lack of bathymetric pattern has been previously observed in several canyons, independently of the geographical region or the canyon-scale environmental conditions, both for the meiofauna higher taxa and nematodes (Danovaro et al., 2009; Bianchelli et al., 2010; Ingels et al., 2013; Romano et al., 2013b; Leduc et al., 2014; Pusceddu et al., 2013; Román et al., 2016), and Blanes Canyon is not an exception (**Chapter 3**).

Conversely, our observations are in contradiction with the general trend in which meiobenthic and nematode diversity are higher on the slopes than in canyons. This trend, has been explained by a higher availability of food in canyons, which would favour colonizer/opportunist species and, thus, a reduced diversity (Garcia et al., 2007; Danovaro et al., 2009; Ingels et al., 2009; Bianchelli et al., 2010; Bianchelli et al., 2013: Eastern Mediterranean; Leduc et al., 2014; Gambi and Danovaro, 2016). Overall, Blanes Canyon harbours a higher diversity than its adjacent western slope. That being said, there are studies supporting our observations, either or meiofauna higher taxa (Bianchelli et al., 2013; Román et al., 2016), nematode species (Danovaro et al., 1999; Bianchelli et al., 2013: NW and Central Mediterranean), macrofauna (Romano et al., 2013a; De Leo et al., 2014), or megafauna (Vetter et al., 2010; Ramirez-Llodra et al., 2010a). These attest the claim of submarine canyons as being considered *hots pots* of benthic biodiversity and biomass. Overall, Blanes Canyon has a higher genera turnover (i.e., β diversity) than the slope, thereby greatly contributing to the total Mediterranean diversity.

The nematode community structure also showed a canyon effect, with clear canyon *vs.* slope differences at 1,500 and 1,750 m depth, and with pronounced bathymetric differences in the canyon in contrast to the slope, where the three depths harbour more similar communities (Fig. 5.3). The same was observed in canyon/slope systems from different marine regions (NE Atlantic: Ingels et al., 2009; Gambi and Danovaro, 2016; Pacific Ocean-New Zealand, Leduc et al., 2014; Rosli et al., 2016; NW Mediterranean, Romano et al., 2013; Ingels et al., 2013; Román et al., 2016). The patterns based on genera abundances (PCO) agree well with those based on environmental variables (PCA), the latter being more similar in the slope stations compared to the canyon ones. Indeed, the deepest, less active canyon station (BC2000) more closely resembled the slope stations than any other canyon station. This agrees with the idea that slope environments tend to be homogeneous, in contrast canyons tending to show a higher heterogeneity (Vetter and Dayton, 1999; Garcia et al., 2007; Romano et al., 2013b; Román et al., 2016). Accordingly, a greater nematode turnover may be expected within canyon than within slope habitats (Ingels et al., 2009).

The dominant genera in our study area conform to the known patterns of dominance in Mediterranean slopes and canyons, including the marked dominance of *Sabatieria* (e.g. Vivier, 1978; Soetaert et al., 1995; Soetaert and Heip, 1995). Vanreusel et al. (2010) reported that the average nematode communities from abyssal plains, canyons and trenches shared several dominant genera with those in slopes. In Blanes Canyon, the dominant genera showed similar relative abundances: *Sabatieria*, *Acantholaimus* and *Halalaimus*. Particularly, *Sabatieria* (15%), *Halalaimus* (8.4%) and *Acantholaimus* (5%), together with *Theristus* (7%), and were also reported as dominant at the NE Catalan margin (Danovaro et al., 2009). In our study, *Sabatieria* was dominant in slope sediment as well as at 2,000 m depth in the canyon. This genus is usually associated with increasing levels of sediment disturbance, possibly due their capacity to thrive in low oxygen environments (Vincx et al., 1990; Schratzberger et al., 2009; Ingels et al., 2011b), as those that may result from high levels of sediment deposition and burial. The reasons why this genus is omnipresent and often dominant, however, remain somewhat unclear. *Sabatieria* also dominated in high hydrodynamic environments such as Gollum Channel (~20 %) (Ingels et al., 2011a). It was moderately dominant (~9 %) in organically enriched sediments in Nazaré Canyon (Ingels et al., 2009), it was not dominant in Cascais Canyon or at the deepest stations of the Setubal Canyon (4,500 m depth) (Ingels et al., 2011b), where the available OM was lower than in Nazaré Canyon. Using the genus-level approach, however, may have been masking species-specific responses since the species of *Sabatieria* respond differently to sediment oxygenation conditions (Moens et al., 2013). In addition, the species of *Sabatieria* are non-selective feeders (Wieser, 1953), which may represent a competitive advantage in exploiting low-quality food (Ingels et al., 2011b) such as those from the Blanes Canyon western slope sediments.

Blanes Canyon communities at 1,500 m depth were characterized by a relatively high abundance of predatory/scavenging genera (such as *Sphaerolaimus* and *Pomponema*), while non-selective deposit feeders (Wieser, 1953) were less numerous compared to the slope. A similar dominance pattern was found in Nazaré Canyon, where it was tentatively explained by the size of these particular feeding types compared to others (Ingels et al., 2009). Large predatory nematodes may be more agile and have superior mobility over smaller predators, being also able to penetrate deeper into sediment and, thus, easily avoid resuspension in the disturbed canyon sediments, thereby enhancing their survival rate. The higher fresh food inputs in the canyon relative to those reaching the slope may be the main reason explaining why the trophic complexity of canyons communities can be sustained. On the other hand, the exclusive presence of *Astomonema* in the canyon, particularly at 1,500 and 1,750 m depth, suggests the existence of sedimentary with reduced conditions, likely due to the very high sedimentation rates. Consequently, respiration and organic carbon burial conditions may be enhanced, thus creating the most appropriate conditions for these nematodes to thrive (Ingels et al., 2011a; Tchesunov et al., 2012; De Leonardis et al., 2008). The high densities at BC1750 were mainly owing to *Molgolaimus*, which may derive from previous depositional events increasing the OM levels at this depth in the canyon (see **Chapter 3**). Accordingly, this genus is often associated with organic enrichment and recent disturbing events (Vanhove et al., 1999; Lee et al; 2001).

5.4.2 Drivers of nematode communities

Nematodes are renowned for their patchy distribution at small (cm) spatial scales, both horizontally and vertically (e.g., Eckman and Thistle, 1988; Gallucci et al., 2009). Our results have shown that small-scale was a more important source of variability for the community structure and structural diversity, both in the canyon and on the slope, compared to bathymetry. Standing stocks showed the same trend, but only in the canyon. These observations are consistent with previous postulates for both meiofaunal and nematode assemblages indicating small-scale (cm) variability is more important than larger-scale (km) in explaining meiofaunal and nematode patterns of distribution (Ingels et al., 2009; Ingels et al., 2011a,b; Ingels and Vanreusel, 2013; Rosli et al., 2016). The large-scale differences in our study (as WD) had contrasting effects on the communities depending on the location. On the slope, WD mainly affected the structural diversity, while in the canyon; the main WD effects were in community structure and standing stocks. The small-scale effects were also illustrated by the occurrence of particular genera in different SL, which lead to higher generic turnover between the 0-1 cm and the 2-5 cm layers, both in the canyon and in the slope. The nematodes showed clear vertical profile gradients in community structure, with genera such as *Acantholaimus* being more common in surface than in deep sediments (e.g., Leduc et al., 2015). In turn, *Sabatieria* thrive in the deep SL suggesting reduced oxygen availability (Soetaert and Heip, 1995, Vanreusel et al., 1997; Muthumbi et al., 2004).

Furthermore, the relationship between nematode descriptors and environmental parameters were much more obvious at small than at large-scale, the latter being absent on the slope, likely as a response to its higher homogeneity and stability compared to Blanes Canyon.

Substrate heterogeneity has often been considered a possible cause of spatial variability in deep-sea benthic organisms and their biodiversity (e.g., Eckman and Thistle, 1988; Rex and Etter; 2010; Vanreusel et al., 2010), acting on various spatial scales and in concert with food availability. Food availability is particularly relevant for nematode distribution at different spatial scales, defining organism habitats at the scale of mm in the sediments up to large oceanographic areas; e.g. from patchy distributions in between the microtopographic features of the seafloor to areas that stretch hundreds of square kilometres and are under the influence of a particular productivity regime in surface waters (Gambi et al., 2014, Rosli et al., 2016). Surface export of primary production to the seafloor may also be influenced by water column processes and hence the food quality and quality arriving to the seabed is in part dependent on the water depth. In this context, small-scale horizontal patchiness may be driven by biotic interactions, variations in microtopography, disturbance, or food availability (Gallucci et al., 2009), whereas vertical patchiness is likely determined by vertical gradients in biogeochemical conditions (e.g., food availability and oxygen concentrations) (Jorissen et al., 1995; Soetaert et al. 2002), or by activity of macrofauna (e.g., predation, bioturbation, competition for food sources) (Braeckman et al. 2011; Gorska et al., 2014). Nematode population descriptors and grain size are often highly correlated, suggesting that large differences in distribution can be explained by sediment structure and heterogeneity, especially in the canyons where habitat heterogeneity is particularly enhanced compared to slope systems (e.g. Leduc et al., 2012b). The complex geology and hydrodynamic regimes associated with canyons may affect the sediment and OM deposition and accumulation, so that the vertical sediment profile acquires a pronounced structure, thereby enhancing the microhabitat´s diversity. Our results showed that food availability and nematode density and structural diversity on the slope were significantly more related at the local scale, whereas in the canyon stations, such a relationship was only expressed between food availability and nematode structural diversity (Table 5.7). This bias may obey to the marked canyon heterogeneity along sampled area (i.e., kilometres of canyon axis) that consequently gave rise to different habitat conditions. The higher homogeneity along the slope leads seabed differences to be more likely present on at the small-scale.

To some extent our results contradict the general idea that OM fluxes from the highly productive surface waters to the slope seafloor exert a considerable control on deep-sea meiobenthic standing stocks and diversity (Soltwedel, 2000; Gambi and Danovaro, 2006; Ingels et al., 2009, Ingels et al., 2011a,b; Leduc et al., 2014; Lins et al., 2014; Lins et al.,2015). Accordingly, we suggest either that our sampling ranges along the slope did not include surface export regimes distinct enough or that other factors might be involved in controlling the observed nematode distribution in the deepest regions of Blanes Canyon and its adjacent slope.

Water and sediment transport dynamics, as well as morphology, may play an important role in controlling canyon biota. In the Blanes area, the velocity of particle fluxes is higher in the canyon than on the adjacent slope, particularly in the upper canyon where it is reinforced by an increased lateral transport in the mid axis region (Zúniga et al., 2009; Lopez-Fernandez et al., 2013).

All our analyses pointed to the deepest canyon station having a nematode community closely resembling those of the nearby slope. This result, together with the fact that this area has a very limited topographic relief, lead us conclude that the deepest canyon station (and the slope ones) lies outside the region suffering the “canyon effect”. Conversely, BC1500 and BC1750 showed more heterogeneous communities and environmental characteristics, in parallel with the complex topography characteristically derived from enhanced transport, deposition and fluxes along the canyon axis and flanks. Accordingly, the high TN throughout the whole vertical profile at both stations suggests the occurrence of high sedimentation rhythms and burial events.

The evident small-scale differences at the community level may derive from the relative vulnerability of nematodes to disturbance. Especially in the canyon, this appears certainly to be related to the ecological impact of bottom trawling activities and its associated sedimentary alterations (see **Chapter 2**). Blanes Canyon, as other submarine canyons along the Catalan margin, supports important bottom trawling fishing grounds in its head and flank regions, where mostly (but not exclusively) the deep-sea red shrimp, *Aristeus antennatus* is targeted (Sardà et al., 1994a; Tudela et al., 2003). Fish trawling activities cause seafloor erosion, with widespread damage throughout the sediment column as a result of increased sediment resuspension and deposition and sediment particle size alteration in the trawled and surrounding areas (Martin et al., 2014a; Martin et al., 2014b; Puig et al., 2012; Puig et al., 2015a, Puig et al., 2015b). Moreover, food availability is also reduced within the sediment in the fishing grounds themselves (Martín et al., 2014b). These habitat modifications may impact the infauna (Pusceddu et al., 2014; Shratzberger et al., 2009) including nematode assemblages. In addition, the generally steep topography of canyon habitats makes them prone to slope instability and turbidity following trawling events on the escarpments and interfluvial areas (Puig et al., 2012), which in turn may remove organic-rich sediment down-slope to deeper parts of the canyon (Puig et al., 2012; Pusceddu et al., 2014), causing potential disturbance to deeper fauna, as well as supporting infaunal proliferations at locations deeper than expected.

The impact of surface sediment remobilization and relocation on the fauna has been recently assessed along the axes of the nearby Arenys, Besòs and Morrás Canyons (Paradis et al., 2017). These authors postulate bottom trawling (i.e., through enhanced sedimentation rates) as a major cause altering natural sedimentary environments, independently of the canyons´-morphology. Trawl-induced sedimentation caused three- to fourfold increase in sedimentation rates in comparison with the natural accumulation. Based on the analysis of the meiofauna along the Blanes Canyon axis, was hypothesized that major trawling impacts in its upper (900 m depth) and mid (1,200 m depth) regions gave rise to the presence of anthropogenic depocenters, subsequently causing increases in infaunal standing stocks (Román et al., 2016). Recent observations on deposition rates in Blanes Canyon revealed that, although sedimentation rates tended to decrease down to 1,200 m depth, it also occurred at 1,500 and 1,800 m depth. Natural accumulation rates would be close to 0.21 and 0.18 $\text{cm}\cdot\text{year}^{-1}$ at 1500 and 1800 m depth, respectively. In turn, the actual rates were 0.88 and 0.65 $\text{cm}\cdot\text{year}^{-1}$, respectively; this being apparently entirely attributable to the resuspension caused by trawling activities (Paradis, personal communication). Down to the deepest canyon region, far away from the sources of anthropogenic disturbance (i.e. at 2,000 m depth), the trawling influence was reduced and the actual sedimentation rates did not differ from the natural ones (Paradis, personal communication). As a consequence, both the sediment characteristics and the nematode community descriptors more closely resemble those of the nearby slope. Our results confirm that the trawling activities may be considered as a major large- and small-scale functional driver in the highly anthropic Blanes submarine canyon. The induced increases in sedimentation rates are thus reflected in the sediment qualitative and quantitative characteristics, as well as in the altered vertical profiles and, ultimately, in the nematode assemblages they contain. Both are mirrored by the nematode assemblages, which did not revert to a "natural" status until dissappearing the influence of trawling.

5.5 SUPPLEMENTARY DATA

Table S5.1. 2- factor PERMANOVA pair-wise test results for significant water depth (WD) and double interactions (LO x WD) based on nematode descriptors: A) Density; B) Genus richness; C) Shannon-Wiener; D) Community structure . Lo: location and WD: water depth; perms: possible permutations; P (MC): Monte-Carlo p-values. Bold P(MC) values: $p < 0.05$; bold italic values: $p < 0.01$. BC: Blanes Canyon; OS: Open slope.

A) Nematode density					
Within WD	**Comparisons**	**t**	**P(perm)**	**perms**	**P(MC)**
1500	BC, OS	0.61619	0.6035	10	0.5706
1750	BC, OS	11.124	0.1012	10	**0.0005**
2000	BC, OS	25.951	0.0984	10	0.0605
Within Lo	**Comparisons**	**t**	**P(perm)**	**perms**	**P(MC)**
BC	1500, 1750	6.071	0.0995	10	***0.0035***
BC	1500, 2000	7.18E-03	1	10	0.9952
BC	1750, 2000	20.831	0.2032	10	0.1043
OS	1500, 1750	4.787	0.1001	10	***0.0096***
OS	1500, 2000	80.955	0.0947	10	***0.0009***
OS	1750, 2000	19.263	0.2081	10	0.1261
B) Genus richness					
Within WD	**Comparisons**	**t**	**P(perm)**	**perms**	**P(MC)**
1500	BC, OS	0.11471	1	7	0.9141
1750	BC, OS	12.649	0.3979	7	0.2802
2000	BC, OS	12.075	0.1061	8	***0.0003***
Within Lo	**Comparisons**	**t**	**P(perm)**	**perms**	**P(MC)**
BC	1500, 1750	0.5547	0.6922	7	0.6129
BC	1500, 2000	1.524	0.2995	8	0.2024
BC	1750, 2000	2	0.1985	5	0.1135
OS	1500, 1750	0.625	0.6927	7	0.5697
OS	1500, 2000	73.744	0.1049	8	***0.0018***
OS	1750, 2000	57.287	0.1028	9	***0.0037***
C) Shannon-Wiener					
	Comparisons	**t**	**P(perm)**	**perms**	
	1500, 1750	9.91E-02	0.9238	8792	
	1500, 2000	35.128	**0.0139**	8764	
	1750, 2000	3.098	**0.0167**	8793	

Table S5.1: *continued*

D) Community structure					
Within WD	**Comparisons**	**t**	**P(perm)**	**perms**	**P(MC)**
1500	BC, OS	2.176	0.0997	10	**0.021**
1750	BC, OS	19.262	0.1029	10	**0.0394**
2000	BC, OS	14.182	0.2034	10	0.1547
Within Lo	**Comparisons**	**t**	**P(perm)**	**perms**	**P(MC)**
BC	1500, 1750	16.869	0.0977	10	**0.0514**
BC	1500, 2000	18.943	0.0996	10	**0.0405**
BC	1750, 2000	17.541	0.0994	10	**0.0308**
OS	1500, 1750	10.588	0.4097	10	0.3726
OS	1500, 2000	12.872	0.2031	10	0.2159
OS	1750, 2000	12.532	0.2023	10	0.2441

Table S5.2. Distance-based linear model (DISTLM) for nematode community structure and selected environmental variables at **regional scale** for canyon habitats.

MARGINAL TESTS				
Variable	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**
Clay (%)	2325.6	2.4481	0.003	0.25911
Silt(%)	2354.4	2.4891	0.009	0.262
Sand(%)	876.55	0.75762	0.679	0.0977
TN(%)	1956	1.9506	0.027	0.21793
% OC	1364.1	1.2545	0.246	0.15198
Chl *a*	1405.4	1.2996	0.217	0.157
Chl*a*:phaeo	1888.3	1.865	0.042	0.21038
CPE	1998	2.0045	0.013	0.22261

SEQUENTIAL TESTS (Best model construction)							
Variable	Adjusted R^2	SS(trace)	Pseudo-F	P	Prop.	Cumul.	res.df
+Silt(%)	0.15693	2354.4	2.4891	0.005	0.26231	0.26231	7
+TN(%)	0.2884	1830.9	2.2933	0.018	0.204	0.4663	6
+% OC	0.31065	923.15	1.1936	0.296	0.103	0.56915	5
+Sand(%)	0.31607	797.74	1.0397	0.487	0.0889	0.65804	4

Marginal tests: explanation of variation for each variable taken alone. Sequential tests: conditional tests of individual variables in constructing the model. Each test examines whether adding the variable contributes significantly to the explained variation. Selection procedure: step-wise, selection criterion: adjusted R^2. Prop.: % variation explained. Cumul.: cumulative variation explained. Chl *a*= Chlorophyll *a*; CPE= chloroplastic phaeopigment equivalents; Chl *a*: phaeo= Chlorophyll a: phaeopigments; TN= total nitrogen content; OC= organic carbon content.

Table S5.3. Distance-based linear model (DISTLM) for nematode community structure and selected environmental variables at **local scale** for canyon habitats.

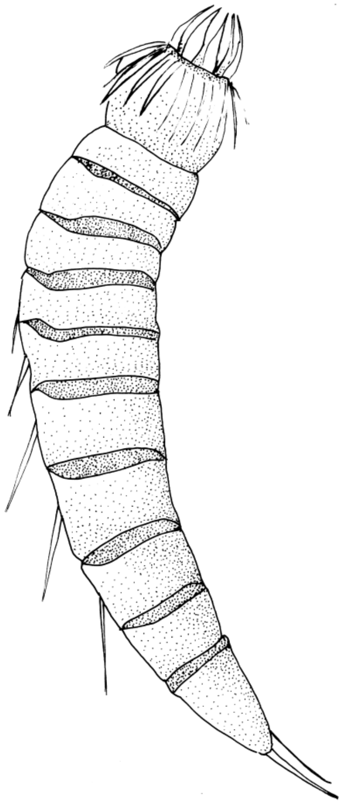
MARGINAL TESTS				
Variable	**SS(trace)**	**Pseudo-F**	**P**	**Prop.**
Clay (%)	12146	66.333	0.001	0.20969
Silt(%)	8724.3	44.333	0.006	0.15062
Sand(%)	3204.4	14.641	0.157	0.0553
TN(%)	3779.4	17.451	0.131	0.0653
% OC	1404.3	0.62119	0.735	0.0242
Chl *a*	2847.3	12.925	0.237	0.0492
Chl*a*:phaeo	2075	0.92889	0.425	0.0358
CPE	4129.2	1.919	0.086	0.0713

SEQUENTIAL TESTS (Best model construction)							
Variable	Adjusted R^2	SS(trace)	Pseudo-F	P	Prop.	Cumul.	res.df
+Clay (%)	0.17808	12146	6.6333	0.001	0.20969	0.20969	25
+Silt(%)	0.18919	2425	1.3425	0.205	0.0419	0.25156	24
+Sand(%)	0.20525	2629.2	1.485	0.15	0.0454	0.29695	23
+CPE	0.2121	2106.3	1.2	0.25	0.0364	0.33332	22
+Chl *a*: phaeo	0.21784	2023.7	1.1614	0.276	0.0349	0.36826	21
+Chl-a	0.26007	3624	2.1985	0.047	0.0626	0.43082	20
+TN(%)	0.2684	2001	1.2277	0.263	0.0345	0.46537	19

Marginal tests: explanation of variation for each variable taken alone. Sequential tests: conditional tests of individual variables in constructing the model. Each test examines whether adding the variable contributes significantly to the explained variation. Selection procedure: step-wise, selection criterion: adjusted R^2. Prop.: % variation explained. Cumul.: cumulative variation explained. Chl *a*= Chlorophyll *a*; CPE= chloroplastic phaeopigment equivalents; Chl *a*: phaeo= Chlorophyll *a*: phaeopigments; TN= total nitrogen content; OC= organic carbon content.

6

Diversity and distribution of deep-sea Kinorhynchs along the Canyon and its adjacent Open Slope



6.1 INTRODUCTION

Deep-sea benthic metazoan meiofauna inhabits the interstitial spaces between sediment grains in vast numbers. This habitat is largely dominated by copepods and, particularly, by nematodes, the latter often representing more than > 85% of the total abundance (Lambshead, 2004; Mokievskii et al., 2007; Vincx et al., 1994). This biased pattern of dominance frequently masks the changes and the relative importance of the "rare" meiofaunal taxa. However, when considered, rare meiofaunal taxa have demonstrated important differences in deep-sea habitats, such as submarine canyons and their adjacent open slopes (Bianchelli et al.; 2010).

The Kinorhyncha, the so-called "mud dragons" is an exclusively meiobenthic phylum of marine invertebrates (Higgins and Thiel, 1988; Kristensen and Higgins, 1991), considered part of the "rare" meiofauna. Since the first kinorhynch record approximately 170 years ago by Dujardin, (1851), more than 250 species nested in 29 genera have been described (Herranz et al., 2014; Herranz et al., 2017; Pardos et al. 2016 a,b; Sánchez et al., 2014a,b; Sánchez et al., 2016; Sørensen et al. 2015, Yamasaki 2016). Kinorhynchs are known to occur from polar to tropical regions and from intertidal to abyssal depths, with the deepest recording being at 7,800 m in the Atacama Trench, South Pacific Ocean (Danovaro et al., 2002).

Even though kinorhynchs are known to occur in high diversity in the deep-sea (Neuhaus and Blanche, 2006), they are usually reported in ecological papers at "phylum" level, or, simply among "other groups" (i.e., those representing < 1 – 2 % of the total deep-sea meiofauna), resulting in a loss of valuable information (see Table 13 in Neuhaus, 2013). Studies on deep-sea Kinorhyncha identified at the species level have been scarce and scattered covering different regions of the world: the Guinea and Angola Basins (ca. 5,000 m depth), the Kuril-Kamchatka Trench (ca. 5,200 m depth), New Zealand (ca. 600-3,000 m), the Southern Atlantic Ocean (c.a. 2,300 m depth) and Fieberling Guyot off the Pacific coast of North America (500 to 700 m depth). All these are taxonomic studies describing new genera (Neuhaus and Blasche, 2006; Sánchez et al., 2014b; Sørensen, 2008) and species (Bauer-Nebelsick 1996; Nehaus and Blasche, 2006; Sánchez et al., 2014a,b; Sørensen, 2008; Sørensen and Landers, 2014; Sørensen et al., 2016; Neuhaus and Sorensen 2013; Adrianov and Maiorova 2016).

Despite that the available information on mud dragon diversity and distribution is gradually increasing, information on biogeographical patterns is still highly incomplete and biased, usually reflecting sampling effort rather than real distributions. Also, species distributions at a local scale and their relationships with abiotic factors are still poorly understood, because this information is practically absent for deep-sea environments.

During the last decade, intensive sampling campaigns along the Iberian Peninsula coasts have provided a significant increase in the knowledge on kinorhynch diversity (Herranz et al., 2011; Sánchez et al., 2012; Sánchez-Tocino et al., 2010; Neves et al. 2016). However, most of these studies were restricted to subtidal zones (i.e., down to a maximum of 100 m depth). This particular range is mirrored by most Mediterranean surveys dealing with kinorhynchs (Zelinka, 1928; Nebelsick, 1990; Dal Zotto and Todaro, 2016; Yildiz et al. 2016). Several ecological studies have reported the presence of kinorhynchs from a wide range of deep-sea Mediterranean habitats, from continental slopes to bathyal and abyssal plains, including submarine canyons (e.g., Danovaro et al., 2009; Bianchelli et al., 2010; Gambi et al., 2010; Romano et al., 2013b).

Submarine canyons are steep-sided valleys cut into the oceans´ continental slopes. They form complex systems in terms of topography, hydrography and sedimentology and have been proven to be hot spots for meiofaunal assemblages density, biomass and diversity by funnelling organic matter from surface waters and continental shelf areas to the deep sea where it is deposited (Romano et al., 2013b; Ingels et al. 2011a, 2013, Román et al. 2016). Kinorhynchs have often been reported as part of these meiofaunal assemblages. However, nothing is known about the overall deep-sea diversity of mud dragons in submarine canyons.

In previous studies carried out in the Blanes Canyon by Romano et al. (2013b) and in this present work in **Chapter 3,** deep-sea kinorhynchs have been regularly reported at low densities, grouped within the “rare taxa”, but have never been studied in depth.

Framed within the above-mentioned Iberian surveys, the NW Mediterranean littoral has already been qualitatively sampled looking for kinorhynchs biodiversity, which revealed the shallow waters (i.e., from 11 to 60 m depth) off Blanes as a *hot spot* for mud dragons diversity (Sánchez et al., 2012).

In this chapter the kinorhynch communities from Blanes Canyon and its adjacent open slope were analysed, and with this study we are providing the first quantitative study describing the local density and biodiversity patterns of deep-sea kinorhynch communities from a submarine canyon system. The purpose of this study is twofold. First, to analyse the distribution of kinorhynch densities along a bathymetric gradient (500 – 2,000 m depth) and throughout the vertical sediment profile, as well as to assess the relationships between kinorhynch densities and sediment environmental variables. Second, to increase the knowledge on kinorhynch diversity and species distribution in the area, complementing previous shallow water studies. Future publications will specifically address the taxonomy of the species and describe the morphotypes found during the present study, including the formal description of the taxa here identified as undescribed.

6.2 MATERIAL AND METHODS

6.2.1 Sampling

In this study we considered the same sampling design and stations as targeted in **Chapter 3**. It includes, all the sampling cruises carried out during the DOSMARES project (spring and autumn 2012 and 2013), and covers both canyon (BC) and its adjacent slope (OS) from ca. 500 to ca. 2000 m depth (BC) and five more on the adjacent western slope (OS) (see **Chapter 2** section **2.2** and Table 2.1for further sampling details) (Fig. 6.1)

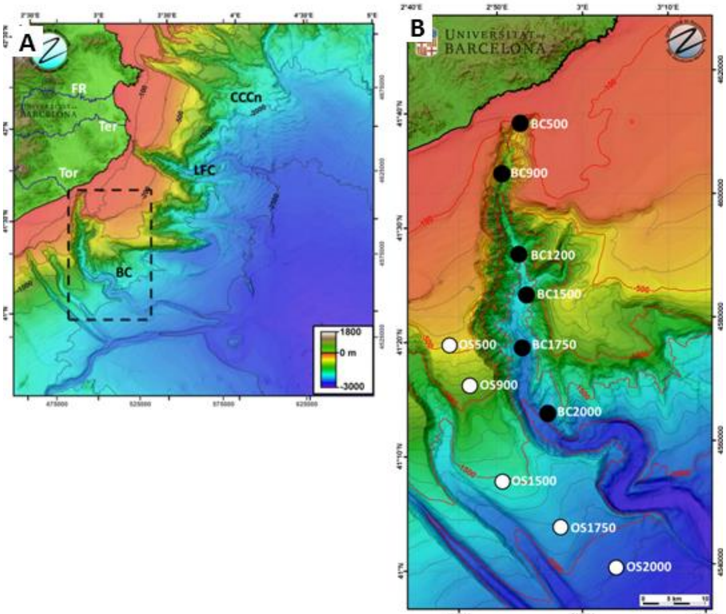


Figure 6.1. (**A)** Detailed bathymetric map of the the north-Catalan margin showing the position of the three main submarine canyons. (**B)** Detailed bathymetric map of Blanes Canyon system showing the position of the sampling stations (white dots). ToR: Tordera River, TeR: Ter River, FR: Fluvià River; BC: Blanes canyon, OS: Open slope. (Canals et al., 2004).

Three multicorer deployments (replicates) were conducted at each sampling station. From each multicore deployment, one core was used for meiofaunal and three for sediment analyses, which were all carefully sub-sampled on board by means of a small PVC tube (36 mm of diameter, 5 cm sediment depth) taken from the center of the core to maintain a consistent sample surface area for all replicates. The three sediment layers from each subcore (see **Chapter 2**, section **2.2** for further details) were used separately in the analyses. The methods to determine the quantity and quality of sediments variables are fully described in **Chapter 2**.

6.2.2 Environmental data

Grain size (clay, silt and sand fractions), Chlorophyll *a* (Chl *a,* μg/g) and chlorophyll degradation products in the sediment, organic carbon concentration (OC%) and total nitrogen concentration (TN%) sediment variables were considered in the present studied. Moreover, Chloroplastic Pigments Equivalents (CPE: sum of Chl *a* and its degradation products as phaeopigments) were used to estimate surface-produced organic matter (OM). The ratio Chl *a:* phaeopigments (Chl *a*: phaeo) is used as a proxy to estimate the freshness of photosynthetically derived OM (Thiel, 1978).

6.2.3 Meiofauna and kinorhynch analyses

The techniques to analyse meiofauna samples are fully detailed in **Chapter 2** section **2.4.1**. Kinorhynch specimens were sorted from the remaining meiofauna, counted under a stereomicroscope (50 x magnifications), and subsequently preserved in 70% ethanol until identification. Specimens for light microscopy observations (LM) were dehydrated through a graded series of ethanol and transferred to glycerin prior tomounting in Fluoromount G®. They were examined and photographed using a Zeiss Axioplan 2 microscope with differential interference contrast optics (DIC) equipped with a Zeiss-Axiocam503-color camera. Specimens for scanning electron microscopy (SEM) were cleaned by exposing them to ultrasound intervals of 5–10 s, and then dehydrated through a graded series of ethanol, critical point dried, mounted on aluminium stubs, sputter coated with platinum-palladium, and imaged with a Hitachi S4700 field emission scanning electron microscope. Coating and SEM imaging were performed at the Bioimaging Facility at the University of British Columbia (UBC).

Terminology follows Sørensen and Pardos (2008). General kinorhynch classification follows Sørensen et al. (2015). Undescribed morphotypes were designed as sp. Juveniles of all species were undistinguishable and, thus, were considered altogether. All examined material is currently deposited at the MH´s personal collection (UBC).

6.2.3 Data processing

Regression analyses were performed to reveal the form, distribution and significance of the functional relationship between density and depth, for both canyon and slope samples. Additionally, Spearman Rank correlations were used to assess the strength of the relationship between the selected environmental variables (clay, silt, sand, OC, TN, Chl *a*, CPE, and Chl *a*: phaeo) and density for both canyon and slope habitats separately, by means of XLSTAT (Addinsoft) software.

6.3 RESULTS

6.3.1 Environmental variables

The environmental data used herein were fully described in **Chapter 3**. The main trends relevant for the kinorhynch assemblages are as follows. The sediments collected were typically muddy but showed a higher heterogeneity in the canyon than in the slope. The highest sand contents were found at BC1200. Clay contents were higher at the slope and increased with the increasing depth and along the vertical sediment profile (Table S1). Food quantity (as Chl *a* and CPE) tended to decrease with increasing depth along the slope, and were mainly concentrated at shallower stations in the canyon, where a particularly high peak of CPE occurred at 1,200 m depth. Moreover, food quantity was mostly concentrated in the surface sediment layer (0-1 cm) and, decreased along the vertical sediment profile (Table S1).

6.3.2 Kinorhynch densities and spatial distribution

We collected 295 kinorhynch specimens, which represented less than 2% (canyon) and 1% (slope) of the total meiofauna. Thus, they were considered as "other taxa" in **Chapter 3**. However, among these "other taxa", its relative abundance was 41 % in the canyon, and 15 % on the slope (Fig. 6.2).

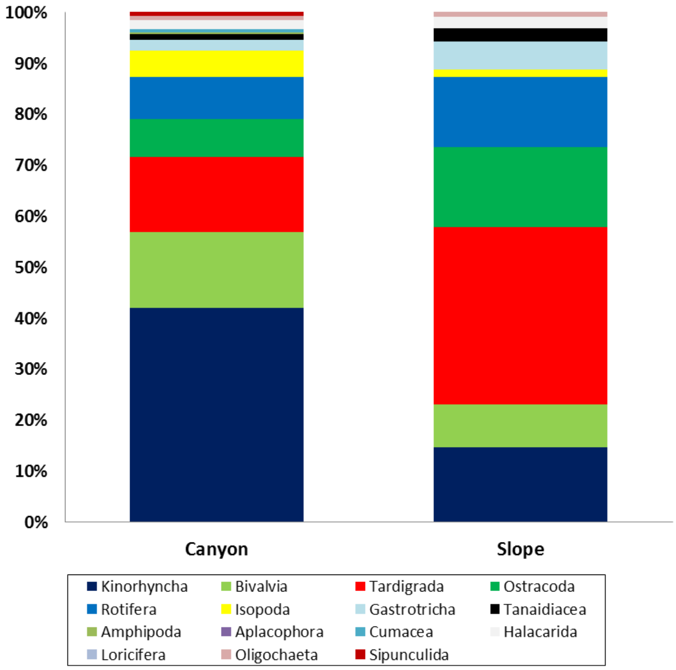
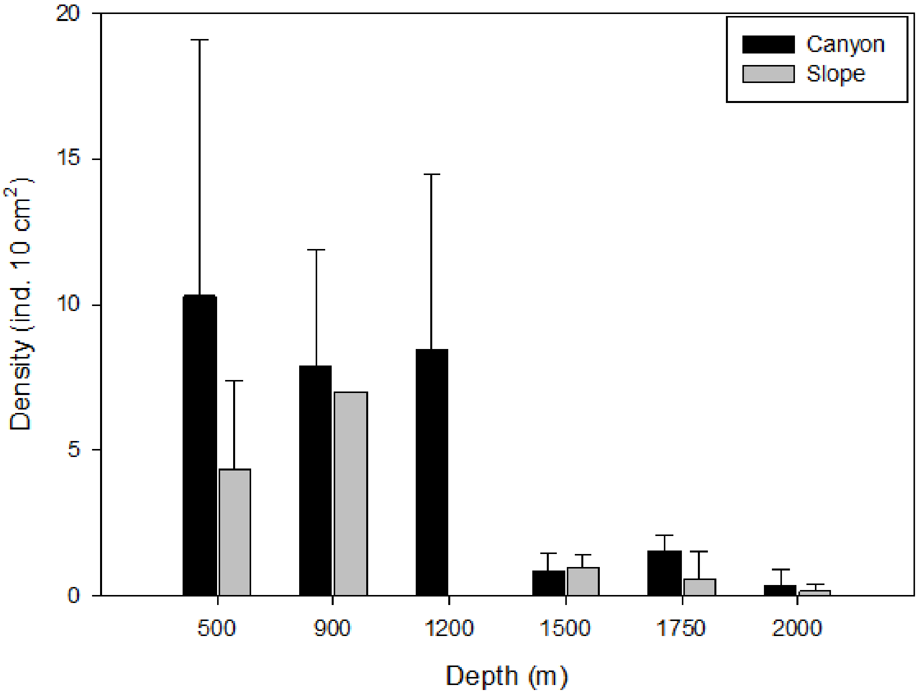


Figure 6.2. Relative abundance (as average of all samples) of taxa including less than 2% of the total meiofauna for the canyon and slope

Overall, mean densities (as sum of the three sediment layers) significantly decreased with increasing water depth in the canyon (R= 0.328) and on the slope (R= 0.582) (Fig. 6.3). The highest densities were registered in autumn 2012 and spring 2013 (Table 6.3), particularly at BC500 (47 in total; 23.5 ± 17.6 on average; Table 6.1). The lowest densities were registered at BC2000 and OS2000 (two and one specimens, respectively).



.**Figure 6.3.** Kinorhynch overall bathymetric density distribution (0-5 cm sediment depth, average ± standard deviation) distribution for canyon and slope stations.

Along the vertical sediment profile, kinorhynchs were 1) mainly concentrated in the 0-1 cm layer (Fig. 6.4; Table 6.1) and 2) almost equally distributed in 0-1 cm and 1-2 cm layers at BC500. Kinorhynchs penetrated deeper into the sediment in the canyon than on the slope. Accordingly, we found them at the 2-5 cm layer until 1,500 m depth in the canyon. On the slope, they were scarcely present in the 1-2cm layer (and only at OS900 and OS1500) and were absent from the 2-5 cm layer (Fig. 6.4).

Canyon densities were positively correlated with OC, Chl *a*, CPE, and Chl *a*: phaeo as well as with TN (Table 6.2). On the slope, densities were 1) positively correlated with OC, Chl *a*, CPE, Chl *a*: phaeo and with silt content, and 2) negatively correlated with clay content (Table 6.2).

Table 6.1. Average density of kinorhynchs (ind. 10 cm^{-2}) at each period.sampled in the Blanes Canyon system. NS: not sampled.

Station	Spring 2012	Autumn 2012	Spring 2013	Autumn 2013
BC500	6.0	23.5	6.0	8.3
BC900	3.0	6.3	11.0	11.3
BC1200	2.0	9.3	9.3	NS
BC1500	1.5	1.3	0.3	0.3
BC1750	1.5	1.3	1.3	0.0
BC2000	1.0	0.0	NS	0.0
OS500	NS	NS	4.3	NS
OS900	NS	NS	7.0	NS
OS1500	NS	0.0	1,30	NS
OS1750	0.0	0.0	2.0	0.0
OS2000	0.0	0.0	NS	NS

Table 6.2. Coefficients for the Spearman correlations between kinorhynch densities and sediment variables. Bold: $p < 0.05$. TN: total nitrogen, OC: organic carbon; Chl *a*: Chlorophyll a; CPE: Chloroplastic Pigment Equivalents; Chl *a*: phaeo: Chlorophyll *a*: phaeopigments ratio.

Variables	Canyon	Slope
Clay	0.060	**-0.463**
Silt	-0.020	**0.249**
Sand	0.055	0.113
TN	**0.172**	0.032
OC	**0.197**	**0.415**
Chl *a*	**0.447**	**0.394**
CPE	**0.354**	**0.516**
Chl *a*: phaeo	**0.254**	**0.259**

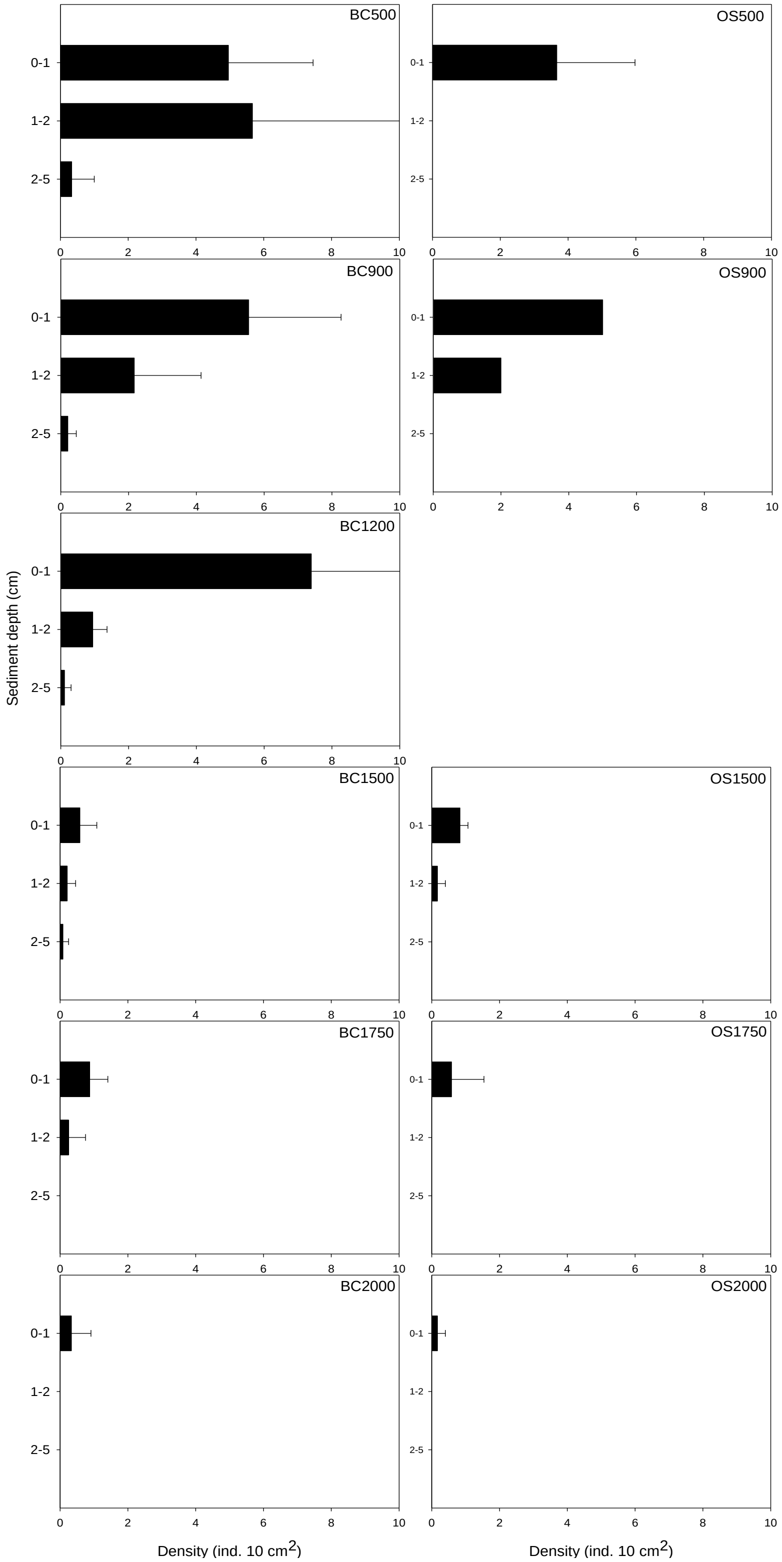


Figure 6.4. Vertical profile in the sediment of the kinorhynch density (average values of all samples).

6.3.3 Taxonomic considerations

Nine different species nested in four genera and three families were recorded, all of them belonging to the Class Cyclorhagida (Zelinka, 1896) Sørensen et al. 2015 (Figs 6.5, 6.6). The taxonomic information on each species recorded follows below.

Order Echinorhagata Sørensen et al., 2015

Family Echinoderidae (Zelinka, 1894)

Genus *Echinoderes* Claparède, 1863

Echinoderes lusitanicus Neves et al., 2016 (Fig 6.5D-E).

Echinoderes lusitanicus was easily identified by showing: tubes on subdorsal and ventrolateral positions on segment 2, lateroventral on segment 5, lateral accessory on segment 8 and laterodorsal on segment 10, and five long middorsal spines on segments 4-8, lateroventral spines on segments 8-9 and short lateral terminal spines and minute lateral accessory spines in females (Fig. 6.5D-E).

The remaining echinoderid specimens found in this study belong to 5 unknown species, named *Echinoderes* sp.1 to sp.5 until formal descriptions are published. However, we provide their diagnostic characters below:

Echinoderes sp.1 (Fig. 6.6D):

Morphotype characterized by 5 long middorsal spines (segments 4-8), lateroventral spines on segments 6-9; lateroventral tubes on segment 5 and laterodorsal on segment 10; glandular cell outlets type 2 subdorsal, laterodorsal and ventrolateral on segment 2, and midlateral on segments 5 and 8.

The combination of all this characters makes these specimens easily distinguishable from the more than 50 known species having a similar middorsal and lateroventral spine pattern.

Echinoderes sp.2 (Fig. 6.6E):

Morphotype characterized by a single short middorsal spine on segment 4, lateroventral spines on segments 6-7, absence of tubes; glandular outlets type 2 in subdorsal, laterodorsal, sublateral and ventrolateral positions on segment 2, and sublateral on segments 5 and 8.

Echinoderes sp.3 (Fig. 6.6B):

Morphotype characterized by three long middorsal spines on segments 4, 6, 8; lateroventral tubes on segment 5, lateral accessory on segment 8 and laterodorsal on segment 10; lateroventral spines on segments 6-9; glandular cell outlets type 2 in subdorsal and sublateral positions on segment 2.

Echinoderes sp.4 (Fig. 6.6F):

Morphotype characterized by three middorsal spines on segments 4, 6, 8; laterodorsal and ventrolateral tubes on segment 2, lateroventral on segment 5, lateral accessory on segment 8 and laterodorsal on segment 10; short lateroventral spines on segments 6-9; elongated tergal extensions.

Echinoderes sp.5 (Fig. 6.6G-H):

Morphotype characterized by three very long middorsal spines on segments 4, 6, 8; long tubes in ventrolateral position on segment 5, lateral accessory position on segment 9 and laterodorsal on segment 10; very long and thin lateroventral spines on segments 8 and 9; lateral accessory spines in females very robust.

Order Kentrorhagata Sørensen et al. 2015
Family Centroderidae Zelinka, 1896
Genus *Centroderes* Zelinka, 1907
Centroderes spinosus Reinhardt, 1881 (Fig. 5A-C)

Centroderes spinosus was easy to identify by lateroventral tubes on segments 2 and 5 and lateroventral spines on segments 8 and 9; the spine on segment 8 is very thick and conical in contrast with the thin acicular spine on segment 9 (Fig. 6.5A, C).

Genus *Condyloderes* Higgins, 1969
Condyloderes sp. (Fig. 6.6C):

Morphotype characterized by a unique cuspidate and acicular spine formula (cuspidates: ventrolateral on segments 5 and 8, sublateral on segment 9; acicular: middorsal segments 1-10, lateroventral segments 1-10; subdorsal segment 10; midterminal and lateral terminal segment 11) that differentiates it from all its congeners, including two undescribed morphotypes recently collected in the Mediterranean coast of Italy (Dal Zotto and Todaro, 2016).

Family Semnoderidae Remane, 1929
Genus *Sphenoderes* Higgins, 1969
Sphenoderes sp. (Fig. 6.6A):

Morphotype showing striking similarities (shared spine and sensory spot formula) with *Sphenoderes aspidochelone* Sørensen and Landers, 2017, which was collected from 80 to 300 m depth in the Gulf of Mexico (Sørensen and Landers, 2017). More detailed observations and, ideally, molecular data will be required to confirm whether our specimens belong to this species.

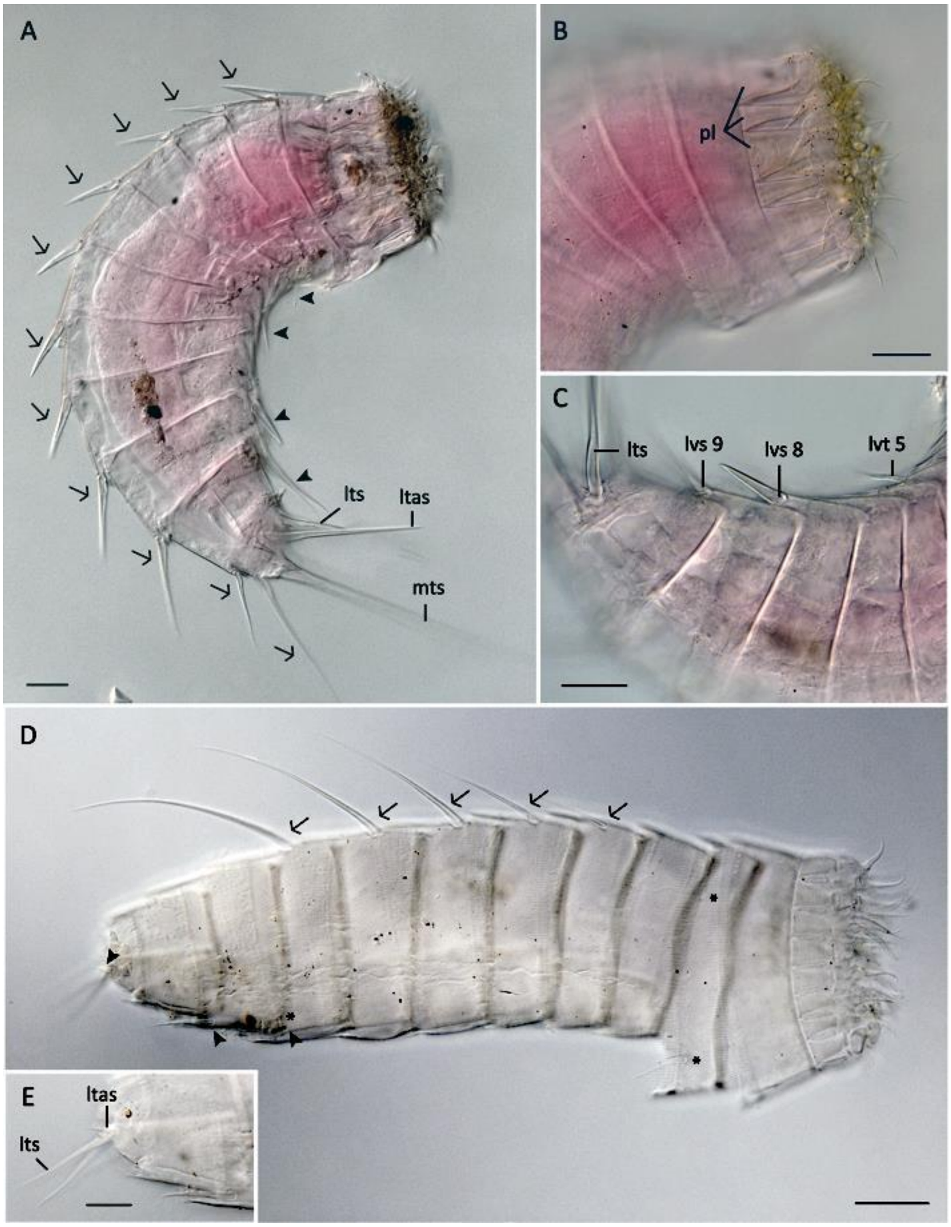


Figure 6.5. DIC micrographs. *Centroderes spinosus* (**A-C**): **A.** Lateral overview, dorsal side is left; **B.** Detail of neck and segments 1-4, lateral view; **C.** Details segments 5-11, lateral view. *Echinoderes lusitanicus* (**D-E**): **D**. Lateral overview, dorsal side is up; **E**. Detail segments 9-11, lateral view (arrows point on middorsal spines; arrowheads point onlateral ventral spines; asterisks point ontubes). lt: lateral terminal spine; ltas: lateral terminal accessory spine; lvs: lateroventral spine; lvt: lateroventral tube; mts: midterminal spine; pl: neck placids. Digits mark the segment number. Scale bar A-D: 20 μm**,** E: 10 μm**.**

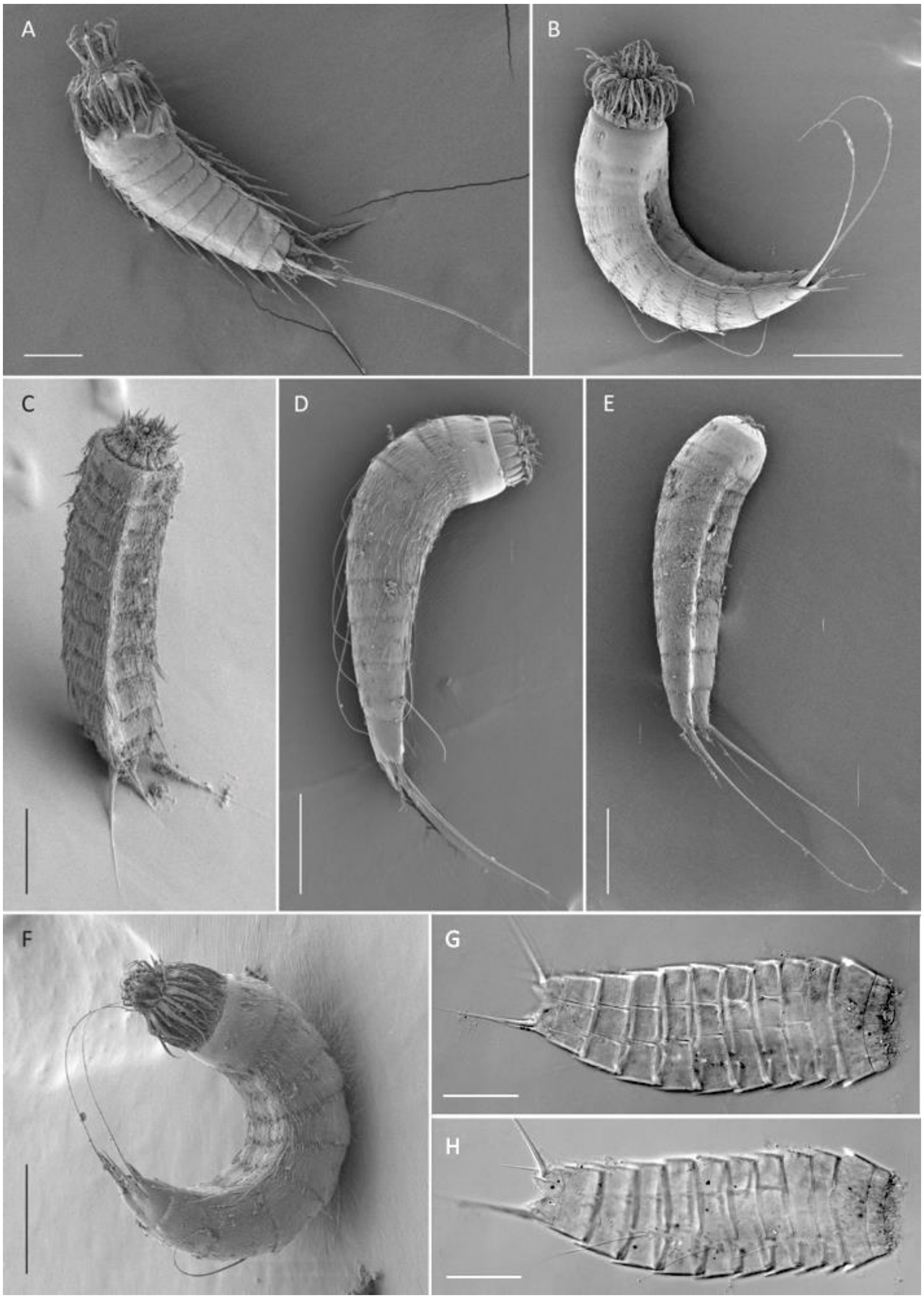


Figure 6.6. SEM micrographs: **A**. *Sphenoderes* sp., lateral view; **B**. *Echinoderes* sp.3, lateral view; **C.** *Condyloderes* sp., lateral view; **D.** *Echinoderes* sp.1, lateral view; **E**. *Echinoderes* sp.2, lateral view; **F**. *Echinoderes* sp.4, lateral view. DIC micrographs of *Echinoderes* sp.5 (**G-H**): **G**. Ventral view; **H**. Dorsal view. Scale bar: 50 μm.

6.3.4 Diversity and species distribution

The number of species decreased with increasing water depth both in the canyon and on the slope (Table 6.3, Fig. 6.7). All species occurred in the canyon, and BC1200 was the most diverse station (six species), followed by BC500 and BC900 (five species) (Figs. 6.5, 6.7). Only three species occurred on the slope (Fig. 6.7). Juveniles occurred in all stations, except for the deepest canyon one (BC2000, Table 6.3, Fig. 6.7).

The most speciose and abundant genus found in this study was *Echinoderes* Claparède, 1863 with six species, five of them currently undescribed (Table 6.3, Fig .6.5-6). The species of *Echinoderes* mainly occurred in the canyon, except for the two specimens of *Echinoderes.* sp. 1 found at OS1500. *Echinoderes* sp. 1 (101 specimens) and *Echinoderes.* sp. 2 (21 specimens) were found all along the canyon down to BC1750 (Fig. 6.7). *Echinoderes* sp. 3 and *Echinoderes* sp. 5 were found only once, respectively at BC900 and, BC1200, whereas *Echinoderes* sp. 4 (5 specimens) was found at the upper canyon stations, from BC500 to BC1200, and *Echinoderes lusitanicus* was found at BC500 and BC1500 (Fig. 6.7).

Centroderes spinosus was only recorded in the upper canyon (1 specimen at BC900). *Condyloderes* sp. (4 specimens) was present in the shallowest stations both in the canyon and slope (BC500, BC900 and OS500).

Sphenoderes sp. was recorded both in the canyon (4 specimens) and in the slope (8 specimens) and was present at mid and deep stations in the canyon, and in shallow and mid-depth stations in the slope (Fig. 6.7). Juveniles occurred in all samples, both in the slope and canyon (Table 6.3). In general, juvenile and adult abundance were paired, being both higher in the canyon and showing the highest abundances in shallower stations. Juveniles from slope stations were likely belonging to belong to *Sphenoderes* sp., *Condyloderes* sp. and *Echinoderes* sp.1.

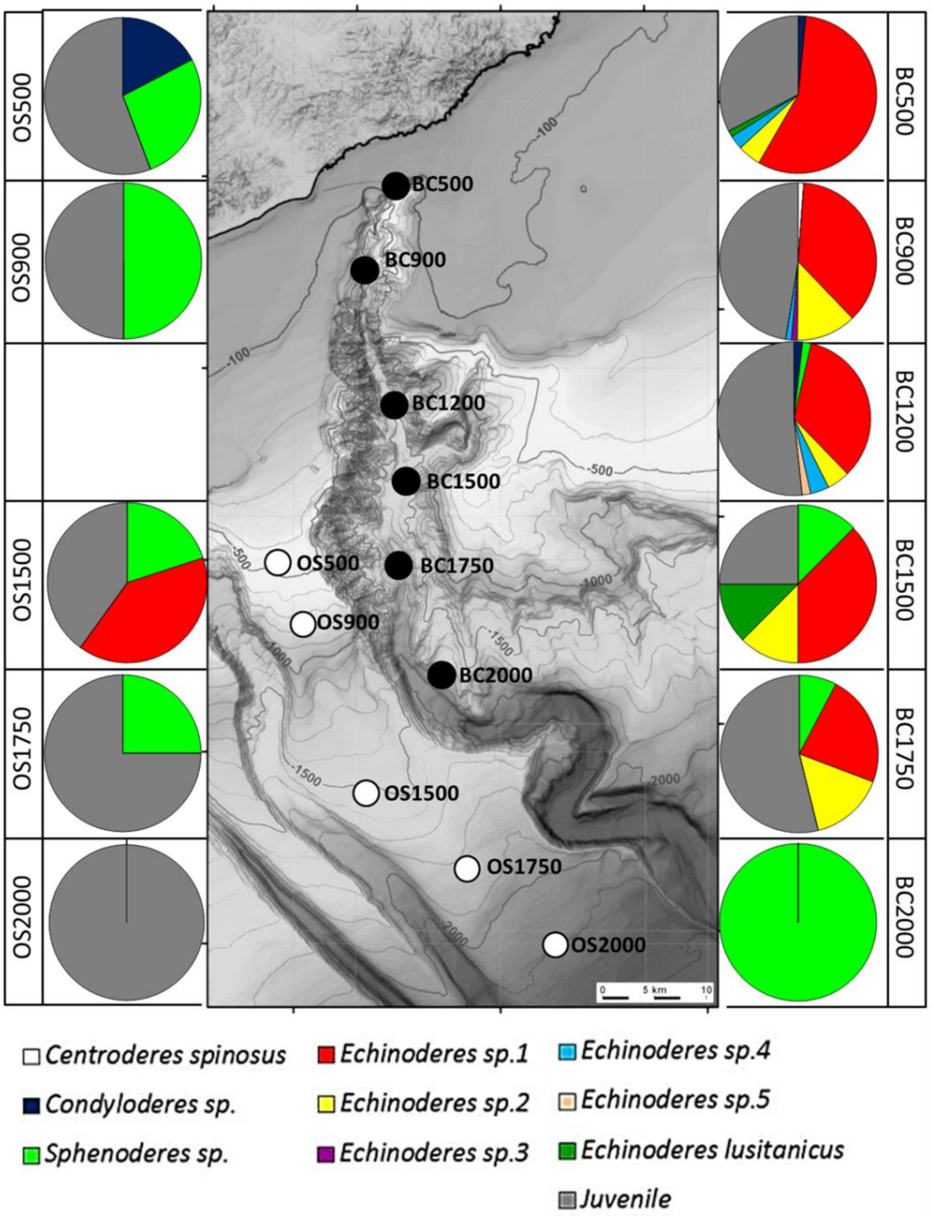


Figure 6.7. Relative abundance of the adult and juvenile kinorhynch in the Blanes Canyon and its adjacent western slope.

6.4 DISCUSSION

6.4.1 Spatial distribution

Our results show an inverse correlation between kinorhynch density and depth, both in the canyon and on the slope. Indeed, kinochrynchs are much denser in shallower depths (i.e., from 500 to 900/1,200 m). This pattern is congruent with previous studies in bathyal areas (Gutzmann et al., 2004), including shelf-slope (Soetaert et al., 1991a; Tietjen, 1971), slope (Ingels et al., 2009), and submarine canyons (Ingels et al., 2009, 2011a, b; Ramalho et al., 2014; Rosli et al., 2009; Soetaert et al., 1991a). The only exception occurred in the Atacama Trench (South Pacific Ocean), where densities increased considerably with depth, reaching a maximum of ca. 50 ind. $10cm^2$ at 7,800 m depth (Danovaro et al., 2002).

The decrease of meiofaunal density with increasing depth is a well-known pattern that has been correlated with a depth-related scarcity of potential food (e.g., phytodetritus) sinking down from the euphotic zone (Thiel, 1983, 1987; Tietjen, 1992; Danovaro et al 1995; Soltwedel, 2000; Pape et al., 2013; Lins et al., 2014).

In our study, we demonstrate that the relationship with depth, play an important role in shaping the kinorhynch distribution along Blanes Canyon and its adjacent open slope. This relationship, was clearly mediated by food availability, both in terms of quantity (Chl *a*, CPE, OC) and quality (Chl *a*: phaeo), as the densities showed positive relationships with all these food-related variables. Grain size characteristics of the sediment also influenced the slope assemblages. Particularly, there was a negative relationship between density and clay contents.

Densities and relative abundances were much higher in Blanes Canyon than on the slope, which is congruent with the overall higher meiofaunal densities and biomasses found in submarine canyons, compared to the nearby slopes (Ingels et al., 2009; Leduc et al., 2014; Rosli et al., 2016; Soetaert et al., 1991a, **Chapter 3** and **5**). This trend, mainly obeys to the distribution of the most dominant taxa, the nematodes. However, it may also apply to kinorhynchs, showed high numbers in canyons such as Nazaré (Bianchelli et al., 2010; Ingels et al., 2009), Cap de Creus (Bianchelli et al., 2010) and Calvi (Soetaert et al., 1991a), as well as in previous reports from the Blanes area (Romano et al., 2013b). Submarine canyons offer a complex habitat with specific hydrographic, sedimentological and geochemical characteristic (Flexas et al., 2008) act as major conduits for shelf sediment transport, and shows more intense bottom currents than the adjacent slopes

(Zuñiga et al., 2009; Lopez-Fernandez et al., 2013). These characteristics clearly influenced the associated faunal assemblages (e.g., **Chapter 5**; Ingels et al., 2009; Leduc et al., 2014) and kinorhynch were not and exception.

Overall, kinorhynchs are mostly found in the upper 2-3 centimetres in muddy sediments, and tend to decrease towards the deepest sediment layers in which nematodes become dominant (Kristensen and Higgins, 1991; Giere, 2009). Their presence and density may be influenced by the availability of oxygen, as they seem to be more sensitive to its low O_2 concentrations than nematodes (Grego et al., 2014; Kristensen and Higgins, 1991). This may explain the higher occurrences in surface layers, as well as the decreasing abundance as sediment compactness increases in deeper layers (as expressed by a corresponding high clay content; Table S1). These high densities in surface layers, and in the upper canyon (Table S1) may also be related to a higher food availability (Kristensen and Higgins, 1991; Shimanaga et al., 2000), expressed in terms of Chl *a*, CPE and OC. As water depth increases, sediment-mixing activity decreases (Soetaert et al., 1991a), leading to a reduced penetration of both OM and oxygen into the sediment. Therefore, sediment grain size and phaeopigment (Chl *a,* CPE) availability in the subsurface and deeper layers are likely the most influencing variables explaining the distribution of kinorhynchs in the Blanes Canyon system.

Our results also showed differences in the distribution along the vertical sediment profile. Kinorhynchs from the canyon were able to penetrate deeper into the sediment, compared with those from the slope. This might be related with the higher clay content of the slope. Moreover, Chl *a* was higher in the canyon axis than on the slope (Table S1). The high occurrences in subsurface and deeper layers have been related with the hydrodynamic regime typical of submarine canyons. Being more complex than that on the slope, it might affect the deposition and accumulation rates of sediment and organic matter. Similar sediment patterns were found by Ingels et al. (2009) in the Nazaré Canyon and its adjacent slope, where CPE and, particularly, Chl *a* reached deeper sediment layers (5 cm), while Chl *a* was only found in the first cm layer along the slope, a distribution that was mirrored by the associated kinorhynch assemblages. In addition, the high densities observed in Blanes Canyon in autumn 2012 and spring 2013 coincided with increased nutrient availability at the surface waters. In 2012 this increase was reflected in the increase of CPE reaching the canyon, through its axis and gullies after a cascading event, while in spring 2013, an increase of surface productivity ("spring bloom") in terms of Chl *a* concentrations was observed (see **Chapter 3**).

6.4.2 Diversity and species distribution

All specimens of kinorhynch found in the Blanes Canyon system belong to the Cyclorhagida, a diverse taxa including more than 150 species grouped in 17 genera. Our specimens belong to four genera (i.e., *Echinoderes*, *Centroderes*, *Condyloderes* and *Sphenoderes*). *Echinoderes* was represented by six taxa (one species and five undescribed morphotypes), which is not surprising because it is by far the most speciose genus of kinorhynchs, containing more than 30% of the group´s diversity (Neuhaus, 2013; Herranz et al., 2017). Nevertheless, there are no deep-sea species of *Echinoderes* described so far, although the genus was reported from 1,000-2,400 m depth near Costa Rica (Neuhaus and Lüter, 2003) and from 5,000 to 5,600 m depth, as "not examined *Echinoderes"* among the material collected during the German DIVA2 expedition in the Angola and Guinea Basins, Atlantic Ocean (Sørensen, 2008; Sánchez et al., 2014a, b).

Among all the species of *Echinoderes* found in our samples, only *Echinoderes lusitanicus* (Fig. 6D-E) was known to science. The species was recently described from the south Atlantic coast of Portugal, from muddy sediments at 100 m depth (Neves et al., 2016). Our specimens were collected at 900 and 1,500 m depth, while its absence at 1,200 m depth may be a sampling artefact. Our findings extend the known distribution of *E. lusitanicus* to the Mediterranean waters. The Mediterranean coast of the Iberian Peninsula has been intensively sampled for kinorhynchs during the last decade, including the Blanes area (summarized in Sánchez et al. 2012). However, most surveys were restricted to shallow waters (i.e., less than 100 m depth), which could explain why the species has not been previously found and may also indicate a preference for deeper waters.

The remaining *Echinoderes* specimens were undescribed morphotypes (*E*. sp.1 – *E*. sp.5). The most abundant was *E*. sp.1, which was present in all canyon stations except the deepest one (BC2000). *Echinoderes* sp.2 had a very similar distribution, but with lower abundances. Both taxa seem to be more affected by food availability than by depth. *Echinoderes* sp.2 has also been recorded in muddy sediments at 117 m depth in Naples (Italy) (M. Herranz pers. obs.). This may suggest that *E*. sp.2 has a broad distribution and therefore, we might expect to find it in other shallow and deep Mediterranean areas. Conversely, *Echinoderes* sp.3 to sp.5 showed low abundances and distributions restricted to the shallowest canyon samples (BC500 to BC1200). These species appear to be more sensitive to a combined oxygen and food availability, as well as to the presence of fine sediments. Therefore, we suggest that its rarity in our samples could be explained by a preference for areas shallower than those sampled in the Blanes Canyon system. This hypothesis seems to be supported by the finding of *Echinoderes* sp.5 from Naples (from 30 to 100 m depth) and in Faro, Portugal (muddy sediments at 90 to 100 m depth) (M. Herranz pers. obs.).

Centroderes spinosus is the second "previously known" species found in the present study, where, it was collected in a single locality (BC900) inside the canyon. The species is known to have a wide distribution, including the North Sea, the NW Atlantic Ocean, the Mediterranean Sea and the Black Sea, and has been collected from 14 to 444 m depth (Sanchez et al., 2012; Dal Zotto and Todaro 2016, see Neuhaus, 2013 for additional references). Our finding at 900 m depth increases its known depth range, confirming an occurrence not limited by depth. Our single record, however, does not allow us to hypothesize on the factors driving its presence in Blanes Canyon. However, based on the variety of sediments in which *C. spinosus* occurs (see Neuhaus 2013; Dal Zotto and Todaro, 2016; Sánchez et al. 2012), we could also discard sediment type as selecting factor. Overall, it seems that *C. spinosus* may be resilient species, able to thrive in different environmental conditions as reflected by its wide distribution.

The specimens of *Condyloderes* were found at different depths both in the canyon and slope shallowest stations (BC500, BC900 and OS500) (Table 4, Fig. 5C). Currently, *Condyloderes* accommodates six different species: *Condyloderes multispinosus* McIntyre, 1962; *C. paradoxus* Higgins, 1969; *C. setoensis* Adrianov et al., 2002; *C. storchi* Martorelli and Higgins, 2004; *C. megastigma* Sørensen et al., 2010 and *C. kurilensis* Adrianov and Maiorova, 2016. Among them, only the latter was recorded from abyssal depths (5,222 m). The specimens from the Blanes Canyon system are the second deep-sea report for the genus. However, the apparent preference of the genus for the deep sea might be a sampling artefact, as the existing studies are biased toward coastal shallow areas. In fact, one specimen of *Condyloderes* sp. has also been found in muddy sediments at 117 m depth off Naples (M. Herranz pers. obs.), which strongly suggest that it might not be restricted to deep waters. Moreover, two other undescribed morphotypes of *Condyloderes* have been previously reported, from Mediterranean shallow waters in the Thyrrhenian and

Ligurian seas (as *Condyloderes* sp.1 and sp.2 in Dal Zotto and Todaro, 2016). Both were collected in silty sediments ranging from 30 to 130 m depth, which agrees with the overall preference of *Condyloderes* for finer sediment.

The last undescribed morphotype found in the Blanes Canyon system belongs to the rare genus *Sphenoderes*, which accommodates only three species: *Sphenoderes indicus* Higgins, 1969; *S. poseidon* Sørensen, 2010 and the recently described *S. aspidochelone* Sørensen and Landers, 2017. All of them were collected in muddy areas of the Bay of Bengal, the Korea Strait and the Gulf of Mexico, respectively. Therefore, our report is the first for the genus in European waters and, more specifically, in the Mediterranean Sea. Interestingly, *Sphenoderes* sp. does not follow the general trends showed by the other kinorhynchs from Blanes Canyon. It was found in depth range, and was also present in both in canyon and slope sediments (Table 6.3), showing the highest abundance in the shallowest slope stations (OS500 and OS900). Therefore, contrary to most kinorhynch species, *Sphenoderes* sp. seems to be less affected by the lower amount of food sources and the higher clay contents characterizing the slope.

6.4.3 Previous studies in Blanes area

The previous studies on kinorhynchs from shallow waters off Blanes coast (ranging from 11 to 55 m depth) revealed a high diversity, with thirteen species nested in six different genera: *Echinoderes*, *Centroderes*, *Semnoderes*, *Antygomonas*, *Meristoderes* and *Pycnophyes* (summarized in Sánchez et al., 2012) (Table 6.4). Of them, *Echinoderes* and *Centroderes* were the most common. However, only *C. spinosus* was coincident with the present study.

This result is demonstrating how dissimilar the kinorhynch communities can be at a very local scale, but also underscoring the influence of the canyon to the whole meiobenthic ecosystem as reported previously (Romano et al., 2013b; **Chapter 3**). Despite the proximity of these sampled shallow areas to the canyon head (c.a. 1-2 km), the depth ranges were quite different, (11-55 m vs. 500 m depth of the shallowest station in our study). This highlights the abrupt modification of the seafloor topography and dynamics caused by the presence of the canyon (Ingels et al. 2013). The enhanced erosion-accumulation processes typical of a canyon, combined with the distance from land, cause modifications in sediment composition, which tends to change from sandier

Table 6.4. Kinorhynch diversity and bathymetric distribution off Blanes coast.

Species	Depth (m)										
	Sánchez et al. (2012)					Present study					
	11	28.4	30	37.5	55.4	500	900	1200	1500	1750	2000
*Echinoderes hispanicus*Pardos et al., 1998		■	■	■							
Echinoderes sp			■	■							
Echinoderes isabelae Gª Ordonez et al., 2008			■	■							
Semnoderes armiger Zelinka, 1928					■						
Meistoderes macracanthus Herranz et al. 2012	■	■	■	■							
Pycnophyes aulacodes Sánchez et al., 2011				■							
Pycnophyes carinatus Zelinka, 1928				■							
Pycnophyes communis Zelinka, 1928				■							
Pycnophyes dentatus (Reinhard, 1881)		■		■							
Pycnophyes zelinkaei Southerns, 1914			■		■						
Pycnophyes sp.3		■		■							
Antygomonas incomitata Nebelsick, 1990	■		■								
Centroderes spinosus (Reinhard, 1881) Zelinka, 1928					■		■				
Condyloderes sp.						■		■			
Sphenoderes sp.						■	■	■	■	■	■
Echinoderes sp.1						■	■	■	■	■	
Echinoderes sp.2						■	■	■	■	■	
Echinoderes sp.3							■				
Echinoderes sp.4						■	■	■			
Echinoderes sp.5								■			
Echinoderes lusitanicus (Neves, Sørensen & Herranz, 2016)						■			■		

(in shallower areas) to muddier (in deeper areas), most likely altering the kinorhynch communities. Therefore, some species reported from shallow waters (highly associated with sandy sediments) are logically absent from the canyon. This seems to be the case of *Antygomonas incomitata*, Nebelsick 1990 and *Meristoderes macracanthus* Herranz et al. 2012 (Herranz et al. 2012; Nebelsick 1990; Sánchez et al., 2012), which also occur along the coasts of Italy (Dal Zotto and Todaro, 2016). Thus, we suggest that they may show a wide Mediterranean distribution, although restricted to shallow sandy areas.

The lack of some relatively abundant and wide-spread Mediterranean species in Blanes Canyon, such as *Semnoderes armiger* (considered one of the most widely distributed kinorhynchs in European waters) might be resulting from undersampling. Factors such as an increased structural heterogeneity, the complex hydrodynamic patterns, and the downward food transport, could likely explain the divergence between the communities from the canyon and the nearby shallow open areas. However, we certainly expected some of the dominant species from the canyon to occur in the surrounding shallower areas and *vice versa*, as supported by the presence of *E.* sp.2 and *E.* sp.5 in shallow depths off Naples and Faro (M. Herranz per. obs.) and in line with the hypothesized sampling bias. Therefore, we may expect the diversity of the kinorhynch communities from the Blanes coasts, both from shallow waters and, particularly, from deep bottoms in and around the canyon, to increase as a result of an intensive sampling effort, which emerges an objective for future studies.

6.5 SUPPLEMENTARY DATA

Table S6.1. Average values of all environmental variables per station and sediment layer.

Station	Sediment depth (cm)	Clay (%)	Silt (%)	Sand (%)	TN (%)	OC (%)	Chl *a* (µg/g)	CPE (µg/g)	Chl *a*: phaeo
BC500	0-1	20.78	71.37	7.84	0.10	0.96	0.86	12.31	0.08
	1-2	20.29	68.20	11.52	0.10	0.97	0.63	7.21	0.10
	2-5	19.45	68.42	12.13	0.10	1.02	0.37	6.71	0.07
BC900	0-1	19.28	73.38	7.35	0.09	0.84	0.07	1.56	0.06
	1-2	18.57	72.35	9.07	0.09	0.82	0.03	0.89	0.03
	2-5	18.68	70.65	10.67	0.09	0.80	0.02	0.84	0.02
BC1200	0-1	18.27	67.30	14.43	0.09	0.76	0.10	1.84	0.05
	1-2	16.87	66.42	16..71	0.08	0.74	0.04	1.35	0.05
	2-5	16.36	65.11	18.54	0.08	0.77	0.03	0.97	0.05
BC1500	0-1	16.37	69.92	13.71	0.08	0.76	0.05	0.98	0.05
	1-2	17.70	73.50	8.79	0.08	0.77	0.03	0.82	0.04
	2-5	17.46	69.83	12.71	0.09	0.78	0.02	0.61	0.04
BC1750	0-1	19.28	75.60	5.12	0.09	0.82	0.05	1.18	0.04
	1-2	17.93	74.96	7.12	0.09	0.81	0.03	0.68	0.04
	2-5	19.78	72.69	7.53	0.09	0.80	0.01	0.51	0.02
BC2000	0-1	19.17	75.04	5.80	0.09	0.77	0.54	0.58	0.03
	1-2	18.90	75.33	5.77	0.09	0.76	0.22	0.25	0.01
	2-5	21.96	70.78	7.26	0.09	0.78	0.18	0.20	0.01
OS500	0-1	14.83	65.23	19.94	0.07	0.73	0.13	0.10	0.11
	1-2	14.49	65.03	20.48	0.07	0.64	0.05	0.06	0.06
	2-5	13.74	58.40	27.86	0.06	0.62	0.06	0.11	0.10
OS900	0-1	17.13	80.68	2.19	0.10	1.04	0.05	0.04	0.04
	1-2	17.30	80.67	2.03	0.09	0.86	0.01	0.02	0.02
	2-5	18.29	79.53	2.18	0.09	0.79	0.00	0.01	0.01
OS1500	0-1	19.99	74.65	5.36	0.08	0.76	0.01	0.01	0.01
	1-2	22.42	71.53	6.05	0.08	0.69	0.01	0.04	0.03
	2-5	26.78	69.17	4.05	0.08	0.62	0.00	0.02	0.01
OS1750	0-1	21.40	72.28	6.32	0.08	0.67	0.01	0.02	0.02
	1-2	25.28	69.66	5.05	0.08	0.60	0.00	0.02	0.03
	2-5	26.70	68.47	4.83	0.07	0.54	0.00	0.01	0.04
OS2000	0-1	20.57	69.13	10.30	0.10	0.72	0.00	0.01	0.01
	1-2	22.48	70.16	7.37	0.09	0.66	0.00	0.01	0.02
	2-5	25.37	69.38	5.24	0.08	0.53	0.00	0.00	0.00

7

General Conclusions

General conclusions and future directions

The results of this Doctoral Thesis provide new insights on the deep-sea metazoan meiofauna living in submarine canyons, being one of the most complete studies on this topic that has been carried out in the Mediterranean deep continental margin. The aims of this research were to better understand the standing stocks, diversity and structure of meiobenthic communities, with particular emphasis on the nematodes, in relation with the main environmental factors driving the functioning of the Blanes Canyon system. The different meiofaunal descriptors analysed (i.e., density, biomass, diversity, structure and feeding types) prove that Blanes Canyon is highly heterogeneous, and confirm that this canyon system is an important density/biomass and biodiversity *hot spots*. The dynamics of water and sediment transport, the morphology of the canyon and the inherent environmental factors of the canyon play an important role in controlling the meiobenthos. However, our results also suggest the relevance of non-"natural" drivers (i.e. of anthropogenic origin) in explaining the canyon variability. Blanes Canyon is a particularly important fishing area. Despite the studied stations were the canyon axis (i.e., far from the areas were fish trawling activities are regularly carried out), we could identify their indirect effects on the sediment variables and, ultimately, on the descriptors of the meiobenthic communities.

The present Thesis also includes the first known study on Mediterranean deep-sea kinorhynchs. This study contributes to enlarge the known distribution of *Centroderes spinosus* and *Echinoderes lusitanicus* and increases the current knowledge on kinorhynch diversity, as seven over the nine species found in the canyon turned to be undescribed.

During the research performed within the framework of this Thesis, several constraints affected the normal development of the initially planned sampling strategy, which it is not unusual for deep-sea research. Among them, the limited ship time and technical problems during deployment of the sampling gear which resulted in insufficient replicates or, even, originally planned stations that were finally not sampled, particularly on the slope. Moreover, for obvious reasons the moorings (including different oceanographic sensors and sediment traps) deployed during the DOS MARES project, were placed in a spatial network that differed from that of the benthic sampling. Therefore, there was a limited knowledge on the hydrodynamic and sediment flux characteristics during the study periods at each sampling station. Despite this limitation, the obtained results highly improve the knowledge of the processes controlling the spatial distribution of meiofaunal standing stocks, structure and biodiversity in Blanes Canyon and its adjacent slope, at different spatial scales (i.e., large (km); small (cm) scales), one of the main objectives of this Thesis.

The results of the studies carried out during this Doctoral research can be summarized in the following conclusions:

1) Within the framework of different multidisciplinary research project (RECS, PROMETEO and DOSMARES) carried out in the Blanes Canyon system, it was shown that the sedimentary environments within and nearby Blanes Canyon are subjected to numerous and heterogeneous processes occurring with distinct frequencies and intensities over time, which lead to cyclic episodes of deposition, resuspension and transport. In addition to this natural variability, the influence of anthropogenic activities, particularly those resulting from the intensive bottom trawling fisheries reveals to be a key factor affecting both geological and biological processes in the canyon system. The result of the studies carried out in the Thesis demonstrated that both the natural and anthropogenic sources of environmental variability are driving the meiofaunal communities.

2) The observed temporal variability on meiofauna in the canyon is only partly explained by the seasonal patterns of food input, derived from phytoplanctonic production, where major oceanographic processes, such as recurrent dense shelf water cascading, seems to play a key role. Moreover, our results based on nematode communities pointed out that the topographical heterogeneous environment associated to the canyon bathymetric, combined with the existence of recurrent, non-seasonal food pulses, are better explaining the observed meiofaunal trends, than seasonal variation of food inputs derived from phytoplanktonic production.

3) Our results confirm that Blanes Canyon exhibits: marked sediment variability, high food availability (i.e., OC, Chl *a*, CPE) compared to the adjacent slope, together with higher density, and diversity, and more marked differences in community composition and distribution, both at higher (i.e., meiofauna) and lower (i.e., nematodes and kinorhynchs) taxonomic level. Our findings additionally support the consideration of submarine canyons as *hotspots* of faunal density, biomass and diversity, and confirm that they play an important role in shaping the patterns and trends shown by benthic fauna.

4) The analysis of the meiofauna spatial distribution proves the existence of clear differences between canyon and slope, with the standing stocks showing a marked bathymetric gradient on the slope and disruption in this gradient along the canyon. The lateral advection and accumulation of food-enriched shallow-water sediments, resuspended as a result of bottom trawling activities along the canyon flanks, then flooding downwards the canyon through lateral gullies, seems to be cause of the increases in meiofauna density and diversity detected at 900 m depth and, particularly, at 1,200 m depth.

5) The nematode communities also mirrored the trends observed in the total meiofauna in lacking a consistent bathymetric gradient in standing stocks and diversity along the canyon axis. The standing stocks peaked at 1,200 and 1,750 m depth, mainly as a result of the overall high nematode densities in the surface sediment layer (i.e. 0 - 1 cm), while such a general trend did not occur for the biomass

6) The analysis of the nematode community structure allowed us to identify three different faunal zones along the canyon, which were 1) supported by the environmental descriptors, 2) followed the canyon topographic heterogeneity, and 3) responded to the food inputs from canyon walls and adjacent margins.

7) As it occurred for the whole meiofauna, the nematode communities from Blanes Canyon revealed to be not only controlled by the sedimentary characteristics and available food sources, but also by the canyon topography and hydrodynamic regime. In the specific case of the Blanes Canyon, moreover, the anthropogenic pressure derived specifically from fish trawling activities in the canyon system surroundings, play also a key role in explaining the meiobenthic distribution patterns.

8) Among all spatial gradients analysed in canyon and slope environments, the greatest effect on nematode community structure and structural diversity were related with the small-scale (cm), especially, by the highest numbers at the surface layer leading to a marked tendency of diversity to decrease along the sediment profile. Trophic conditions in the sediment, as well as the associated biochemical gradients (e.g. oxygen availability), seemed to have a greater influence on nematode community structure, especially in the highly heterogeneous canyon habitats. Moreover, different genera display a different ability to reside (or penetrate into) the different sediment layers, as illustrated by the high abundance of *Sabatieria* in the deepest layers.

9) The presence of chemosynthetic nematodes of the genus *Astomonema* in the canyon (BC500-BC1750), especially at deeper layers, points to the presence of the hydrogen sulphides by the lack of oxygen in the sediments. Due to high burial rates in the canyon sediments compared to the slope, oxygen levels may quickly drop a few centimetres below the sediment surface as high amounts of organic matter are being degraded. So, the identification of nematodes provided useful information on biochemical conditions such as sulphate reduction.

10) The effect of bottom trawling on meiofaunal assemblages and, more specifically, on the nematodes, were detected in the canyon axis until 1,750 m depth. The high sedimentation rates observed suggest that trawling activities should be considered as a major large- and small-scale driver in the Blanes Canyon. In fact, our results support that the high variability in nematode communities at small-scale (i.e. along the vertical sediment profile) lead them to be highly vulnerable to trawling induced disturbances altering sedimentation rates. Down to the deeper canyon region, at 2,000 m depth the trawling influence almost disappeared and the actual sedimentation rates did not differ from the natural ones. This explained that, both the sediment characteristics and the nematode assemblage descriptors are more similar to those of the nearby slope.

11) The less abundant and more dispersed kinorhynchs showed a contrasting pattern. Both the canyon and the slope showed a general decrease of density and diversity with increasing water depth, likely related to food availability. However, there were clear differences between canyon and slope communities in terms of density and diversity, higher each time values in the canyon. Also, the distribution along the vertical profile was different, with higher numbers in subsurface (1-2 cm) and deep (2-5 cm) layers in the canyon, likely as a response of the higher food sources and lower compactness in canyon sediments.

12) Our results allowed us to avoid suggesting generalizations in the description of meiofauna patterns in the canyon, highlighting the fact that different taxa may show different bathymetric-related responses, this being particularly relevant in the case of the rare taxa (e.g., the kinorhynchs).

13) The overall results of this Thesis strongly suggest that the Blanes Canyon system is not a purely natural environment. Not only the canyon flanks have been long-time used as fishing grounds (to the extreme that their topography may has been softened as it also occurred in the nearby La Fonera Canyon) but also because the indirect effects appear to be regularly driving the structure and possibly also the functioning of the associated benthic communities, including meiofauna, all along the canyon axis until, at least 1,750 m depth.

The results of this Thesis constitute a step forward in our knowledge on submarine canyon ecosystems. In particular, our research provided novel data on faunal distribution patterns, specifically dealing with structure, abundance/biomass, and biodiversity of canyon associated communities. Conservation of deep-sea environments, such as canyons, requires improving our understanding on the functioning of these ecosystems, as well on their biological and ecological role, and the, threats to which they are subjected. By identifying the factors mostly influencing the observed patterns, we will be able to provide essential tools to develop and implement sustainable policies in the exploitation of deep-sea biological resources. This certainly offers a wide field for future research, especially in Blanes Canyon that is currently providing great human ecological services. Future studies would have to consider specific analyses of the potential impacts derived from trawling activities on meiofaunal communities (in terms of structure, standing stocks and biodiversity), including trawled and non-trawled areas on the canyon flanks but also indirectly affected areas inside the canyon axis, in parallel with analyses of the sedimentation rates as tracers of the actual sediment dynamics.

Future studies must also consider to obtain better insights on biodiversity and taxonomy, especially by addressing specific taxa at the species level, to support the above-mentioned qualification of Blanes Canyon as *hot spot* of diversity, but also to be able provide more precise information on deep-sea species distribution and biodiversity estimates. A future approach combining morphological and molecular (e.g. DNA barcoding) methods, may also be a promising solution to unravel the biodiversity estimates. This is particularly relevant for highly abundant and diverse groups like nematodes, in which classical taxonomic identifications are highly complex and particularly time-costly. However such an approach, may also contribute to increase the species libraries of less well represented, but not less interesting, taxa such as (in the particular case of our study at Blanes Canyon), the kinorhynchs.

Last but not least, the postulated role of meiofaunal communities as a resource for higher tropic levels leads us to consider the carbon flow along the benthic food web in the canyon as another major topic to be elucidated. Techniques like isotope analyses (as an approach to trophic dynamics) or the analyses of fatty acids would be very helpful in this matter. Such an approach would include the transfer from lower levels (i.e. bacteria) to meiofauna and from this compartment to macro- and megafauna (including species of high commercial interest such as the red shrimp *Aristeus antennatus*). Accordingly, this would allow to understand the full relevance of canyon ecosystems in the oceans' functioning and, particularly, to improve the actual knowledge on the roles of the different meiofaunal taxa in deep-sea ecosystems.

References

Adrianov, A., Maiorova, A., 2016. Condyloderes kurilensis sp. nov.(Kinorhyncha: Cyclorhagida)—a new deep water species from the abyssal plain near the Kuril-Kamchatka Trench. Russian Journal of Marine Biology 42, 11-19.

Adrianov, A., Murakami, C., Shirayama, Y., 2002. Taxonomic study of the Kinorhyncha in Japan. II. Condyloderes setoensis, a new species (Kinorhyncha. Proceedings of the Biological Society of Washington 115, 205-216.

Aguzzi, J., Company, J., Bahamon, N., Flexas, M., Tecchio, S., Fernandez-Arcaya, U., Garcia, J., Mecho, A., Koenig, S., Canals, M., 2013. Seasonal bathymetric migrations of deep-sea fishes and decapod crustaceans in the NW Mediterranean Sea. Progress in Oceanography 118, 210-221.

Ahumada-Sempoal, M.-A., Flexas, M. d. M., Bernardello, R., Bahamon, N., Cruzado, A., 2013. Northern Current variability and its impact on the Blanes Canyon circulation: A numerical study. Progress in Oceanography 118, 61-70.

Almeida, M., Frutos, I., Company, J. B., Martin, D., Romano, C., Cunha, M. R., 2017. Biodiversity of suprabenthic peracarid assemblages from the Blanes Canyon region (NW Mediterranean Sea) in relation to natural disturbance and trawling pressure. Deep Sea Research Part II: Topical Studies in Oceanography 137, 390-403.

Allen, S., Durrieu de Madron, X., 2009. A review of the role of submarine canyons in deep-ocean exchange with the shelf. Ocean Science 5, 607-620.

Aller, J. Y., 1997. Benthic community response to temporal and spatial gradients in physical disturbance within a deep-sea western boundary region. Deep Sea Research Part I: Oceanographic Research Papers 44, 39-69, doi:Doi: 10.1016/s0967-0637(96)00092-1.

Amaro, T., Amaro, T., Allcock, A. L., Aslam, T., Davies, J. S., De Stigter, H. C., Danovaro, R., De Stigter, H. C., Howell, K. L., Gambi, C., Gooday, A. J., Gunton, L. M., Hall, R., Howell, K. L., Ingels, J., Kiriakoulakis, K., Kershaw, C. E., Wilson, A. M., Robert, K., Stewart, H., Van Rooij, D., White, M., Wilson, A. M., 2016. The Whittard Canyon – A case study of submarine canyon processes. Progress in oceanography 146, 38-57.

Amblas, D., Canals, M., Urgeles, R., Lastras, G., Liquete, C., Hughes Clarke, J., Casamor, J., Calafat, A., 2006. Morphogenetic mesoscale analysis of the northeastern Iberian margin, NW Mediterranean Basin. Marine geology 234, 3-20.

Anderson, M., Gorley, R. N., Clarke, R. K., 2008. Permanova+ for Primer: Guide to Software and Statisticl Methods.

Anderson, M. J., 2005. Permutational multivariate analysis of variance. Department of Statistics, University of Auckland, Auckland 26, 32-46.

Andrassy, I., 1956. The determination of volume and weight of nematodes. Acta Zoologica (Hungarian Academy of Science) 2, 1-15.

Azam, F., Fenchel, T., Field, J., Gray, J., Meyer-Reil, L., Thingstad, F., 1983. The ecological role of water-column microbes in the sea. Marine ecology progress series. Oldendorf 10, 257-263.

Baker, K. D., Wareham, V. E., Snelgrove, P. V., Haedrich, R. L., Fifield, D. A., Edinger, E. N., Gilkinson, K. D., 2011. Distributional patterns of deep-sea coral assemblages in three submarine canyons off Newfoundland, Canada. Marine Ecology Progress Series 445, 235-249.

Bauer-Nebelsick, M., 1996. Antygomonas oreas sp. n., a new deep sea kinorhynch from the Pacific Ocean (Kinorhyncha: Cyclorhagida). Annalen des Naturhistorischen Museums in Wien. Serie B für Botanik und Zoologie, 5-22.

Benedetti-Cecchi, L., 2001. Variability in abundance of algae and invertebrates at different spatial scales on rocky sea shores. Marine Ecology Progress Series 215, 79-92.

Bergamasco, A., Malanotte-Rizzoli, P., 2010. The circulation of the Mediterranean Sea: a historical review of experimental investigations. Advances in Oceanography and Limnology. 1, 11-28.

Bessette, S., Fagervold, S. K., Romano, C., Martin, D., Le Bris, N., Galand, P. E., 2014. Diversity of bacterial communities on sunken woods in the Mediterranean Sea. Journal of Marine Science and Technology 22, 60-66.

Béthoux, J. P., Morin, P., Ruiz-Pino, D. P., 2002. Temporal trends in nutrient ratios: chemical evidence of Mediterranean ecosystem changes driven by human activity. Deep Sea Research Part II: Topical Studies in Oceanography 49, 2007-2016.

Bianchelli, S., Gambi, C., Pusceddu, A., Danovaro, R., 2008. Trophic conditions and meiofaunal assemblages in the Bari Canyon and the adjacent open slope (Adriatic Sea). Chemistry in ecology 24, 101-109.

Bianchelli, S., Gambi, C., Zeppilli, D., Danovaro, R., 2010. Metazoan meiofauna in deep-sea canyons and adjacent open slopes: A large-scale comparison with focus on the rare taxa. Deep Sea Research Part I: Oceanographic Research Papers 57, 420-433.

Bianchelli, S., Gambi, C., Mea, M., Pusceddu, A., Danovaro, R., 2013. Nematode diversity patterns at different spatial scales in bathyal sediments of the Mediterranean Sea. Biogeosciences 10, 5465-5479.

Billett, D. S. M., Lampitt, R. S., Rice, A. L., Mantoura, R. F. C., 1983. Seasonal sedimentation of phytoplankton to the deep-sea benthos. Nature 302, 520-522.

Bonnin, J., Heussner, S., Calafat, A., Fabres, J., Palanques, A., 2008. Comparison of horizontal and downward particle fluxes across canyons of the Gulf of

Lions (NW Mediterranean): Meteorological and hydrodynamical forcing. Continental Shelf Research 28, 1957-1970.

Braeckman, U., Van Colen, C., Soetaert, K., Vincx, M., Vanaverbeke, J., 2011. Contrasting macrobenthic activities differentially affect nematode density and diversity in a shallow subtidal marine sediment. Marine Ecology Progress Series 422, 179-191.

Buchaca, T., Catalan, J., 2008. On the contribution of phytoplankton and benthic biofilms to the sediment record of marker pigments in high mountain lakes. Journal of Paleolimnology 40, 369-383.

Buhl-Mortensen, L., Vanreusel, A., Gooday, A. J., Levin, L. A., Priede, I. G., Buhl-Mortensen, P., Gheerardyn, H., King, N. J., Raes, M., 2010. Biological structures as a source of habitat heterogeneity and biodiversity on the deep ocean margins. Marine Ecology 31, 21-50.

Canals, M., Casamor, J.L., Urgeles, R., Farrán, M., Calafat, A.M., Amblás, D., Willmott, V., Estrada, F., Sánchez-Vida, l.A., Arnau, P., Frigola, J., Colás, S., 2004. Mapa del relleu submarí de Catalunya (Seafloor relief map of Catalonia), 1:250.000. Institut Cartogràfic de Catalunya, 1 map.

Canals, M., Puig, P., de Madron, X., Heussner, S., Palanques, A., Fabres, J., 2006. Flushing submarine canyons. Nature 444, 354-7.

Canals, M., Company, J., Martín, D., Sanchez-Vidal, A., Ramirez-Llodra, E., 2013. Integrated study of Mediterranean deep canyons: novel results and future challenges. Elsevier.

Carman, K. R., Sherman, K. M., Thistle, D., 1987. Evidence that sediment type influences the horizontal and vertical distribution of nematodes at a deep-sea site. Deep Sea Research Part A. Oceanographic Research Papers 34, 45-53.

Cartes, J., Company, J., Maynou, F., 1994. Deep-water decapod crustacean communities in the Northwestern Mediterranean: influence of submarine canyons and season. Marine Biology 120, 221-229.

Cartes, J., Fanelli, E., Papiol, V., Maynou, F., 2010. Trophic relationships at intrannual spatial and temporal scales of macro and megafauna around a submarine canyon off the Catalonian coast (western Mediterranean). Journal of Sea Research 63, 180-190.

Cartes, J. E., Maynou, F., Fanelli, E., Romano, C., Mamouridis, V., Papiol, V., 2009. The distribution of megabenthic, invertebrate epifauna in the Balearic Basin (western Mediterranean) between 400 and 2300 m: Environmental gradients influencing assemblages composition and biomass trends. Journal of Sea Research 61, 244-257.

Castro-Jiménez, J., Rotllant, G., Ábalos, M., Parera, J., Dachs, J., Company, J. B., Calafat, A., Abad, E., 2013. Accumulation of dioxins in deep-sea crustaceans, fish and sediments from a submarine canyon (NW Mediterranean). Progress in Oceanography 118, 260-272.

Claparède, A., 1863. Zur Kenntnis der Gattung Echinoderes Duj. Beobachtungen über Anatomie und Entwicklungsgeschichte wirbelloser Thiere an der Küste von Normandie angestellt. Leipzig: Verlag von Wilhelm Engelmann.

Clarke, K. R., Gorley, R. N., 2006. PRIMER V6: user manual-tutorial. Plymouth Marine Laboratory.

Company, J. B., Puig, P., Sardà, F., Palanques, A., Latasa, M., Scharek, R., 2008. Climate Influence on Deep Sea Populations. PLoS ONE 3, e1431.

Company J.B., Ramirez-Llodra E., Sardà F., Puig P., Canals M., Calafat A., Palanques A., Solé M., Sanchez-Vidal A., Martín J., Aguzzi J., Lastras G., Tecchio S., Koenig S., Fernandez- Arcaya U., Mechó A, Fernández P., 2012. Submarine canyons in the Catalan Sea (NW Mediterranean): megafaunal biodiversity patterns and anthropogenic threats Mediterranean Submarine Canyons: Ecology and Governance. In: (Ed.) WM (ed). IUCN, Gland (Switzerland) and Malaga (Spain).

Coull, B. C., Ellison, R. L., Fleeger, J. W., Higgins, R. P., Hope, W. D., Hummon, W. D., Rieger, R. M., Sterrer, W. E., Thiel, H., Tietjen, J. H., 1977. Quantitative estimates of the meiofauna from the deep sea off North Carolina, USA. Marine Biology 39, 233-240.

Cunha, M. R., Paterson, G. L. J., Amaro, T., Blackbird, S., de Stigter, H. C., Ferreira, C., Glover, A., Hilário, A., Kiriakoulakis, K., Neal, L., Ravara, A. A., Rodrigues, C. F., Tiago, Á., Billett, D. S. M., 2011. Biodiversity of macrofaunal assemblages from three Portuguese submarine canyons (NE Atlantic). Deep Sea Research Part II: Topical Studies in Oceanography 58, 2433-2447, doi:10.1016/j.dsr2.2011.04.007.

Curdia, J., Carvalho, S., Ravara, A., Gage, J., Rodrigues, A., Quintino, V., 2004. Deep macrobenthic communities from Nazaré submarine canyon (NW Portugal).

d'Onghia, G., Politou, C. Y., Bozzano, A., Lloris, D., Rotllant, G., Sion, L., Mastrototaro, F., 2004. Deep-water fish assemblages in the Mediterranean Sea. Scientia Marina 68, 87-99.

Dal Zotto, M., Todaro, M. A., 2016. Kinorhyncha from Italy, a revision of the current checklist and an account of the recent investigations. Zoologischer Anzeiger-A Journal of Comparative Zoology 265, 90-107.

Dana, J., 1863. Manual of geology: Philadelphia, 798 p. Virginia and North by WC Mansfield: Virginia and North Prof. Paper, 199-8.

Danovaro, R., 1995. Meiofauna of the deep Eastern Mediterranean Sea: distribution and abundance in relation to bacterial biomass, organic matter composition and other environmental factors. Progress In Oceanography 36, 329-341.

Danovaro, R., 2009. α-, β-, γ-, δ-and ε-diversity of deep-sea nematodes in canyons and open slopes of northeast Atlantic and Mediterranean margins. Marine Ecology Progress Series 396, 197-209.

Danovaro, R., Gambi, C., Della Croce, N., 2002. Meiofauna hotspot in the Atacama Trench, eastern South Pacific Ocean. Deep-sea research. Part 1. Oceanographic research papers 49, 843-857.

Danovaro, R., Dinet, A., Duineveld, G., Tselepides, A., 1999. Benthic response to particulate fluxes in different trophic environments: a comparison between the Gulf of Lions–Catalan Sea (western-Mediterranean) and the Cretan Sea (eastern-Mediterranean). Progress in Oceanography 44, 287-312.

Danovaro, R., Gambi, C., Lampadariou, N., Tselepides, A., 2008. Deep-sea nematode biodiversity in the Mediterranean basin: Testing for longitudinal, bathymetric and energetic gradients. Ecography 31, 231-244.

Danovaro, R., Batista Company, J., Corinaldesi, C., D'Onghia, G., Galil, B., Company, J., Gambi, C., Gooday, A., Lampadariou, N., Luna, G., Morigi, C., Olu, K., Polymenakou, P., Ramirez Llodra, E., Sabbatini, A., Sard, F., Sibuet, M., Tselepides, A., Gratwicke, B., 2010. Deep-Sea Biodiversity in the Mediterranean Sea: The Known, the Unknown, and the Unknowable. PLoS ONE 5, e11832.

Danovaro, R., Carugati, L., Corinaldesi, C., Gambi, C., Guilini, K., Pusceddu, A., Vanreusel, A., 2013. Multiple spatial scale analyses provide new clues on patterns and drivers of deep-sea nematode diversity. Deep-sea research. Part 2, Topical studies in oceanography 92, 97-106.

Decraemer W., Coomans A., Baldwin J., 2013. Morphology of Nematoda. In: Schmidt Rhaesa A (ed.) Handbook of zoology: Gastrotricha, Cycloneuralia ad Gnatifera, vol. 2: Nematoda. P. 1-59.

De Grisse, A. T., 1969. Redescription ou modifications de quelques technique utilis [a] es dan l'etude des n [a] ematodes phytoparasitaires.

De Leo, F., Smith, C., Rowden, A., Bowden, D., Clark, M., 2010. Submarine canyons: hotspots of benthic biomass and productivity in the deep sea. Proceedings - Royal Society. Biological sciences 277, 2783-92.

De Leo, F., Vetter, E., Smith, C., Rowden, A., McGranaghan, M., 2014. Spatial scale-dependent habitat heterogeneity influences submarine canyon macrofaunal abundance and diversity off the Main and Northwest Hawaiian Islands. Deep-sea research. Part 2, Topical studies in oceanography 104, 267-290.

De Leonardis, C., Sandulli, R., Vanaverbeke, J., Vicx, M., De Zio, S., 2008. Meiofauna and nematode diversity in some Mediterranean subtidal areas of the Adriatic and Ionian Sea. Scientia Marina 72, 5-13.

De Mesel, I., Lee, H., Vanhove, S., Vincx, M., Vanreusel, A., 2006. Species diversity and distribution within the deep-sea nematode genus

Acantholaimus on the continental shelf and slope in Antarctica. Polar Biology 29, 860-871.

De Stigter, H., Boer, W., Jesus, C., Thomsen, L., 2007. Recent sediment transport and deposition in the Nazare Canyon, Portuguese continental margin. Marine Geology 246, 144-164.

De Bovee, F., Guidi, L. D., Soyer, J., 1990. Quantitative distribution of deep-sea meiobenthos in the Northwestern mediterranean (Gulf of Lions). Continental Shelf Research 10, 1123-1145, doi:10.1016/0278-4343(90)90077-y.

Duineveld, G., Lavaleye, M., Berghuis, E., De Wilde, P., 2001. Activity and composition of the benthic fauna in the Whittard Canyon and the adjacent continental slope (NE Atlantic). Oceanologica Acta 24, 69-83.

Dujardin, F., 1851. Sur un petit animal marin, l'Echinodère formant un type intermédiaire entre les Crustacés et les Vers. Ann Sci Nat (3) Zool 15.

Duros, P., Fontanier, C., Metzger, E., Pusceddu, A., Cesbron, F., 2011. Live (stained) benthic foraminifera in the Whittard Canyon, Celtic margin (NE Atlantic). Deep-sea research. Part 1. Oceanographic research papers 58, 128-146.

De Madron, X. D., Abassi, A., Heussner, S., Monaco, A., Aloisi, J. C., Radakovitch, O., Giresse, P., Buscail, R., Kerherve, P., 2000. Particulate matter and organic carbon budgets for the Gulf of Lions (NW Mediterranean). Oceanologica acta 23, 717-730.

Durrieu de Madron, X., Houpert, L., Puig, P., Sanchez-Vidal, A., Testor, P., Bosse, A., Estournel, C., Somot, S., Bourrin, F., Bouin, M.-N., 2013. Interaction of dense shelf water cascading and open-sea convection in the northwestern Mediterranean during winter 2012. Geophysical Research Letters 40, 1379-1385.

Eckman, J. E., Thistle, D., 1988. Small-scale spatial pattern in meiobenthos in the San Diego Trough. Deep Sea Research Part A. Oceanographic Research Papers 35, 1565-1578.

Epping, E., van der Zee, C., Soetaert, K., Helder, W., 2002. On the oxidation and burial of organic carbon in sediments of the Iberian margin and Nazaré Canyon (NE Atlantic). Progress in Oceanography 52, 399-431.

Etter, R. J., Grassle, J. F., 1992. Patterns of species diversity in the deep sea as a function of sediment particle size diversity. Nature 360, 576-578.

Estrada M., 1996 Primary production in the Northwestern Mediterranean. Scientia Marina 60:55-64.

European Union, 2011. Commission Implementing Regulation (EU) No 404/2011 of 8 April 2011 laying down detailed rules for the implementation of Council Regulation (EC) No 1224/2009 establishing a Community control system for ensuring compliance with the rules of the Common Fisheries Policy. Official Journal of the European Union L112: 1- 153.

Fabiano, M., Danovaro, R., 1999. Meiofauna distribution and mesoscale variability in two sites of the Ross Sea (Antarctica) with contrasting food supply. Polar Biology 22, 115-123.

Fagervold, S., Bessette, S., Romano, C., Martin, D., Plyuscheva, M., Le Bris, N., Galand, P. E., 2013. Microbial communities associated with the degradation of oak wood in the Blanes submarine canyon and its adjacent open slope (NW Mediterranean). Progress in Oceanography 118, 137-143.

Farrugio, H., 2012. A refugium for the spawners of exploited Mediterranean marine species: the canyons of the continental slope of the Gulf of Lion. Mediterranean Submarine Canyons: Ecology and Governance, 45-49.

Fernandez-Arcaya, U., Rotllant, G., Ramirez-Llodra, E., Recasens, L., Aguzzi, J., Flexas, M. d. M., Sanchez-Vidal, A., López-Fernández, P., García, J. A., Company, J., 2013. Reproductive biology and recruitment of the deep-sea fish community from the NW Mediterranean continental margin. Progress in Oceanography 118, 222-234.

Fernandez-Arcaya, U., Ramirez-Llodra, E., Aguzzi, J., Allcock, A. L., Davies, J. S., Dissanayake, A., Harris, P., Howell, K., Huvenne, V. A. I., Macmillan-Lawler, M., Martín, J., Menot, L., Nizinski, M., Puig, P., Rowden, A. A., Sanchez, F., Van den Beld, I. M. J., 2017. Ecological Role of Submarine Canyons and Need for Canyon Conservation: A Review. Frontiers in Marine Science 4, doi:10.3389/fmars.2017.00005.

Flach, E., Muthumbi, A., Heip, C., 2002. Meiofauna and macrofauna community structure in relation to sediment composition at the Iberian margin compared to the Goban Spur (NE Atlantic). Progress in Oceanography 52, 433-457.

Flexas, M., De Madron, X. D., Garcia, M., Canals, M., Arnau, P., 2002. Flow variability in the Gulf of Lions during the MATER HFF experiment (March–May 1997). Journal of Marine Systems 33, 197-214.

Flexas, M. M., Boyer, D. L., Espino, M., Puigdefabregas, J., Rubio, A., 2008. Circulation over a submarine canyon in the NW Mediterranean. Journal of Geophysical Research 113.

Fonseca, G., Soltwedel, T., Vanreusel, A., Lindegarth, M., 2010. Variation in nematode assemblages over multiple spatial scales and environmental conditions in Arctic deep seas. Progress in Oceanography 84, 174-184.

Forbes, E., 1843. Report on the Mollusca and Radiata of the Aegean Sea: And on Their Distribution, Considered as Bearing on Geology. Rep. Br. Assoc. Adv. Sci. 1843 129–193.

Gage, J., Ormond, R., Gage, J., Angel, M., 1997. High benthic species diversity in deep-sea sediments: the importance of hydrodynamics. Marine Biodiversity. Cambridge University Press, Cambridge.

Gage, J., 2003. Food inputs, utilisation, carbon flow and energetics. In:P.A., Tyler (ed.), Ecosystems of the World (Ecosystems of the Deep Ocean), Elsevier, Amsterdam, 313–426, 2003.

Gallucci, F., Moens, T., Fonseca, G., 2009. Small-scale spatial patterns of meiobenthos in the Arctic deep sea. Marine Biodiversity 39, 9-25.

Gambi, C., Danovaro, R., 2006. A multiple-scale analysis of metazoan meiofaunal distribution in the deep Mediterranean Sea. Deep Sea Research Part I: Oceanographic Research Papers 53, 1117-1134.

Gambi, C., Danovaro, R., 2016. Biodiversity and life strategies of deep-sea meiofauna and nematode assemblages in the Whittard Canyon (Celtic margin, NE Atlantic Ocean). Deep-sea research. Part 1. Oceanographic research papers 108, 13-22.

Gambi, C., Vanreusel, A., Danovaro, R., 2003. Biodiversity of nematode assemblages from deep-sea sediments of the Atacama Slope and Trench (South Pacific Ocean). Deep-sea research. Part 1. Oceanographic research papers 50, 103-117.

Gambi, C., Lampadariou, N., Danovaro, R., 2010. Latitudinal, longitudinal and batrhymetric paterns of abundance, biomass of metazoan meiofauna: importance of the rare taxa and anomalies in the deep Mediterranean sea. Advance Oceanography Limnology 1.

Gambi, C., Pusceddu, A., Benedetti Cecchi, L., Danovaro, R., 2014. Species richness, species turnover and functional diversity in nematodes of the deep Mediterranean Sea: searching for drivers at different spatial scales. Global ecology and biogeography 23, 24-39.

Garcia, R., Thomsen, L., 2008. Bioavailable organic matter in surface sediments of the Nazare canyon and adjacent slope (Western Iberian Margin). Journal of Marine Systems 74, 44-59.

Garcia, R., Koho, K. A., De Stigter, H. C., Epping, E., Koning, E., Thomsen, L., 2007. Distribution of meiobenthos in the Nazaré canyon and adjacent slope (western Iberian Margin) in relation to sedimentary composition. Marine Ecology Progress Series 340, 207-220. Gerritsen, H. D., Minto, C., Lordan, C., 2013. How much of the seabed is impacted by mobile fishing gear? Absolute estimates from Vessel Monitoring System (VMS) point data. ICES Journal of Marine Science: Journal du Conseil 70, 523-531.

Giere, O., 2009. Meiobenthology: the Microscopic Motile Fauna of Aquatic Sediments. Springer-Verlag, Berlin.

Gili, J. M., Bouillon, J., Palanques, A., Puig, P., 1999. Submarine canyons as habitats of prolific plankton populations: three new deep-sea

Hydroidomedusae in the western Mediterranean. Zoological Journal of the Linnean Society 125, 313-329.

Gili, J. M., Pages, F., Bouillon, J., Palanques, A., Puig, P., 2000. A multidisciplinary approach to the understanding of hydromedusan populations inhabiting Mediterranean submarine canyons. Deep-sea research. Part 1. Oceanographic research papers 47, 1513-1533.

Glover, A. G., Gooday, A. J., Bailey, D. M., Billett, D. S. M., Chevaldonne, P., DesbruyÃ¨res, D., ChevaldonnÃ©, P., ColaÃ§o, A., Copley, J., Cuvelier, D., Kalogeropoulou, V., Klages, M., Lampadariou, N.,

Gooday, A. J., Pfannkuche, O., Lambshead, P. J. D., 1996. An apparent lack of response by metazoan meiofauna to phytodetritus deposition in the bathyal north-eastern Atlantic. Journal of the Marine Biological Association of the United Kingdom 76, 297-310.

Gorska, B., Grzelak, K., Kotwicki, L., Hasemann, C., Schewe, I., Górska, B., Soltwedel, T., Włodarska Kowalczuk, M., 2014. Bathymetric variations in vertical distribution patterns of meiofauna in the surface sediments of the deep Arctic ocean (HAUSGARTEN, Fram strait). Deep-sea research. Part 1. Oceanographic research papers 91, 36-49.

Granata, T. C., Vidondo, B., Duarte, C. M., Satta, M. P., García, M., 1999. Hydrodynamics and particle transport associated with a submarine canyon off Blanes (Spain), NW Mediterranean Sea. Continental Shelf Research 19, 1249-1263.

Gremare, A., Medernach, L., deBovee, F., Amouroux, J. M., Vetion, G., Grémare, A., deBovée, F., Vétion, G., Albert, P., 2002. Relationships between sedimentary organics and benthic meiofauna on the continental shelf and the upper slope of the Gulf of Lions (NW Mediterranean). Marine Ecology Progress Series 234, 85-94.

Guidi Guilvard, L., Dallot, S., 2014. Metazoan Meiobenthos temporal fluctuations in the deep NW Mediterranean Sea (DYFAMED-BENTHOS 1993–1995). Contribution of spatial variability and disturbance. Deep-sea research. Part 1. Oceanographic research papers 92, 127-140.

Guilini, K., Bezerra, T., Deprez, T., Fonseca, G., Holovachov, O., Leduc, D., Miljutin, D., Moens, T., Sharma, J., Smol, N., 2016. NeMys: world database of free-living marine nematodes.

Guillén, J., Bourrin, F., Palanques, A., De Madron, X. D., Puig, P., Buscail, R., 2006. Sediment dynamics during wet and dry storm events on the Têt inner shelf (SW Gulf of Lions). Marine Geology 234, 129-142.

Gunton, L. M., Gooday, A. J., Glover, A. G., Bett, B. J., 2015. Macrofaunal abundance and community composition at lower bathyal depths in different branches of the Whittard Canyon and on the adjacent slope (3500m; NE

Atlantic). Deep Sea Research Part I: Oceanographic Research Papers 97, 29-39.

Gutzmann, E., Arbizu, P. M., Rose, A., Veit-Köhler, G., 2004. Meiofauna communities along an abyssal depth gradient in the Drake Passage. Deep Sea Research Part II: Topical Studies in Oceanography 51, 1617-1628.

Harris, P., Whiteway, T., 2011. Global distribution of large submarine canyons: Geomorphic differences between active and passive continental margins. Marine Geology 285, 69-86.

Harris, P. T., Macmillan Lawler, M., Rupp, J., Baker, E. K., 2014. Geomorphology of the oceans. Marine geology 352, 4-24.

Hasemann, C., Soltwedel, T., Voolstra, C., 2011. Small-Scale Heterogeneity in Deep-Sea Nematode Communities around Biogenic Structures. PLoS One 6, e29152-e29152.

Heip, C., Soetaert, K., 1988. Data processing, evaluation and analysis. Introduction to the study of meiofauna.

Heip, C., Vinx, M., Vranken, G., 1985. The Ecology of Marine Nematode. Oceanography. Mar. Bio. Ann. Rev 23, 92.

Herranz, M., Thormar, J., Benito, J., Sánchez, N., Pardos, F., 2012. *Meristoderes* gen. nov., a new kinorhynch genus, with the description of two new species and their implications for echinoderid phylogeny (Kinorhyncha: Cyclorhagida, Echinoderidae). Zoologischer Anzeiger-A Journal of Comparative Zoology 251, 161-179.

Herranz, M., Pardos, F., 2013. *Fissuroderes sorenseni* sp. nov. and *Meristoderes boylei* sp. nov.: first Atlantic recording of two rare kinorhynch genera, with new identification keys. Zool Anz A J Comp Zool 253, doi:10.1016/j.jcz.2013.09.005.

Herranz, M., Sánchez, N., Pardos, F., Higgins, R. P., 2013. New Kinorhyncha from Florida coastal waters. Helgoland Marine Research 68, 59.

Herranz M., Yangel E., Leander S., 2017. Echinoderes hakaiensis sp. nov. a new mud dragon (Kinorhyncha, Echinoderidae) from the northeastern Pacific Ocean with the redescription of Echinoderes pennaki Higgins, 1960. Marine Biodiversity (*in press*).

Hessler, R. R., Sanders, H. L., 1967. Faunal diversity in the deep-sea. Deep Sea Research and Oceanographic Abstracts, Vol. 14. Elsevier, pp. 65IN2571-70IN2878.

Heussner, S., de Madron, X. D., Calafat, A., Canals, M., Carbonne, J., Delsaut, N., Saragoni, G., 2006. Spatial and temporal variability of downward particle fluxes on a continental slope: Lessons from an 8-yr experiment in the Gulf of Lions (NW Mediterranean). Marine Geology 234, 63-92.

Higgins, R. P., 1969. Indian Ocean Kinorhyncha: 1-*Condyloderes* and *Sphenoderes,* new Cyclorhagid Genera.

Higgins, R. P., Thiel, H., 1988. Introduction to the study of meiofauna. Smithsonian Institution Press, London.

Hintzen, N. T., Bastardie, F., Beare, D., Piet, G. J., Ulrich, C., Deporte, N., Egekvist, J., Degel, H., 2012. VMStools: open-source software for the processing, analysis and visualisation of fisheries logbook and VMS data. Fisheries Research 115, 31-43.

Hoste, E., Vanhove, S., Schewe, I., Soltwedel, T., Vanreusel, A., 2007. Spatial and temporal variations in deep-sea meiofauna assemblages in the Marginal Ice Zone of the Arctic Ocean. Deep-sea research. Part 1. Oceanographic research papers 54, 109-129.

Houpert, L., Durrieu de Madron, X., Testor, P., Bosse, A., D'Ortenzio, F., Bouin, M., Dausse, D., Le Goff, H., Kunesch, S., Labaste, M., 2016. Observations of open-ocean deep convection in the northwestern Mediterranean Sea: Seasonal and interannual variability of mixing and deep water masses for the 2007-2013 Period. Journal of Geophysical Research: Oceans 121, 8139-8171.

Huang, Z., Nichol, S. L., Harris, P. T., Caley, M. J., 2014. Classification of submarine canyons of the Australian continental margin. Marine Geology 357, 362-383.

Huvenne, V. A., Tyler, P. A., Masson, D. G., Fisher, E. H., Hauton, C., Hühnerbach, V., Le Bas, T. P., Wolff, G. A., 2011. A picture on the wall: innovative mapping reveals cold-water coral refuge in submarine canyon. PloS one 6, e28755.

Ingels, J., Vanreusel, A., 2013b. The importance of different spatial scales in determining structural and functional characteristics of deep-sea infauna communities. Biogeosciences 10, 4547-4563, doi:10.5194/bg-10-4547-2013.

Ingels, J., Tchesunov, A. V., Vanreusel, A., 2011a. Meiofauna in the Gollum Channels and the Whittard Canyon, Celtic Margin—How Local Environmental Conditions Shape Nematode Structure and Function. PLoS ONE 6, e20094, doi:10.1371/journal.pone.0020094.

Ingels, J., Kiriakoulakis, K., Wolff, G., Vanreusel, A., 2009. Nematode diversity and its relation to the quantity and quality of sedimentary organic matter in the deep Nazare Canyon, Western Iberian Margin. Deep-sea research. Part 1. Oceanographic research papers 56, 1521-1539.

Ingels, J., Billett, D. S. M., Kiriakoulakis, K., Wolff, G. A., Vanreusel, A., 2011b. Structural and functional diversity of Nematoda in relation with environmental variables in the Set√∫bal and Cascais canyons, Western Iberian Margin. Deep Sea Research Part II: Topical Studies in Oceanography 58, 2354-2368, doi:10.1016/j.dsr2.2011.04.002.

Ingels, J., Vanreusel, A., Romano, C., Coenjaerts, J., Flexas, M. M., Zúñiga, D., Martin, D., 2013. Spatial and temporal infaunal dynamics of the Blanes submarine canyon-slope system (NW Mediterranean); changes in nematode standing stocks, feeding types and gender-life stage ratios. Progress in Oceanography 118, 159-174.

Jensen, P., 1984. Measuring carbon content in nematodes. Helgoländer Meeresuntersuchungen 38, 83-86.

Jensen, P., Jensen, 1988. Nematode assemblages in the deep-sea benthos of the Norwegian Sea. Deep-sea research. Part A, Oceanographic research papers 35, 1173-1184.

Jorissen, F. J., de Stigter, H. C., Widmark, J. G., 1995. A conceptual model explaining benthic foraminiferal microhabitats. Marine micropaleontology 26, 3-15.

Koenig, S., Fernández, P., Company, J. B., Huertas, D., Solé, M., 2013. Are deep-sea organisms dwelling within a submarine canyon more at risk from anthropogenic contamination than those from the adjacent open slope? A case study of Blanes canyon (NW Mediterranean). Progress in Oceanography 118, 249-259.

Koho, K. A., Garcia, R., de-Stigter, H. C., Epping, E., Koning, E., 2008. Sedimentary labile organic carbon and pore water redox control on species distribution of benthic foraminifera: A case study from Lisbon-Setubal Canyon (southern Portugal). Progress In Oceanography 79, 55-82.

Kristensen, R. M., 1991. Kinorhyncha. Microscopic anatomy of invertebrates 4, Aschelminthes. Wiley-Liss, New York. 377–404.

Lambshead, P., Tietjen, J., Ferrero, T., Jensen+, P., 2000. Latitudinal diversity gradients in the deep sea with special reference to North Atlantic nematodes. Marine Ecology Progress Series 194, 159-167.

Lambshead, P., Chen, Z., Chen, W., Chen, S., Dickson, S., 2004. Marine nematode diversity.

Lambshead, P. J. D., 2002. Latitudinal diversity patterns of deep-sea marine nematodes and organic fluxes: A test from the central equatorial pacific. Marine Ecology Progress Series 236, 129-135.

Lambshead, P. J. D., Boucher, G., 2003. Marine nematode deep-sea biodiversity – hyperdiverse or hype? Journal of Biogeography 30, 475-485, doi:10.1046/j.1365-2699.2003.00843.x.

Lambshead, P. J. D., Ferrero, T. J., Wollf, G. A., 1995. Comparison of the vertical distribution of nematodes from two contrasting abyssal sites in the Northeast Atlantic subject to different seasonal inputs of phytodetritus. Internationale Revue der gesamten Hydrobiologie und Hydrographie 80, 327-331.

Lampadariou, N., Tselepides, A., 2006. Spatial variability of meiofaunal communities at areas of contrasting depth and productivity in the Aegean Sea (NE Mediterranean). Progress in Oceanography 69, 19-36.

Lampitt, R., 1985. Evidence for the seasonal deposition of detritus to the deep-sea floor and its subsequent resuspension. Deep Sea Research Part A. Oceanographic Research Papers 32, 885-897.

Landers, S. C., Sørensen, M. V., 2016. Two new species of Echinoderes (Kinorhyncha, Cyclorhagida), E. romanoi sp. n. and E. joyceae sp. n., from the Gulf of Mexico. ZooKeys 594, doi:10.3897/zookeys.594.8623.

Lastras, G., Canals, M., Amblas, D., Lavoie, C., Church, I., 2011. Understanding sediment dynamics of two large submarine valleys from seafloor data: Blanes and La Fonera canyons, northwestern Mediterranean Sea. Marine Geology 280, 20-39.

Leduc, D., Probert, P., Nodder, S., 2010. Influence of mesh size and core penetration on estimates of deep-sea nematode abundance, biomass, and diversity. Deep Sea Research Part I: Oceanographic Research Papers 57, 1354-1362.

Leduc, D., Rowden, A., Bowden, D., Probert, P. K., Pilditch, C., Nodder, S. D., 2012a. Unimodal relationship between biomass and species richness of deep-sea nematodes: implications for the link between productivity and diversity. Marine ecology progress series 454, 53-64.

Leduc, D., Rowden, A., Probert, P. K., Pilditch, C., Nodder, S., Vanreusel, A., Witbaard, R., 2012b. Further evidence for the effect of particle-size diversity on deep-sea benthic biodiversity. Deep-sea research. Part 1. Oceanographic research papers 63, 164-169.

Leduc, D., Rowden, A. A., Nodder, S. D., Berkenbusch, K., Probert, P. K., Hadfield, M. G., 2014. Unusually high food availability in Kaikoura Canyon linked to distinct deep-sea nematode community. Deep Sea Research Part II: Topical Studies in Oceanography 104, 310-318, doi:http://dx.doi.org/10.1016/j.dsr2.2013.06.003.

Leduc, D., Nodder, S., Berkenbusch, K., Rowden, A., 2015. Effect of core surface area and sediment depth on estimates of deep-sea nematode genus richness and community structure. Marine Biodiversity 45, 349-356.

Lee, H., Vanhove, S., Peck, L. S., Vincx, M., 2001. Recolonisation of meiofauna after catastrophic iceberg scouring in shallow Antarctic sediments. Polar Biology 24, 918-925.

Levin, L., Sibuet, M., 2012. Understanding Continental Margin Biodiversity: A New Imperative. Annual Review of Marine Science 4, 79-112.

Levin, L. A., Dayton, P. K., 2009. Ecological theory and continental margins: where shallow meets deep. Trends in ecology & evolution 24, 606-617.

Levin, L. A., Sibuet, M., Gooday, A. J., Smith, C. R., Vanreusel, A., 2010. The roles of habitat heterogeneity in generating and maintaining biodiversity on continental margins: an introduction. Marine Ecology 31, 1-5.

Levin, L. A., Etter, R. J., Rex, M. A., Gooday, A. J., Smith, C. R., Pineda, J., Stuart, C. T., Hessler, R. R., Pawson, D., 2001. Environmental Influences on Regional Deep-Sea Species Diversity 1. Annual Review of Ecology and Systematics 32, 51-93.

Lins, L., Vanreusel, A., van Campenhout, J., Ingels, J., 2013. Selective settlement of deep-sea canyon nematodes after resuspension — an experimental approach. Journal of Experimental Marine Biology and Ecology 441, 110-116.

Lins, L., da Silva, M. C., Hauquier, F., Esteves, A. M., Vanreusel, A., 2015. Nematode community composition and feeding shaped by contrasting productivity regimes in the Southern Ocean. Progress in Oceanography 134, 356-369.

Lins, L., Guilini, K., Veit-Köhler, G., Hauquier, F., Alves, R. M. d. S., Esteves, A., Vanreusel, A., 2014. The link between meiofauna and surface productivity in the Southern Ocean. Deep Sea Research Part II: Topical Studies in Oceanography 108, 60-68.

Lopez Fernandez, P., Calafat, A., Sanchez Vidal, A., Canals, M., Flexas, M. M., Mar Flexas, M., Cateura, J., Company, J., 2013. Multiple drivers of particle fluxes in the Blanes submarine canyon and southern open slope: Results of a year round experiment. Progress in oceanography 118, 95-107.

Maiorano, P., Tselepides, A., Politou, C. Y., Plaity, W., Rotllant, G., Sardá, F., 2004. Deep-sea decapod crustaceans in the western and central Mediterranean Sea: preliminary aspects of species distribution, biomass and population structure. Scientia Marina 68, 73-86.

Mare, M. F., 1942. A study of a marine benthie community with special reference to tho micro-organisms. Journal of the Marine Biological Association Plymouth 25, 517-554.

Martín, J., Palanques, A., Puig, P., 2006. Composition and variability of downward particulate matter fluxes in the Palamós submarine canyon (NW Mediterranean). Journal of Marine Systems 60, 75-97.

Martín, J., Puig, P., Palanques, A., Ribó, M., 2014a. Trawling-induced daily sediment resuspension in the flank of a Mediterranean submarine canyon. Deep Sea Research Part II: Topical Studies in Oceanography 104, 174-183.

Martín, J., Puig, P., Palanques, A., Masqué, P., García-Orellana, J., 2008. Effect of commercial trawling on the deep sedimentation in a Mediterranean submarine canyon. Marine Geology 252, 150-155.

Martín, J., Puig, P., Masqué, P., Palanques, A., Sánchez-Gómez, A., 2014b. Impact of bottom trawling on deep-sea sediment properties along the flanks of a submarine canyon. PloS one 9, e104536.

Martorelli, S., Higgins, R. P., 2004. Kinorhyncha from the stomach of the shrimp Pleoticus muelleri (Bate, 1888) from Comodoro Rivadavia, Argentina. Zoologischer Anzeiger-A Journal of Comparative Zoology 243, 85-98.

Masson, D., Huvenne, V., De Stigter, H., Wolff, G., Kiriakoulakis, K., Arzola, R., Blackbird, S., 2010. Efficient burial of carbon in a submarine canyon. Geology 38, 831-834.

Maurer, D., Robertson, G., Gerlinger, T., 1994. Comparison of Community Structure of Soft-Bottom Macrobenthos of the Newport Submarine Canyon, California and the Adjoining Shelf. Internationale Revue der Gesamten Hydrobiologie und Hydrographie 79, 591-603.

McClain, C. R., Barry, J. P., 2010. Habitat heterogeneity, disturbance, and productivity work in concert to regulate biodiversity in deep submarine canyons. Ecology 91, 964-976.

McClain, C. R., Hardy, S. M., 2010. The dynamics of biogeographic ranges in the deep sea. Proceedings of the Royal Society of London B: Biological Sciences 277, 3533-3546. doi:10.1098/rspb.2010.1057

McClain, C. R., Schlacher, T. A., 2015. On some hypotheses of diversity of animal life at great depths on the sea floor. Marine Ecology 36, 849-872.

McIntyre, A., 1962. The class kinorhyncha (Echinoderida) in British waters. Journal of the Marine Biological Association of the United Kingdom 42, 503-509.

Mecho, A., Billett, D. S., Ramírez-Llodra, E., Aguzzi, J., Tyler, P. A., 2014. First records, rediscovery and compilation of deep-sea echinoderms in the middle and lower continental slope of the Mediterranean Sea. Scientia Marina 78, 281-302.

Mecho, A., Fernandez-Arcaya, U., Ramirez-Llodra, E., Aguzzi, J., 2015. Reproductive biology of the seastar *Ceramaster grenadensis* from the deep north-western Mediterranean Sea. Journal of the Marine Biological Association of the United Kingdom 95, 805-815.

Menot, L., Sibuet, M., Carney, R. S., Levin, L. A., Rowe, G. T., Billett, D. S., Poore, G., Kitazato, H., Vanreusel, A., Galéron, J., 2010. New perceptions of continental margin biodiversity. In Life in the World's Oceans: Diversity, Distribution, and Abundance, edited by: McIntyre, AD, 79–103. Cens. Mar. Life. Oxford, UK: Wiley-Blackwell.

Miljutin, D. M., Gad, G., Miljutina, M. M., Mokievsky, V. O., Fonseca-Genevois, V., Esteves, A. M., 2010. The state of knowledge on deep-sea nematode taxonomy: how many valid species are known down there? Marine Biodiversity 40, 143-159.

Millot, C., 1999. Circulation in the western Mediterranean Sea. Journal of Marine Systems 20, 423-442.

Moens, T., Braeckman, U., Derycke, S., Fonseca, G., Gallucci, F., Gingold, R., Guilini, K., Ingels, J., Leduc, D., Vanaverbeke, J., Van Colen, C., Vanreusel, A., Vincx, M., 2014. Ecology of free-living marine nematodes, in: Schmidt-Rhaesa, A. (Ed.), Handbook of Zoology: Gastrotricha, Cycloneuralia and Gnathifera. Nematoda 2, 109–152.

Mokievskii, V., Udalov, A., Azovskii, A., 2007. Quantitative distribution of meiobenthos in deep-water zones of the World Ocean. Oceanology 47, 797-813.

Moranta, J., Stefanescu, C., Massutí, E., Morales-Nin, B., Lloris, D., 1998. Fish community structure and depth-related trends on the continental slope of the Balearic Islands (Algerian basin, western Mediterranean). Marine Ecology Progress Series 171, 247-259.

Muthumbi, A., Vincx, M., 1996. Nematodes from the Indian Ocean: description of six new species of the genus Molgolaimus Ditlevsen, 1921 (Nematoda: Desmodoridae). Bulletin. Institut Royal des Sciences Naturelles de Belgique. Koninklijk Belgisch Instituut voor Natuurwetenschappen.

Muthumbi, A., Vanreusel, A., Duineveld, G., Soetaert, K., Vincx, M., 2004. Nematode Community Structure along the Continental Slope off the Kenyan Coast, Western Indian Ocean. International review of hydrobiology 89, 188-205.

Nebelsick, M., 1990. Antygomonas incomitata gen. et sp. n.(Cyclorhagida, Kinorhyncha) and its phylogenetic relationships. Zoologica Scripta 19, 143-152.

Neuhaus, B., Lüter, C., 2003. Ausfahrten SO 144–143 und SO 158: Zoobenthische Untersuchungen im zentralamerikanischen Ostpazifik. Statusseminar 2003 Meeresforschung mit FS Sonne, 12.-14. M.rz 2003 in Hamburg, Abstracts, pp. 123–126

Neuhaus, B., 2004. Description of Campyloderes cf. vanhoeffeni (Kinorhyncha, Cyclorhagida) from the Central American East Pacific deep sea with a review of the genus. Meiofauna Mar 13, 3-20.

Neuhaus, B., 2013. Kinorhyncha (=Echinodera). In: Schmidt-Rhaesa, A., (Ed.), Handbook of zoology. Gastrotricha, Cycloneuralia and Gnathifera, vol 1: Nematomorpha, Priapulida, Kinorhyncha, Loricifera. De Gruyter, Berlin.

Neuhaus, B., Blasche, T., 2006. *Fissuroderes*, a new genus of Kinorhyncha (Cyclorhagida) from the deep sea and continental shelf of New Zealand and from the continental shelf of Costa Rica. Zoologischer Anzeiger 245, 19-52. doi:10.1016/j.jcz.2006.03.003.

Neuhaus, B., Sørensen, M. V., 2013. Populations of Campyloderes sp.(Kinorhyncha, Cyclorhagida): One global species with significant morphological variation? Zoologischer Anzeiger-A Journal of Comparative Zoology 252, 48-75.

Neves, R., Sørensen, M. V., Herranz, M., 2016. First account on kinorhynchs from Portugal, with the description of two new species: Echinoderes lusitanicus sp. nov. and E. reicherti sp. nov. Mar. Biol Res.

Nieuwenhuize, J., Maas, Y. E., Middelburg, J. J., 1994. Rapid analysis of organic carbon and nitrogen in particulate materials. Marine Chemistry 45, 217-224.

Palanques, A., de Madron, X., Puig, P., Fabres, J., Guillen, J., 2006a. Suspended sediment fluxes and transport processes in the Gulf of Lions submarine canyons. The role of storms and dense water cascading. Marine Geology 234, 43-61.

Palanques, A., Martín, J., Puig, P., Guillén, J., Company, J., Sardà, F., 2006b. Evidence of sediment gravity flows induced by trawling in the Palamós (Fonera) submarine canyon (northwestern Mediterranean). Deep Sea Research Part I: Oceanographic Research Papers 53, 201-214.

Palanques, A., Masqué, P., Puig, P., Sanchez-Cabeza, J. A., Frignani, M., Alvisi, F., 2008. Anthropogenic trace metals in the sedimentary record of the Llobregat continental shelf and adjacent Foix Submarine Canyon (northwestern Mediterranean). Marine Geology 248, 213-227.

Palanques, A., García-Ladona, E., Gomis, D., Martín, J., Marcos, M., Pascual, A., Puig, P., Gili, J.-M., Emelianov, M., Monserrat, S., 2005. General patterns of circulation, sediment fluxes and ecology of the Palamós (La Fonera) submarine canyon, northwestern Mediterranean. Progress in Oceanography 66, 89-119.

Pape, E., Bezerra, T. N., Jones, D. O. B., Vanreusel, A., 2013a. Unravelling the environmental drivers of deep-sea nematode biodiversity and its relation with carbon mineralisation along a longitudinal primary productivity gradient. Biogeosciences 10, 3127-3143, doi:10.5194/bg-10-3127-2013.

Pape, E., Manini, E., Bezerra, T., Vanreusel, A., Jones, D. O. B., 2013b. Benthic-Pelagic Coupling: Effects on Nematode Communities along Southern European Continental Margins. PLoS ONE 8, e59954.

Paradis, S., Puig, P., Masqué, P., Juan-Díaz, X., Martín, J., Palanques, A., 2017. Bottom-trawling along submarine canyons impacts deep sedimentary regimes. Scientific Reports 7.

Pardos, F., Sánchez, N., Herranz, M., 2016a. Two sides of a coin: The phylum Kinorhyncha in Panama. I) Caribbean Panama. Zoologischer Anzeiger-A Journal of Comparative Zoology 265, 3-25.

Pardos, F., Herranz, M., Sánchez, N., 2016b. Two sides of a coin: the phylum Kinorhyncha in Panama. II) Pacific Panama. Zoologischer Anzeiger-A Journal of Comparative Zoology 265, 26-47.

Pasqual, C., Sanchez Vidal, A., Zuniga, D., Calafat, A., Canals, M., 2010. Flux and composition of settling particles across the continental margin of the Gulf of Lion: the role of dense shelf water cascading. Biogeosciences 7, 217-231.

Paterson, G. L. J., Glover, A. G., Cunha, M. R., Neal, L., de Stigter, H. C., Kiriakoulakis, K., Billett, D. S. M., Wolff, G. A., Tiago, A., Ravara, A., Lamont, P., Tyler, P., 2011. Disturbance, productivity and diversity in deep-sea canyons: A worm's eye view. Deep Sea Research Part II: Topical Studies in Oceanography 58, 2448-2460, doi:http://dx.doi.org/10.1016/j.dsr2.2011.04.008.

Pedrosa-Pàmies, R., Sanchez-Vidal, A., Calafat, A., Canals, M., Durán, R., 2013. Impact of storm-induced remobilization on grain size distribution and organic carbon content in sediments from the Blanes Canyon area, NW Mediterranean Sea. Progress in Oceanography 118, 122-136.

Pielou, E. C., 1969. An introduction to mathematical ecology. An introduction to mathematical ecology.

Plante-Cuny, M.-R., Bodoy, A., 1987. Biomasse et production primaire du phytoplancton et du microphytobenthos de deux biotopes sableux (Golfe de Fos, France). Oceanologica acta 10, 223-237.

Platt, H., Warwick, R., 1988. Freeliving marine nematodes: Part II. British Chromadorida. Synopses of the British Fauna No. 38. EJ Brill, Dr. W. Backhuys for the Linnean Society of London and the Estuarine and Brackish-water Sciences Associatio.

Puig, P., Palanques, A., Martín, J., 2014. Contemporary sediment-transport processes in submarine canyons. Annual review of marine science 6, 53-77.

Puig, P., Palanques, A., Guillén, J., García-Ladona, E., 2000. Deep slope currents and suspended particle fluxes in and around the Foix submarine canyon (NW Mediterranean). Deep Sea Research Part I: Oceanographic Research Papers 47, 343-366, doi:http://dx.doi.org/10.1016/S0967-0637(99)00062-X.

Puig, P., Martín, J., Masqué, P., Palanques, A., 2015a. Increasing sediment accumulation rates in La Fonera (Palamós) submarine canyon axis and their relationship with bottom trawling activities. Geophysical Research Letters 42, 8106-8113.

Puig, P., Masqué, P., Martín, J., Paradis Vilar, S., Juan Díaz, X., Toro, M., Palanques, A., 2015b. Changes in sediment accumulation rates within NW Mediterranean submarine canyons caused by bottom trawling activities.

Puig, P., Canals, M., Company, J., Martin, J., Amblas, D., Martín, J., Lastras, G., Palanques, A., Calafat, A., 2012. Ploughing the deep sea floor. Nature 489, 286-289.

Pusceddu, A., 2010. Organic matter in sediments of canyons and open slopes of the Portuguese, Catalan, Southern Adriatic and Cretan Sea margins. Deep-sea research. Part 1. Oceanographic research papers 57, 441-457.

Pusceddu, A., Bianchelli, S., Martin, J., Puig, P., Palanques, A., Martín, J., Masqué, P., Danovaro, R., Masque, P., 2014. Chronic and intensive bottom trawling impairs deep-sea biodiversity and ecosystem functioning. Proceedings of the National Academy of Sciences of the United States of America 111, 8861-8866.

Pusceddu, A., Mea, M., Gambi, C., Bianchelli, S., Canals, M., Sanchez-Vidal, A., Calafat, A., Heussner, S., De Madron, X. D., Avril, J., 2010. Ecosystem effects of dense water formation on deep Mediterranean Sea ecosystems: an overview. Advances in Oceanography and Limnology 1, 67-83.

Pusceddu, A., Mea, M., Canals, M., Heussner, S., Durrieu de Madron, X., Sanchez-Vidal, A., Bianchelli, S., Corinaldesi, C., Dell'Anno, A., Thomsen, L., Danovaro, R., 2013. Major consequences of an intense dense shelf water cascading event on deep-sea benthic trophic conditions and meiofaunal biodiversity. Biogeosciences 10, 2659-2670, doi:10.5194/bg-10-2659-2013.

Ramalho, S., Adao, H., Kiriakoulakis, K., Wolff, G., Vanreusel, A., Adão, H., Ingels, J., 2014. Temporal and spatial variation in the Nazaré Canyon (Western Iberian margin): Inter-annual and canyon heterogeneity effects on meiofauna biomass and diversity. Deep-sea research. Part 1. Oceanographic research papers 83, 102-114.

Ramirez-Llodra, E., De Mol, B., Company, J. B., Coll, M., Sardà, F., 2013. Effects of natural and anthropogenic processes in the distribution of marine litter in the deep Mediterranean Sea. Progress in Oceanography 118, 273-287, doi:http://dx.doi.org/10.1016/j.pocean.2013.07.027.

Ramírez-Llodra, E., Ballesteros, M., Dantart, L., Sardà, F., 2008. Spatio-temporal variations of biomass and abundance in bathyal non-crustacean megafauna in the Catalan Sea (North-western Mediterranean). Marine Biology 153, 297-309.

Ramirez Llodra, E., Baptista Company, J., Sarda, F., Rotllant, G., 2010a. Megabenthic diversity patterns and community structure of the Blanes submarine canyon and adjacent slope in the Northwestern Mediterranean: a human overprint? Marine Ecology 31, 167-182.

Ramirez Llodra, E., Brandt, A., Danovaro, R., De Mol, B., Escobar, E., German, C. R., Levin, L. A., Martinez Arbizu, P., Menot, L., Buhl Mortensen, P., Narayanaswamy, B. E., Smith, C. R., Tittensor, D. P., Tyler, P. A., Vanreusel, A., Vecchione, M., 2010b. Deep, diverse and definitely different: unique attributes of the world's largest ecosystem. Biogeosciences 7, 2851-2899.

Reinhard, W., 1881. Über Echinoderes und Desmoscolex der Umgegend von Odessa. Zool. Anz 4, 588-592.

Remane, A., 1929. Dritte Klasse des Cladus Nemathelminthes, Kinorhyncha = Echinodera. pp. 187–248 in Krumbach, T. (ed.) Handbuch der Zoologie. Eine Naturgeschichte der St.mme des Tierreichse. Sechste Lieferung. Walter de Gruyter & Co, Berlin.

Rex, M. A., 1981. Community structure in the deep-sea benthos. Annual Review of Ecology and Systematics 12, 331-353.

Rex, M. A., Etter, R. J., 2010. Deep-sea biodiversity: pattern and scale. Harvard University Press.

Rex, M. A., Etter, R. J., Morris, J. S., Crouse, J., McClain, C. R., Johnson, N. A., Stuart, C. T., Deming, J. W., Thies, R., Avery, R., 2006. Global bathymetric patterns of standing stock and body size in the deep-sea benthos. Marine Ecology Progress Series 317, 1-8.

Román, S., Vanreusel, A., Romano, C., Ingels, J., Puig, P., Company, J. B., Martin, D., 2016. High spatiotemporal variability in meiofaunal assemblages in Blanes Canyon (NW Mediterranean) subject to anthropogenic and natural disturbances. Deep Sea Research Part I: Oceanographic Research Papers 117, 70-83, doi:http://dx.doi.org/10.1016/j.dsr.2016.10.004.

Romano, C., Voight, J., Company, J. B., Plyuscheva, M., Martin, D., 2013a. Submarine canyons as the preferred habitat for wood-boring species of Xylophaga (Mollusca, Bivalvia). Progress in Oceanography 118, 175-187.

Romano, C., Coenjaerts, J., Flexas, M. M., Zúñiga, D., Vanreusel, A., Company, J. B., Martin, D., 2013b. Spatial and temporal variability of meiobenthic density in the Blanes submarine canyon (NW Mediterranean). Progress in Oceanography 118, 144-158.

Rosenzweig, M. L., 1995. Species diversity in space and time. Cambridge University Press.

Rosli, N., Leduc, D., Rowden, A., Clark, M., Probert, P. K., Berkenbusch, K., Neira, C., 2016. Differences in meiofauna communities with sediment depth are greater than habitat effects on the New Zealand continental margin: implications for vulnerability to anthropogenic disturbance. PeerJ 4, e2154-e2154.

Rowe, G. T., Polloni, P. T., Haedrich, R. L., 1982. The deep-sea macrobenthos on the continental margin of the northwest Atlantic Ocean. Deep Sea Research Part A. Oceanographic Research Papers 29, 257-278, doi:http://dx.doi.org/10.1016/0198-0149(82)90113-3.

Rucabado, J., Lloris, D., Stefanescu, C., 1991. OTSB14: Un arte de arrastre bentónico para la pesca profunda (por debajo de los 1000 metros).

Rumolo, P., Cartes, J. E., Fanelli, E., Papiol, V., Sprovieri, M., Mirto, S., Gherardi, S., Bonanno, A., 2015. Seasonal variations in the source of sea bottom organic matter off Catalonia coasts (western Mediterranean): links with hydrography and biological response. Journal of oceanography 71, 325-343.

Ryan, J. P., Chavez, F. P., Bellingham, J. G., 2005. Physical-biological coupling in Monterey Bay, California: topographic influences on phytoplankton ecology. Marine Ecology Progress Series 287, 23-32.

Sánchez-Tocino, L., Tierno de Figueroa, J., López-Rodríguez, M., Liébanas, G., 2011. First record of Echinoderes dujardinii Claparède, 1863 (Kinorhyncha, Cyclorhagida) in Iberian Peninsula coast waters. Zoologica Baetica 22, 179-184.

Sanchez-Vidal, A., Llorca, M., Farré, M., Canals, M., Barceló, D., Puig, P., Calafat, A., 2015. Delivery of unprecedented amounts of perfluoroalkyl substances towards the deep-sea. Science of The Total Environment 526, 41-48, doi:http://dx.doi.org/10.1016/j.scitotenv.2015.04.080.

Sanchez-Vidal, A., Canals, M., Calafat, A. M., Lastras, G., Pedrosa-Pàmies, R., Menéndez, M., Medina, R., Hereu, B., Romero, J., Alcoverro, T., 2012. Impacts on the deep-sea ecosystem by a severe coastal storm. PLoS One 7, e30395.

Sánchez, N., Pardos, F., Sørensen, M. V., 2014a. Deep-sea Kinorhyncha: two new species from the Guinea Basin, with evaluation of an unusual male feature. Organisms Diversity & Evolution 14, 349-361.

Sánchez, N., Pardos, F., Sørensen, M. V., 2014b. A new kinorhynch genus, Mixtophyes (Kinorhyncha: Homalorhagida), from the Guinea Basin deep-sea, with new data on the family Neocentrophyidae. Helgoland Marine Research 68, 221.

Sánchez, N., Herranz, M., Benito, J., Pardos, F., 2012. Kinorhyncha from the Iberian Peninsula: new data from the first intensive sampling campaigns. Zootaxa 3402, 24-44.

Sánchez, N., Yamasaki, H., Pardos, F., Sørensen, M. V., Martínez, A., 2016. Morphology distangles the systematics of a ubiquitous but elusive meiofaunal group (Kinorhyncha: Pycnophyidae). Cladistics.

Sanders, H. L., 1968. Marine benthic diversity: a comparative study. The American Naturalist 102, 243-282.

Sarda, F., Cartes, J., Norbis, W., 1995. Spatio-temporal structure of the deep-water shrimp Aristeus antennatus (Decapoda: Aristeidae) population in the western Mediterranean. Oceanographic Literature Review 5, 399.

Sardà, F., Cartes, J., Norbis, W., 1994a. Spatio-temporal structure of the deep-water shrimp *Aristeus antennatus*(Decapoda: Aristeidae) population in the western Mediterranean. Fishery Bulletin 92, 599-607.

Sardà, F., Cartes, J., Company, J., 1994b. Spatio-temporal variations in megabenthos abundance in three different habitats of the Catalan deep-sea (Western Mediterranean). Marine Biology 120, 211-219.

Sardà, F., Calafat, A., Flexas, M. M., Tselepides, A., Canals, M., Espino, M., A., T., 2004. An introduction to Mediterranean deep-sea biology. Scientia marina 68, 32.

Sardà, F., Company, J. B., Bahamón, N., Rotllant, G., Flexas, M. M., Sánchez, J. D., Zúñiga, D., Coenjaerts, J., Orellana, D., Jordà, G., Puigdefábregas, J., Sánchez-Vidal, A., Calafat, A., Martín, D., Espino, M., 2009. Relationship between environment and the occurrence of the deep-water rose shrimp Aristeus antennatus (Risso, 1816) in the Blanes submarine canyon (NW Mediterranean). Progress In Oceanography 82, 227-238 doi:10.1016/j.pocean.2009.07.001.

Sardou, J., Etienne, M., Andersen, V., 1996. Seasonal abundance and vertical distributions of macroplankton and micronekton in the Northwestern Mediterranean Sea. Oceanologica acta 19, 645-656.Schlacher, T. A., Williams, A., Althaus, F., Schlacher-Hoenlinger, M. A., 2010. High-resolution seabed imagery as a tool for biodiversity conservation planning on continental margins. Marine Ecology 31, 200-221.

Schratzberger, M., Jennings, S., 2002. Impacts of chronic trawling disturbance on meiofaunal communities. Marine biology 141, 991-1000.

Schratzberger, M., Lampadariou, N., Somerfield, P., Vandepitte, L., Berghe, E. V., 2009. The impact of seabed disturbance on nematode communities: linking field and laboratory observations. Marine Biology 156, 709.

Schratzberger, M., Ingels, J., 2017. Meiofauna matters: The roles of meiofauna in benthic ecosystems. Journal of Experimental Marine Biology and Ecology.

Sevastou, K., Lampadariou, N., Polymenakou, P., Tselepides, A., 2013. Benthic communities in the deep Mediterranean Sea: exploring microbial and meiofaunal patterns in slope and basin ecosystems. Biogeosciences 10, 4861-4878.

Shannon, C., 1948. A mathematical theory of communication, bell System technical Journal 27: 379-423 and 623–656. Mathematical Reviews (MathSciNet): MR10, 133e.

Shepard, F. P., Dill, R. F., 1966. Submarine canyons and other sea valleys. Rand McNally.

Shimanaga, M., Kitazato, H., Shirayama, Y., 2000. Seasonal patterns of vertical distribution between meiofaunal groups in relation to phytodetritus deposition in the bathyal Sagami Bay, central Japan. Journal of oceanography 56, 379-387.

Shirayama, Y., 1984. The abundance of deep-sea meiobenthos in the Western Pacific in relation to environmental-factors. Oceanologica Acta 7, 113-121.

Shirayama, Y., Horikoshi, M., 1982. Vertical distribution of smaller macrobenthos and larger meiobenthos in the sediment profile in the deep-sea system of Suruga Bay (Central Japan). Journal of the Oceanographical Society of Japan 38, 273-280.

Smith, C. R., Baco, A. R., 2003. Ecology of whale falls at the deep-sea floor. Oceanography and marine biology 41, 311-354.

Smith, C. R., De Leo, F. C., Bernardino, A. F., Sweetman, A. K., Arbizu, P. M., 2008. Abyssal food limitation, ecosystem structure and climate change. Trends in Ecology & Evolution 23, 518-528.

Snelgrove, P. V. R., Smith, C. R., Gibson, Ansell, A. D., Atkinson, R. J. A., Atkinson, R. J., Barnes, H., Barnes, M., Gibson, R. N., 2002. A riot of species in an environmental calm: The paradox of the species-rich deep-sea floor Oceanography And Marine Biology.

Soetaert, K., Heip, C., 1995. Nematode assemblages of deep-sea and shelf break sites in the North Atlantic and Mediterranean Sea. Marine ecology progress series 125, 171-183.

Soetaert, K., Heip, C., Vincx, M., 1991a. The meiobenthos along a Mediterranean deep-sea transect off Calvi (Corsica) and in an adjacent canyon. Marine Ecology 12, 227-242.

Soetaert, K., Heip, C., Vincx, M., 1991b. Diversity of nematode assemblages along a Mediterranean deep-sea transect. Marine Ecology Progress Series 75, 275-282.

Soetaert, K., Vincx, M., Heip, C., 1995. Nematode community structure along a Mediterranean shelf-slope gradient. Marine Ecology 16, 189-206.

Soetaert, K., Herman, P. M., Middelburg, J. J., 1996. Dynamic response of deep-sea sediments to seasonal variations: a model. Limnology and Oceanography 41, 1651-1668.

Soetaert, K., Muthumbi, A., Heip, C., 2002. Size and shape of ocean margin nematodes: morphological diversity and depth-related patterns. Marine ecology progress series 242, 179-193.

Soetaert, K., Vanaverbeke, J., Heip, C., Herman, P. M. J., Middelburg, J. J., 1997. Nematode distribution in ocean margin sediments of the Goban Spur (northeast Atlantic) in relation to sediment geochemistry. Deep-sea research. Part 1. Oceanographic research papers 44, 1671-1683.

Soetaert, K., Franco, M., Lampadariou, N., Muthumbi, A., Steyaert, M., Vandepitte, L., vanden Berghe, E., Vanaverbeke, J., 2009. Factors affecting nematode biomass, length and width from the shelf to the deep sea. Marine ecology progress series 392, 123-132.

Soltwedel, T., 2000. Metazoan meiobenthos along continental margins: a review. Progress in Oceanography 46, 59-84.

Soltwedel, T., Hasemann, C., Quéric, N.-V., von Juterzenka, K., 2005. Gradients in activity and biomass of the small benthic biota along a channel system in the deep Western Greenland Sea. Deep-sea research. Part 1. Oceanographic research papers 52, 815-835.

Sørensen, M. V., 2008. A new kinorhynch genus from the Antarctic deep sea and a new species of Cephalorhyncha from Hawaii (Kinorhyncha: Cyclorhagida: Echinoderidae). Org Divers Evol 8.

Sørensen, M. V., Pardos, F., 2008. Kinorhynchs systematic and biology—an introduction to the study of kinorhynchs, inclusive identification keys to genera. Meiofauna Mar 6.

Sørensen, M. V., Landers, S. C., 2017. Description of a new kinorhynch species, *Paracentrophyes sanchezae* n. sp.(Kinorhyncha: Allomalorhagida) from the Gulf of Mexico, with differential notes on one additional, yet undescribed species of the genus. Zootaxa 4242, 61-76.

Sørensen, M. V., Rho, H. S., Kim, D., 2010a. A new species of *Condyloderes* (Cyclorhagida, Kinorhyncha) from Korea. Zoological science 27, 234-242.

Sørensen, M. V., Rho, H. S., Kim, D., 2010b. A new species of the rare genus *Sphenoderes* (Cyclorhagida, Kinorhyncha), with differential notes on *S. indicus* Higgins, 1969. Marine Biology Research 6, 472-484.

Sørensen, M. V., Herranz, M., Landers, S. C., 2016. A new species of *Echinoderes* (Kinorhyncha: Cyclorhagida) from the Gulf of Mexico, with a redescription of *Echinoderes bookhouti* Higgins, 1964. Zool Anz A J Comp Zool.

Sørensen, M. V., Zotto, M., Rho, H. S., Herranz, M., Sánchez, N., Pardos, F., Yamasaki, H., 2015. Phylogeny of Kinorhyncha based on morphology and two molecular loci. PLoS One 10, doi:10.1371/journal.pone.0133440.

Soyer, J., 1985. Mediterranean Sea meiobenthos. NATO conference series. I, Ecology 8, 85-108.

Stefanescu, C., Lloris, D., Rucabado, J., 1993. Deep-sea fish assemblages in the Catalan Sea (western Mediterranean) below a depth of 1000 m. Deep Sea Research Part I: Oceanographic Research Papers 40, 695-707.

Stefanescu, C., Nin-Morales, B., Massuti, E., 1994. Fish assemblages on the slope in the Catalan Sea (Western Mediterranean): influence of a submarine canyon. Journal of the Marine Biological Association of the United Kingdom 74, 499-512.

Sutton, T., Porteiro, F., Heino, M., Byrkjedal, I., Langhelle, G., Anderson, C., Horne, J., Søiland, H., Falkenhaug, T., Godø, O., 2008. Vertical structure, biomass and topographic association of deep-pelagic fishes in relation to a mid-ocean ridge system. Deep Sea Research Part II: Topical Studies in Oceanography 55, 161-184.

Tassone, A., Roca, E., Muñoz, J., Cabrera, L., Canals, M., 1994. Evolución del sector septentrional del margen continental catalán durante el Cenozoico. Acta geológica hispánica 29, 3-37.

Tchesunov, A., Ingels, J., Popova, E., 2012. Marine free-living nematodes associated with symbiotic bacteria in deep-sea canyons of north-east Atlantic Ocean. Journal of the Marine Biological Association of the United Kingdom 92, 1257-1271.

Tecchio, S., Ramírez-Llodra, E., Aguzzi, J., Sanchez-Vidal, A., Flexas, M. M., Sardà, F., Company, J. B., 2013. Seasonal fluctuations of deep megabenthos:

Finding evidence of standing stock accumulation in a flux-rich continental slope. Progress in Oceanography 118, 188-198.

Tesi, T., Puig, P., Palanques, A., Goni, M. A., 2010. Lateral advection of organic matter in cascading-dominated submarine canyons. Progress In Oceanography 84, 185-203.

Thiel, H., 1975. The size and structure of the deep-sea benthos. International Revue ges Hydrobiol.

Thiel, H., 1978. Benthos in upwelling regions. Upwelling ecosystems. Berlin: Springer, 124-138.

Thiel, H., 1983. Meiobenthos and nanobenthos of the deep sea. The sea 8, 167-230.

Thistle, D., 2003. The deep-sea floor: an overview. Ecosystems of the World, 5-38.

Thistle, D., Yingst, J., Fauchald, K., 1985. A deep-sea benthic community exposed to strong near-bottom currents on the Scotian Rise (western Atlantic). Marine Geology 66, 91-112.

Thomson, C. W., 1873. The Depths of the Sea. Macmillan and Co., London.

Thurber, A. R., Sweetman, A. K., Narayanaswamy, B. E., Narayanaswamy, B. E., Ingels, J., Hansman, R. L., 2014. Ecosystem function and services provided by the deep sea. Biogeosciences 11, 3941-3963.

Tietjen, J. H., 1971. Ecology and distribution of deep-sea meiobenthos off North Carolina. Deep Sea Research and Oceanographic Abstracts, Vol. 18. Elsevier, pp. 941-944 in 2957.

Tietjen, J. H., 1992. Abundance and biomass of metazoan meiobenthos in the deep-sea food chains and the global carbon cycle. NATO ASI series. Series C, Mathematical and physical sciences 360, 45-62.

Tselepides, A., Lampadariou, N., 2004. Deep-sea meiofaunal community structure in the Eastern Mediterranean: are trenches benthic hotspots? Deep-sea research. Part 1. Oceanographic research papers 51, 833-847.

Tselepides, A., Lampadariou, N., Hatziyanni, E., 2004. Distribution of meiobentbos at bathyal depths in the Mediterranean Sea. A comparison between sites of contrasting productivity. Scientia Marina 68, 39-51.

Tubau, X., Canals, M., Lastras, G., Rayo, X., Rivera, J., Amblas, D., 2015. Marine litter on the floor of deep submarine canyons of the Northwestern Mediterranean Sea: The role of hydrodynamic processes. Progress in oceanography 134, 379-403.

Tudela, S., Sardà, F., Maynou, F., Demestre, M., 2003. Influence of submarine canyons on the distribution of the deep-water shrimp, Aristeus antennatus (Risso, 1816) in the NW Mediterranean. Crustaceana 76, 217-225.

Tunnicliffe, V., Juniper, S. K., Sibuet, M., 2003. Reducing environments of the deep-sea floor. Ecosystems of the World, 81-110.

Tyler, P., 1995. Conditions for the existence of life at the deep-sea floor: an update. Oceanography and Marine Biology: An Annual Review.

Tyler, P., Amaro, T., Arzola, R., Cunha, M., de Stigter, H., 2009. Europe's Grand Canyon Nazare Submarine Canyon. Oceanography 22, 46-57.

Tyler, P. A., 2003. Ecosystems of the deep oceans. Elsevier.

Van Dover, C., 2000. The ecology of deep-sea hydrothermal vents. Princeton University Press.

Van Gaever, S., Galeron, J., Sibuet, M., Vanreusel, A., 2009. Deep-sea habitat heterogeneity influence on meiofaunal communities in the Gulf of Guinea. Deep-sea research. Part 2, Topical studies in oceanography 56, 2259-2269.

Vanaverbeke, J., Soetaert, K., Heip, C., Vanreusel, A., 1997. The metazoan meiobenthos along the continental slope of the Goban Spur (NE Atlantic). Journal of Sea Research 38, 93-107, doi:10.1016/s1385-1101(97)00038-5.

Vanhove, S., Arntz, W., Vincx, M., 1999. Comparative study of the nematode communities on the southeastern Weddell Sea shelf and slope (Antarctica). Marine ecology progress series 181, 237-256.

Vanhove, S., Vermeeren, H., Vanreusel, A., 2004. Meiofauna towards the South Sandwich Trench (750–6300m), focus on nematodes. Deep-sea research. Part 2, Topical studies in oceanography 51, 1665-1687.

Vanreusel, A., Vincx, M., Vangansbeke, D., Gijselinck, W., 1992. Structural-analysis of the meiobenthos communities of the shelf-break area in 2 stations of the Gulf-of-Biscay (NE Atlantic). Belgian journal of zoology 122, 185-202.

Vanreusel, A., VandenBossche, I., Thiermann, F., 1997. Free-living marine nematodes from hydrothermal sediments: similarities with communities from diverse reduced habitats. Marine Ecology Progress Series 157, 207-219.

Vanreusel, A., Vincx, M., Schram, D., Van Gansbeke, D., 1995. On the vertical distribution of the metazoan meiofauna in shelf breack and upper slope habitats of the NE Atlantic. Internationale Revue der gesammten Hydrobiologie 80, 313-326.

Vanreusel, A., Fonseca, G., Danovaro, R., da Silva, M., Esteves, A., 2010. The contribution of deep-sea macrohabitat heterogeneity to global nematode diversity. Marine Ecology 31, 6-20.

Vetter, E., Dayton, P., 1998. Macrofaunal communities within and adjacent to a detritus-rich submarine canyon system. Deep Sea Research Part II: Topical Studies in Oceanography 45, 25-54.

Vetter, E., Dayton, P., 1999. Organic enrichment by macrophyte detritus, and abundance patterns of megafaunal populations in submarine canyons. Marine Ecology Progress Series 186, 137-148.

Vetter, E., Smith, C., De Leo, F., 2010. Hawaiian hotspots: enhanced megafaunal abundance and diversity in submarine canyons on the oceanic islands of Hawaii. Marine ecology 31, 183-199.

Vincx, M., 1996. Meiofauna in Marine and Freshwater Sediment. CAB INTERNATIONAL. Methods for the Examination of Organnismal Diversity in Soils ans Sediments.

Vincx, M., Meire, P., Heip, C., 1990. The distribution of nematodes communities in the Southern Bight of the North Sea. Cahiers de biologie marine 31, 107-129.

Vincx, M., Bett, B. J., Dinet, A., Ferrero, T., Gooday, A. J., 1994. Meiobenthos of the deep northeast Atlantic. Advances in Marine Biology 30, 2-88.

Vivier, M. H., 1978. Influence d'un deversement industriel profond sur la nematofaune (Canyon de Cassidaigne, Mediterranee). Téthys 8, 307-321.

Weaver, P. P., Billett, D. S., Boetius, A., Danovaro, R., Freiwald, A., Sibuet, M., 2004. Hotspot ecosystem research on Europe's deep-ocean margins. Oceanography 17, 132-143.

Wefer, G., Billet, D., Hebbeln, D., Jorgensen, B. B., Schlüter, M., Weering, T., 2003. Ocean margin systems. Springer.

Wei, C.-L., Rowe, G., Escobar Briones, E., Boetius, A., Soltwedel, T., Caley, M. J., Soliman, Y., Huettmann, F., Qu, F., Yu, Z., Pitcher, C. R., Haedrich, R., Wicksten, M., Rex, M., Baguley, J., Sharma, J., Danovaro, R., MacDonald, I., Nunnally, C., Deming, J., Montagna, P., Lévesque, M., Weslawski, J., Wlodarska Kowalczuk, M., Ingole, B., Bett, B., Yool, A., Bluhm, B., Iken, K., Narayanaswamy, B., Romanuk, T. N., 2010. Global Patterns and Predictions of Seafloor Biomass Using Random Forests. PLoS One 5, e15323-e15323.

Wieser, W., 1953. Die Beziehung zwischen Mundhöhlengestalt, Ernährungsweise und Vorkommen bei freilebenden marinen Nematoden: eine ökologisch-morphologische Studie.

Wigley, R.L., McIntyre, A.D., 1964. Some quantitative comparisons of offshore meiobenthos and macrobenthos south of Martha's Vineyard. Limnol. Oceanogr. 9, 485–493. doi:10.2307/2833569

Wilson, A. M., Kiriakoulakis, K., Raine, R., Gerritsen, H. D., Blackbird, S., Allcock, A. L., White, M., 2015. Anthropogenic influence on sediment transport in the Whittard Canyon, NE Atlantic. Marine pollution bulletin 101, 320-329.

Würtz, M., 2012. Mediterranean submarine canyons: Ecology and governance. IUCN.

Yamasaki, H., 2016. *Ryuguderes iejimaensis*, a new genus and species of Campyloderidae (Xenosomata: Cyclorhagida: Kinorhyncha) from a submarine cave in the Ryukyu Islands, Japan. Zool Anz A J Comp Zool.

Yıldız, N. Ö., Sørensen, M. V., Karaytuğ, S., 2016. A new species of Cephalorhyncha Adrianov, 1999 (Kinorhyncha: Cyclorhagida) from the Aegean Coast of Turkey. Helgoland Marine Research 70, 24, doi:10.1186/s10152-016-0476-5.

Zelinka, KC., 1896. Demonstration von Tafeln der Echinoderes-Monographie. Verh. Deutsch. Zool. Gesellsch. 6, 197–199.

Zelinka, K., 1894. Über die Organisation von *Echinodere*s. Verh Dtsch Zool Ges 4.

Zelinka, C. 1907. Zur Kenntnis der Echinoderen. Zoologischer Anzeiger 32(5): 130–136.

Zelinka, K., 1928. Monographie der Echinodera. Verlag Wilhelm Engelmann, Leipzig.

Zúñiga, D., Flexas, M. M., Sanchez-Vidal, A., Coenjaerts, J., Calafat, A. i., Jordà, G., García-Orellana, J., Puigdefàbregas, J., Canals, M., Espino, M., Sardà, F., Company, J. B., 2009. Particle fluxes dynamics in Blanes submarine canyon (Northwestern Mediterranean). Progress In Oceanography 82, 239-251, doi:10.1016/j.pocean.2009.07.002.

APPENDIX

APPENDIX 1. Literature review on the main environmental drivers (bold characters) influencing meiofauna diversity, biomass and abundance in submarine canyons. Factors underlying each driver are indicated in italics. Div: diversity. B/A: biomass and abundance. Modified and complete from De Leo (2012).

Environmental driver		**Effects**		**Published literature**	
1) Organic matter supply: quantity and quality		**Div.**	**B/A**	**Article/pub. Year**	**Canyon**
Coastal macrophytic productivity		+	ns	Danovaro et al., 2009	Lacaze-Duthiers
					Cap de Creus
					Adriatic
Increase in POC flux	*Particulate OM serves as food for benthos. Intense organic enrichment causes sediment hypoxia and reduced community complexity and dominant species predominate.*				Cascais
					Nazaré
				Bianchelli et al., 2010	Lacaze-Duthiers
					Cap de Creus
					Adriatic
Distance from shore	*Large distance limit the amount of coastal OM transported to benthic communities being of lesser quality or more degraded when more time is needed for it to Reach the deep canyon part.*				Cascais
					Nazaré
		+	+	Soetaert et al., 1991	Bari
		+	ns	Bianchelli et al., 2013	Lacaze-Duthiers
					Cap de Creus
					Bari
					Samaria
		+	+	Bianchelli et al., 2008	Bari
			+	Rumolo et al., 2015	Buscarró
		-	+	Ingels et al., 2009	Nazaré
				Sotlwedel et al., 2005	Ardencaple
		+	+	Rosli et al., 2016	Pahaua
					Honeycomb

					Campbell
		-	+	Ingels et al., 2011a	Whittard
					Gollum
		-	+	Ingels et al., 2011b	Setubal
					Cascais
		-	-	Romano et al., 2013	Blanes
		ns	-	Ingels et al., 2013	Blanes
		-	-	Ramalho et al., 2014	Nazaré
			-	De Bovèe et al., 1990	Marseille Grand-Rhone Petit Rhone Aude Lacaze-Duthiers
		-	-	Román et al. **(Chapters 3-4-6)**	Blanes
Bioturbation		-	-	Romano et al., 2013	Blanes
2) Physical- Seafloor and Substrate Heterogeneity		Div.	B/A	Article/pub. Year	Canyon
Canyon shape/size topography	*Affects sediment deposition, energy of currents (high/low)* *“U-shaped” canyon can enhance food availability and allows OM retention (depocenter)* *Upper and middle parts may show harsh conditions compared with deeper parts.*	-	+	Leduc et al., 2014	Kaikoura
				De Bovèe et al., 1990	Marseille Grand-Rhone Petit Rhone Aude Lacaze-Duthiers

-	-	Garcia et al., 2007	Nazaré
+	-	Ramalho et al., 2014	Nazaré
*	*	Ingels et al., 2013	Blanes
-	-	Romano et al., 2013	Blanes
+	0	Ingels et al., 2011a	Whittard
			Gollum
		Bianchelli et al., 2008	Bari
+	0	Bianchelli et al., 2010	Lacaze-Duthiers
			Cap de Creus
			Adriatic
			Cascais
			Nazaré
		Leduc et al., 2014	Kaikoura
+	ns	Bianchelli et al., 2013	Lacaze-Duthiers
			Cap de Creus
			Bari
			Samaria
		Román et al. **(Chapters 3-6)**	Blanes

3) Physical- Energy/ levels of Disturbance			Div.	B/A	Article/pub. Year	Canyon
strong winds		*Periocity and intensity of bottom flow regimes affecting sediment depostion and community succesion. High disturbance events can affect sediment slumps and OM inputs, buried and fauna resuspension*	-	-	Pusceddu et al., 2010	La Fonera
wave energy			-	-	Garcia et al., 2007	Nazaré
turbidity flows			-	-	Pusceddu et al., 2013	La Fonera
margin activity			-	-	Ingels et al., 2009	Nazaré
bottom currents			-	-	Ramalho et al., 2014	Nazaré
dense water cascading			-	-	Romano et al., 2013	Blanes
			+	+	Ingels et al., 2011a	Whittard Gollum
			-	-	Van Gaever	Congo
			-	-	**Román et al., 2016**	Blanes
sediment loading		*Sediment loading is observed in shelf-incising canyons providing large and regular supplies of sediment that reach the canyon heads and are transported down-canyon. It is often associated with high OM*	+	+	Bianchelli et al., 2010	Lacaze-Duthiers Cap de Creus
					Román et al. **(Chapters 3-4-5)**	Blanes
bottom trawling	*Direct effect*	*Sediment resuspension and impoverishment burial of fauna.*	-	-	Pusceddu et al., 2014	La Fonera
			-	-	Rosli et al., 2016	Ardencaple Pahaua Honeycomb Campbell
	Indirect effect	*Anthropogenic depocenter, increased it OM*	0	+	Román et al. **Chapter 3-4**	Blanes

APPENDIX 2: Taxonomic account

PHYLUM NEMATODE Potts, 1932
Class ENOPLEA Inglis, 1983
Subclass ENOPLIA Pearse, 1942

Order Enoplida Filipjev, 1929

Suborder Enoplina Chitwood & Chitwood, 1937

Family Thoracostomopsidae Filipjev, 1927

Enoploides Ssaweljev, 1912

Paramesacanthion Wieser, 1953

Family Anoplostomatidae Gerlach & Riemann, 1974

Anoplostoma Bütschli, 1874

Family Phanodermatidae Filipjev, 1927

Crenopharynx Filipjev, 1934

Phanodermopsis Ditlevsen, 1926

Family Anticomidae Filipjev, 1928

Anticoma Bastian, 1865

Odontanticoma Platonova, 1976

Suborder Oncholaimina De Ley & Blaxter, 2002

Family Oncholaimidae Filipjev, 1916

Viscosia de Man, 1890

Family Enchelidiidae Filipjev, 1918

Bathyeurystomina Lambshead & Platt, 1979

Ledovitia Filipjev, 1927

Symplocostoma Bastian, 1865

Suborder Ironina Siddiqi, 1983

Family Ironidae de Man, 1876

Dolicholaimus de Man, 1888

- *Syringolaimus* de Man, 1888
- *Thalassinorus* de Man, 1889

Family Leptosomatidae Filipjev, 1916

- *Cylicolaimus* de Man, 1889

Family Oxystominidae Chitwood, 1935

- *Cricohalalaimus* Bussau, 1993
- *Halalaimus* de Man, 1888
- *Litinium* Cobb, 1920
- *Oxystomina* Filipjev, 1921
- *Thalassoalaimus* de Man, 1893
- *Wieseria* Gerlach, 1956

Order Triplonchida Cobb, 1919

Suborder Tobrilina Tsalolikhin, 1976

Family Rabdodemaniidae Filipjev, 1921

- *Rhabdodemania* Baylis & Daubney, 1926

Class CHROMADOREA

Subclass CHROMADORIA

Order Chromadorida Chitwood, 1933

Suborder Chromadorina Filipjev, 1929

Family Chromadoridae Filipjev, 1917

- *Acantholaimus* Allgén, 1933
- *Actinonema* Cobb, 1920
- *Chromadorella* Filipjev, 1918
- *Chromadorita* Filipjev, 1922
- *Dichromadora* Kreis, 1929
- *Neochromadora* Micoletzky, 1924

Prochromadorella Micoletzky, 1924

Family Neotonchidae Wieser & Hopper, 1966

Nannolaimus Cobb, 1920

Neotonchus Cobb, 1933

Family Cyatholaimidae Filipjev, 1918

Longicyatholaimus Micoletzky, 1924

Marylynnia Hopper, 1977

Metacyatholaimus Stekhoven, 1942

Minolaimus Vitiello, 1970

Paralongicyatholaimus Stekhoven, 1950

Pomponema Cobb, 1917

Family Selachinematidae Cobb, 1915

Choanolaimus de Man, 1880

Gammanema Cobb, 1920

Halichoanolaimus de Man, 1886

Richtersia Steiner, 1916

Synonchiella Cobb, 1933

Order Desmodorida De Coninck, 1965

Suborder Desmodorina De Connick, 1965

Family Desmodoridae Filipjev, 1922

Desmodora de Man, 1889

Desmodorella Cobb, 1933

Molgolaimus Ditlevsen, 1918

Paradesmodora Schuumans Stekhoven, 1950

Familiy Microlaimidae Micoletzky, 1922

Aponema Jensen, 1978

Microlaimus de Man, 1880

Order Desmoscolecida Filipjev, 1929

Family Cyatonematidae Tchesunov, 1990

Cyartonema Cobb, 1920

Family Desmoscolecidae Shipley, 1896

Desmoscolex Claparède, 1863

Greeffiella Cobb, 1922

Tricoma Cobb, 1894

Order Monhysterida Filipjev, 1929

Suborder Monhysterina De Coninck & Schuurmans Stekhoven, 1933

Family Monhysteridae de Man, 1876

Monhystrella Cobb, 1918

Thalassomonhystera Jacobs, 1987

Family Sphaerolaimidae Filipjev, 1918

Metasphaerolaimus Gourbault & Boucher, 1982

Sphaerolaimus Bastian, 1865

Subsphaerolaimus Lorenzen, 1978

Family Xyalidae Chitwood, 1951

Ammotheristus Lorenzen, 1977

Amphimonhystrella Timm, 1961

Capsula Busau, 1993

Daptonema Cobb, 1920

Elzalia Gerlach, 1957

Gnomoxyala Lorenzen, 1977

Linhystera Juario, 1974

Manganonema Bussau, 1993

Metadesmolaimus Schuurmars Stekhoven, 1935

Paramonohystera Steiner, 1916

Paramphimonhystrella Huang & Zhang, 2006

Promonhystera Wieser, 1956

Retrotheristus Lorenzen, 1977

Steineria Micoletzky, 1922

Theristus Bastian, 1865

Suborder Linhomoeina Andrássy, 1974

Family Linhomoeidae Filipjev, 1922

Desmolaimus de Man, 1880

Didelta Cobb, 1920

Disconema Filipjev, 1918

Eleutherolaimus Filipjev, 1922

Linhomoeus Bastian, 1865

Megadesmolaimus Wieser, 1954

Metalinhomoeus de Man, 1907

Paralinhomoeus de Man, 1907

Terschellingia de Man, 1888

Order Araeolaimida De Coninck & Schuurmans Stekhoven, 1933

Family Axonolaimidae Filipjev, 1918

Axonolaimus de Man, 1889

Odontophora Bütschli, 1874

Family Comesomatidae Filipjev, 1918

Cervonema Wieser, 1954

Comesoma Bastian, 1865

Dorylaimopsis Ditlevsen, 1918

Hopperia Vitiello, 1969

Laimella Cobb, 1920

Metacomesoma Wieser, 1954

Pierrickia Vitiello, 1970

Sabatieria Rouville, 1903

Setosabatieria Platt, 1985

Vasostoma Wieser, 1954

Family Coninckiidae Lorenzen, 1981

Coninckia Gerlach, 1956

Family Diplopeltidae Filipjev, 1918

Campylaimus Cobb, 1920

Diplopeltula Gerlach, 1950

Sourtheniella Allgén, 1932

Family Siphonolaimidae Filipjev, 1918

Astomonema Ott, Rieger, Rieger & Enderes, 1982

Order Plectida Malakhov, 1982

Family Leptolaimidae Örley, 1880

Antomicron Cobb, 1920

Leptolaimoides Vitiello, 1971

Family Camacolaimidae Mocoletzky, 1924

Alaimella Cobb, 1920

Deodontolaimus de Man, 1889

Procamacolaimus Gerlach, 1954

Family Ceramonematodae Cobb, 1933

Ceramonema Cobb, 1920

Metadasynemella De Connick, 1942

Pselionema Cobb, 1933

Family Diplopeltoididae Thesunov, 1990

Diplopeltoides Gerlach, 1962

Family Aegiolaimidae Lorenzen, 1981

Aegialoalaimus de Man, 1907

Used sources

Bain, O., Baldwin, J. G., Beveridge, I., Bezerra, T. C., Braeckman, U., Coomans, A., Decraemer, W., Derycke, S., Durette-Desset, M.-C., Fonseca, G., 2014. Nematoda. Walter de Gruyter.

De Ley, P., Decraemer, W., Eyualem, A., 2006. Introduction: summary of present knowledge and research addressing the ecology and taxonomy of freshwater nematodes. Freshwater tematodes: taxonomy and ecology. CABI, pp. 3-30.

Fonseca, G., Decraemer, W., 2008. State of the art of the free-living marine Monhysteridae (Nematoda). Journal of the Marine Biological Association of the UK 88, 1371-1390.

Vanaverbeke, J., Bezerra, T., Braeckman, U., De Groote, A., De Meester, N., Deprez, T., Derycke, S., Gilarte, P., Guilini, K., Hauquier, F., 2014. NeMys: world database of free-living marine nematodes. World Wide Web Electron. Publ. Accesse at http://nemys.ugent.be.

WORMS Editorial Board, 2015. World Register of Marine Species. Available from http://www.marinespecies.org at VLIZ.

APPENDIX 3. List of identified genera and families in the **Chapter 4,** with trophic group (FT) classification according to the definitions of Wieser (1953). 1A: selective deposit feeder, 1B: non-selective deposit feeder, 2A: epistratum feeder, 2B: predator/scavenger, 3: chemosynthetic, symbiosis with sulphur oxidising endosymbiotic bacteria in the gut.

Genus	**FT**	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**BC2000**
Acantholaimus	2A	*	*	*	*	*	*
Actinonema	2A	*	*	*	*	*	*
Aegialoalaimus	1A	*	*	*		*	*
aff. Ammontheristus	1B					*	
aff. Crenopharynx	1A			*	*		*
aff. Daptonema	1B	*	*	*	*		
aff. Metadesmolaimus	1B			*			*
aff. Odontanticoma	2A					*	*
aff. Odontophora	2A	*					
aff. Phanodermella	1A					*	
aff. Phanodermopsis	1A						*
aff. Viscosia	2B	*					
Alaimella	1A			*	*		*
Amphimonhystrella	1B	*	*	*	*	*	*
Anoplostoma	1B						*
Anticoma	1A	*				*	
Antomicron	1A		*		*	*	*
Aponema	2A		*	*	*		*
Astomonema	3	*	*	*	*	*	
Axonolaimus	1B			*	*		
Bathyeurystomina	2B		*	*	*	*	*
Campylaimus	1B	*	*	*	*	*	*
Capsula	1B				*		
Ceramonema	1A			*			*
Cervonema	1B	*	*	*	*	*	*
Choanolaimus	2B						*
Chromadorella	2A	*	*	*		*	*
Chromadoridae	2A	*	*	*	*	*	*
Chromadorita	2A	*	*	*			*
Comesomatidae	1B	*	*	*	*	*	*
Coninckia	1A	*					
Crenopharynx	1A				*		

Genus	**FT**	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**BC2000**
Cyatholaimidae	2A	*	*	*			
Cylicolaimus	2B			*			
Daptonema	1B	*	*	*	*	*	*
Desmodora	2A	*		*		*	*
Desmodorella	2A	*	*	*	*		
Desmolaimus	1B						*
Desmoscolex	1A	*	*		*	*	*
Dichromadora	2A	*	*	*	*	*	*
Didelta	1B					*	
Diplopeltoides	1A			*			*
Diplopeltula	1A	*	*	*	*	*	*
Disconema	1A	*	*	*	*	*	*
Dorylaimopsis	2A	*	*	*	*	*	*
Eleutherolaimus	1B	*					
Elzalia	1B	*	*	*	*	*	*
Enoploides	2B					*	
Ethomolaimidae	1A	*	*		*		
Gammanema	2B				*	*	
Gnomoxyala	1B	*			*		
Greeffiella	1A	*	*		*		
Halalaimus	1A	*	*	*	*	*	*
Halichoanolaimus	2B		*	*	*	*	*
Hopperia	2A	*		*	*	*	*
Laimella	2A	*	*	*	*	*	*
Ledovitia	2B		*		*	*	
Leptolaimidae	1A			*	*		
Leptolaimoides	1A	*	*			*	*
Leptolaimus	1A	*	*	*	*	*	*
Linhomoeus	2A	*	*			*	*
Linhomoeidae	2A	*	*		*		*
Litinium	1A	*			*	*	*
Longicyatholaimus	2A					*	*
Marylynnia	2A	*	*	*	*	*	*
Megadesmolaimus	1B	*				*	*
Metacyatholaimus	2A	*	*	*			
Metadasynemella	1A		*	*		*	*
Metadesmolaimus	1B	*		*	*	*	*
Metalinhomoeus	1B	*	*	*	*	*	*

Genus	**FT**	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**BC2000**
Metasphaerolaimus	2B	*	*	*	*	*	*
Microlaimidae	2A		*				
Microlaimus	2A		*	*	*	*	*
Minolaimus	1A				*	*	*
Molgolaimidae	1A						*
Molgolaimus	1A	*	*	*	*	*	*
Monhysteridae	1B	*	*	*	*		*
Monhystrella	1B	*	*	*	*	*	*
Nannolaimus	1A	*	*	*		*	
Neochromadora	2A						*
Neotonchus	2A	*	*	*	*		*
Odontophora	2A		*	*			
Odotanticoma	2A						*
Oxystomina	1A	*	*	*	*	*	*
Paradesmodora	2A	*	*				
Paralinhomoeus	1B	*					
Paralongicyatholaimus	2A	*	*	*	*	*	*
Paramesacanthion	2B	*	*	*	*	*	*
Paramonohystera	1B	*	*	*	*	*	*
Paramphimonhystrella	1B		*	*	*	*	*
Phanodermopsis	2A				*		
Pierrickia	1B		*			*	
Pomponema	2B	*	*	*	*	*	*
Procamacolaimus	2A						*
Prochromadorella	2A	*	*	*	*		*
Promonhystera	1B	*			*		
Pselionema	1A	*	*	*	*	*	*
Retotheristus	1B	*					
Rhabdodemania	1B	*					
Richtersia	2B	*	*	*			
Sabatieria	1B	*	*	*	*	*	*
Setosabatieria	1B	*	*	*	*	*	
Sourtheniella	1A	*	*			*	*
Sphaerolaimidae	2B	*					
Sphaerolaimus	2B	*	*	*	*	*	*
Steineria	1B	*					
Subsphaerolaimus	1B	*	*				
Symplocostoma	2B					*	

Genus	**FT**	**BC500**	**BC900**	**BC1200**	**BC1500**	**BC1750**	**BC2000**
Synonchiella	2B	*					
Syringolaimus	2B						*
Terschellingia	1A			*	*	*	
Thalassomonhystera	1B	*	*	*	*	*	*
Theristus	2A		*				
Tricoma	1A	*	*	*	*	*	*
Vasostoma	2A	*	*	*		*	*
Viscosia	2B	*	*	*	*		
Wieseria	1A					*	*
Xyalidae	1B	*	*	*	*	*	*

Appendix 4. List of identified genera and families in the study stations of **Chapter 5**, with trophic group (FT) classification according to the definitions of Wieser (1953).

Genus	**FT**	**BC1500**	**BC1750**	**BC2000**	**OS1500**	**OS1750**	**OS2000**
Acantholaimus	2A	*	*	*	*	*	*
Actinonema	2A	*	*	*	*	*	*
Aegialoalaimus	1A		*	*	*		
aff. Ammontheristus	1B		*				
aff. Anoplostoma	1A					*	
aff. Crenopharynx	1B	*		*	*	*	*
aff. Daptonema	1B	*					
aff. Filoncholaimus	2B				*		
aff. Marylynnia	2A					*	
aff. Metacyatholaimus	2A					*	
aff. Metadesmolaimus	1B			*			
aff. Odontanticoma	2A		*	*			*
aff. Phanodermella	2A		*				
aff. Phanodermopsis	1A			*	*		
aff. Rhabdocoma	1A					*	
Alaimella	1A	*		*			
Ammontheristus	2A				*		
Amphimonhystrella	1B	*	*	*	*	*	*
Anoplostoma	1B			*			
Anticoma	1A		*				
Anticomidae indet.	2A						*
Antomicron	1A	*	*	*	*	*	
Aponema	2A	*		*	*		
Astomonema	3	*	*				
Axonolaimus	1B	*					
Bathyeurystomina	2B	*	*	*			
Campylaimus	1B	*	*	*		*	
Capsula	1B	*					
Ceramonema	1A			*			*
Cervonema	1B	*	*	*	*	*	*
Choanolaimus	2B			*			
Chromadorella	2A		*	*	*	*	*
Chromadoridae indet.	2A	*	*	*	*	*	
Chromadorita	2A			*		*	

Genus	**FT**	**BC1500**	**BC1750**	**BC2000**	**OS1500**	**OS1750**	**OS2000**
Comesomatidae indet.	1B	*	*	*		*	
Crenopharynx	1A	*					
Cricohalalaimus	1A					*	*
Cyartonema	1A				*		
Cyatholaimudae indet.	2A				*		*
Daptonema	1B	*	*	*	*	*	*
Deontolaimus	2A				*		
Desmodora	2A		*	*	*	*	*
Desmodorella	2A	*					
Desmodoridae indet.	2A				*		
Desmolaimus	1B			*			
Desmoscolex	1A	*	*	*	*	*	
Dichromadora	2A	*	*	*	*	*	*
Didelta	1B		*				
Diplopeltidae indet.	1A					*	
Diplopeltoides	1A			*			
Diplopeltula	1A	*	*	*	*	*	*
Disconema	2A	*	*	*		*	
Dolicholaimus	2B					*	
Dorylaimopsis	2A	*	*	*			
Elzalia	1B	*	*	*	*	*	
Encheliidae indet.	2B				*		
Enoploides	2B		*				
Ethomolaimidae indet.	1A	*					
Gammanema	2B	*	*				
Gnomoxyala	1B	*					
Greeffiella	1A	*				*	
Halalaimus	1A	*	*	*	*	*	*
Halichoanolaimus	2B	*	*	*	*	*	*
Hopperia	2A	*	*	*	*	*	*
Laimella	2A	*	*	*	*		
Ledovitia	2B	*	*		*		
Leptolaimudae indet.	1A	*			*	*	
Leptolaimoides	1A		*	*		*	
Leptolaimus	1A	*	*	*	*	*	*
Linhomoeus	2A		*	*		*	

Genus	**FT**	**BC1500**	**BC1750**	**BC2000**	**OS1500**	**OS1750**	**OS2000**
Litinium	1A	*	*	*	*	*	*
Longicyatholaimus	2A		*	*	*	*	*
Manganonema	1A				*		
Marylynnia	2A	*	*	*	*	*	*
Megadesmolaimus	1B	*	*	*			
Metacomesoma	1A					*	
Metacyatolaimus	2A				*	*	
Metadasynemella	1A		*	*			
Metadesmolaimus	1B	*	*	*			
Metalinhomoeus	1B	*	*	*			
Metasphaerolaimus	2B	*	*	*	*	*	*
Microlaimidae indet.	2A	*			*	*	
Microlaimus	2A		*	*	*	*	
Minolaimus	1A	*	*	*		*	*
Molgolaimidae indet.	1A			*			
Molgolaimus	2A	*	*	*	*	*	
Monhysteriade indet.	1B	*		*	*	*	*
Monhystrella	1B	*	*	*	*	*	*
Nannolaimus	1A		*				
Neochromadora	2A			*	*	*	
Neotonchus	2A	*		*			
Odotanticoma	2A			*			
Oxystomina	1A	*	*	*	*	*	*
Paracanthonchus	2A	*					*
Paralongicyatholaimus	2A	*	*	*	*	*	*
Paramesacanthion	2B	*	*	*			*
Paramonohystera	1B	*	*	*	*	*	*
Paramphimonhystrella	1B	*	*	*	*		*
Phanodermatidae indet.	1A				*	*	
Phanodermopsis	2A	*					
Pierrickia	1B		*				
Pomponema	2B	*	*	*	*		
Procamacolaimus	2A	*		*			
Prochromadorella	2A	*		*	*	*	
Promonhystera	1B	*					
Pselionema	1A	*	*	*	*	*	*
Richtersia	2B	*			*	*	*

Genus	**FT**	**BC1500**	**BC1750**	**BC2000**	**OS1500**	**OS1750**	**OS2000**
Sabatieria	1B	*	*	*	*	*	*
Setosabatieria	1B	*	*				
Sourtheniella	1A	*	*	*	*	*	
Sphaerolaimus	2B	*	*	*	*	*	*
Symplocostoma	2B	*	*			*	*
Syringolaimus	2B			*	*	*	*
Tarvaia	1B						*
Terschellingia	1A	*	*				
Thalassinorus	2B					*	
Thalassoalaimus	1A				*		
Thalassomonhystera	1B	*	*	*	*	*	*
Theristus	2A						*
Tricoma	1A	*	*	*	*	*	*
Vasostoma	2A	*	*	*			*
Viscosia	2B	*					
Wieseria	1A		*	*	*	*	
Xyalidae indet.	1B	*	*	*	*	*	*

APPENDIX 5. Preliminary analysis on the relationships between temporal and spatial variation of the nematode assemblages from Blanes Canyon

INTRODUCTION

Ecological studies regarding deep-sea free-living nematodes usually address spatial patterns, and these include canyon habitats. Consequently, the temporal patterns of meiofauna communities from submarine canyons are almost lacking (Glover et al., 2010). A pioneer study carried out by Ramalho et al., (2014) addressed assessed the first inter-annual study dataset on meiofaunal communities, with special focus on nematodes, from Nazaré Canyon (Western Iberian margin). As previously mentioned in Chapters 1, 3-5, Ingels et al. (2013) studied the seasonality of the Blanes Canyon nematode standing stocks, feeding types and gender life stage ratios, and did not reported overall differences between canyon and slope stations. However, sampling time played a significant role in explaining the standing stocks.

Several environmental drivers (e.g., surface productivity, seasonal events like storms or DSWC episodes), but also the indirect effects of bottom trawling, may contribute to regulate the meiofauna distribution in the Blanes Canyon system. However, there is still a lack of information about the temporal variation, particularly on the nematode assemblages in terms of community and structural diversity. Indeed, **Chapter 4** results strongly support that trawling, may also influence the nematode communities, particularly at BC1200.

Accordingly, we include in this thesis some preliminary first results on the temporal variation of nematode assemblages, which allowed assessing the following questions:

- Is there any detectable difference in community structure and structural diversity among the studied periods?

- Are the different regions of the canyon affected in a similar way through time?

MATERIAL AND METHODS

This preliminary study was carried out based on samples from 500, 1,200, 1,500 and 1,750 m depth, collected over all DOS MARES cruises at the Blanes Canyon (see section **2.2** in **Chapter 2** for further details). All these samples were processed by the LUDOX procedure used to extract the whole meiofauna from the sediment (see section **2.3.2** in **Chapter 2**).

To assess the temporal variability, the nematodes from the 0-1 cm sediment layer (i.e. the layer showing the highest diversity) were identified. Nematodes were picked out randomly (100-150 individuals), mounted on permanent glycerine slides after a stepwise dehydration in a graded series of ethanol (see section **2.3.3** in **Chapter 2**), and identified down to genus level using pictorial keys (Platt and Warwick. 1988) and the online identification keys/literature available at the Nemys Database (Guilini et al., 2016).

Data analyses

The significance of the differences in nematode structural diversity (as genera richness, expected number of genera (*EG*(51)), and Shannon index) and community structure between sampling cruises and water depths were analysed by means of a two-way crossed design (factors: cruise (fixed) and water depth (fixed)) in uni- or multivariate analysis of variance by permutation (PERMANOVA, Anderson, 2005; Anderson et al., 2008), based on the Euclidean distance matrices. When PERMANOVA permutations numbers were limited to <100, Monte Carlo values were used to infer significance. The data were *a priori* standardized.

Temporal and spatial patterns based on nematode community structure were assessed by a non-Metrical Multidimensional Scaling (nMDS) plot based on Bray-Curtis similarity resemblance matrix. The relative contribution of each genus to the (dis)similarities between cruises and water depths were assessed by SIMPER analyses.

RESULTS

A total of 112 genera were recorded during the four cruises, 18 of them being newly recorded from the canyon. Among them, 5 were previously encountered on the slope in **Chapter 5** (Table A1). Diversity at BC500 was higher in spring, while there was no clear tendency in autumn (Fig. A1). Accordingly, no clear seasonal pattern was detected by the PERMANOVA, where Tm x WD was only significantly different for the genus richness (Table A 2). Pair-wise comparisons indicated that genus richness significantly differed at BC1500 in autumn (2012 *vs.* 2013) and between autumn 2012 and spring 2013.

The most common genera found in all stations were *Halalaimus* (11.2 ± 2.4%), *Cervonema* (6 ± 4.8%), *Molgolaimus* (5.8 ± 3.1%), *Tricoma* (5.7 ± 1.1%) and *Sabatieria* (5.0 ± 2.6%) (Table A3). Overall, *Cervonema* was dominant in upper canyon stations (BC500 and BC1200), which was in consonance with the previous observations in **Chapter 4**. A similarly habitat preference was observed for *Acantholaimus*, but the dominance of this genus apparently continued in deeper stations, particularly at BC1750. Contrastingly, *Molgolaimus*, which appeared to be particularly dominant in the canyon only at 1,750 m depth (**Chapter 4**), revealed to occur also at BC500, BC1200 and BC1500 (Table A3). The patterns of dominance of this genus, however, are still not clear, its presence over time being probably related with the existence of intermittent food inputs in the Canyon (see **Chapter 3**).

Table A1. New nematode genus recorded at Blanes Canyon, including those previously reported on the slope in **Chapter 5**.

New record	Previously cited (**Chapter 5**)
Aff. *Amphimonhystera*	Aff. *Marylynnia*
Aff. *Eleutherolaimus*	*Cricohalalaimus*
Aff. *Paracyatholaimus*	*Cyatonema*
Aff. *Plectus*	*Manganonema*
Aff. *Sourtheniella*	*Paracyatholaimus*
Aff. *Spirobolbolaimus*	
Aff. *Stylotheristus*	
Bolbolaimus	
Comesoma	
Linhystera	
Paracanthonchus	
Paramesonchium	
Parasphaerolaimus	

There were significant differences in nematode communities between sampling periods and water depths, but also for the interaction term (PERMANOVA, Table A2). However, water depth seemed to be more important than sampling periods, as indicated by the estimated component of variation. Subsequent pair-wise comparisons did not show significant differences within each station over time. Overall, these existing patterns are better represented in the nMDS plot (Fig. A2), which revealed clear bathymetric differences overtime in the nematode community composition of the upper (BC500 and BC1200) *vs.* the mid and mid-deep (BC1500 and BC1750) stations. Upper stations were highly variable and, thus, no clear tends could be identified, while there were small variations at BC1500 and BC1750, with the main variation observed between spring 2013 and the rest of sampling periods

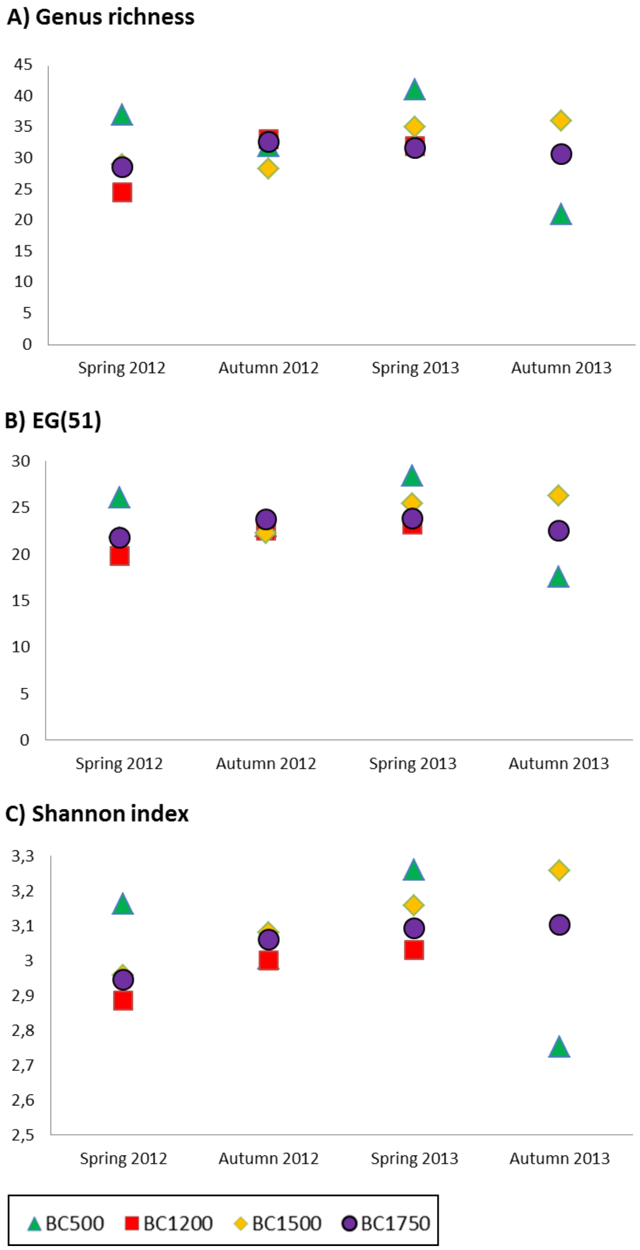


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Figure A1. Nematode structural diversity index per each sampling period (spring and autumn 2012 and 2012) and station (BC500, BC1200, BC1500 and BC1750).

Table A2. Results of the two-way PERMANOVA (factor “cruise” and “water depth” with 4 levels each, both fixed) for the nematode descriptors (i.e., genera richness, expected number of genera (EG(51), Shannon index and community structure) between sampling periods (spring and autumn 2012 and spring and autumn 2013). Bold *p* values indicated significance at the < 0.05 level; df: degrees of freedom; SS: sum of squares; MS: mean squares; Pseudo-F: pseudo-*F* ratio; *p*(Perm): permutation *p-value*; Perms: permutations; ECV: estimated component of variation.

Source	**df**	**SS**	**MS**	**Pseudo-F**	***p*(perm)**	**Unique perms**	**ECV**
Genera richness							
Cruise	3	182.41	60.803	2.7887	0.066	9956	4.6442
Water depth	3	55.882	18.627	0.85435	0.484	9957	-0.3771
Cr x WD	8	588.85	73.607	3.376	**0.0119**	9938	21.764
Residual	22	479.67	21.803				21.803
Total	36	1223.6					
EG (51)							
Cruise	3	55.824	18.608	2.1457	0.13	9998	11.832
Water depth	3	28.916	9.6387	1.1114	0.373	9989	0.11476
Cr x WD	8	157.17	19.646	2.2653	0.065	9978	46.102
Residual	22	190.79	8.6723				86.723
Total	36	410.46					
Shannon index							
Cruise	3	9.93E-02	3.31E-02	0.95235	0.402	9989	-1.97E-04
Water depth	3	9.20E-02	3.07E-02	0.88173	0.472	9998	-4.88E-04
Cr x WD	8	0.41483	5.19E-02	1.4915	0.202	9997	7.18E-03
Residual	22	0.76486	3.48E-02				3.48E-02
Total	36	1.3813					
Community							
Cruise	3	5359.5	1786.5	1.7107	***0.0051***	9865	88.38
Water depth	3	10789	3596.4	3.4437	***0.0001***	9893	303.05
Cr x WD	8	13491	1686.3	1.6148	***0.0009***	9832	250.72
Residual	22	22975	1044.3				1044.3
Total	36	53438					

Table A3. Mean relative abundance of the nematode genera (≥ 3%) per sampling period and water depth.

Spring 2012

BC500	%	BC1200	%	BC1500	%	BC1750	%
Cervonema	13.1	*Halalaimus*	12.5	*Halalaimus*	18.9	*Molgolaimus*	15.8
Halalaimus	11.9	*Tricoma*	7.8	*Molgolaimus*	6.5	*Halalaimus*	8.9
Pomponema	5.9	*Cervonema*	7.6	*Amphimonhystrella*	5.8	*Marylynnia*	8.4
Setosabatieria	5.5	*Actinonema*	5.7	*Leptolaimus*	5.6	*Tricoma*	8.3
Tricoma	5.5	*Daptonema*	4.8	*Desmoscolex*	5.5	*Acantholaimus*	6.2
Sabatieria	3.8	*Monhysteridae*	4.4	*Campylaimus*	5.2	*Oxystomina*	4.7
Prochromadorella	3.4	*Molgolaimus*	3.9	*Sabatieria*	5.0	*Leptolaimus*	4.3
Leptolaimus	3.4	*Acantholaimus*	3.4	*Acantholaimus*	5.0	*Paramesacanthion*	4.0
Desmoscolex	3.4	*Sabatieria*	3.2	*Tricoma*	4.7	*Diplopeltula*	4.0
Metasphaerolaimus	3.4			*Monhystrella*	3.6	*Amphimonhystrella*	3.6
Sphaerolaimus	3.0			*Diplopeltula*	3.2	*Sabatieria*	3.2

Autumn 2012

BC500	%	BC1200	%	BC1500	%	BC1750	%
Cervonema	13.1	*Halalaimus*	19.4	*Halalaimus*	9.9	*Molgolaimus*	11.4
Sabatieria	6.5	*Actinonema*	6.5	*Pomponema*	7.4	*Halalaimus*	10.2
Halalaimus	6.4	*Dichromadora*	6.5	*Acantholaimus*	6.1	*Acantholaimus*	9.8
Sphaerolaimus	6.2	*Cervonema*	5.3	*Sphaerolaimus*	5.9	*Tricoma*	7.8
Setosabatieria	5.6	*Pomponema*	5.1	*Oxystomina*	5.9	*Diplopeltula*	6.3
Daptonema	4.1	*Campylaimus*	4.6	*Tricoma*	4.8	*Amphimonhystrella*	4.9
Pomponema	4.0	*Daptonema*	4.0	*Daptonema*	4.8	*Desmoscolex*	4.9
Chromadorella	4.0	*Diplopeltula*	3.3	*Actinonema*	4.5	*Paramesacanthion*	3.5
Actinonema	3.4			*Sabatieria*	3.9	*Metasphaerolaimus*	3.5
Elzalia	3.2			*Disconema*	3.7	*Marylynnia*	3.4
Metacyatholaimus	2.9			*Molgolaimus*	3.6	*Sphaerolaimus*	3.2
				Pselionema	3.2	*Pselionema*	3.0
				Diplopeltula	3.0		
				Cervonema	3.0		

Table A3. *Continued*

Spring 2013

BC500	%	BC1200	%	BC1500	%	BC1750	%
Halalaimus	16.4	*Molgolaimus*	10.3	*Halalaimus*	11.4	*Sabatieria*	8.2
Daptonema	7.5	*Halalaimus*	9.6	*Sabatieria*	11.3	*Acantholaimus*	6.6
Sabatieria	6.0	*Tricoma*	8.3	*Sphaerolaimus*	6.9	*Marylynnia*	6.1
Cervonema	5.2	*Cervonema*	8.3	*Campylaimus*	5.5	*Antomicron*	5.6
Leptolaimus	5.2	*Microlaimus*	4.7	*Pselionema*	4.6	*Pselionema*	5.6
Tricoma	5.2	*Actinonema*	3.0	*Tricoma*	4.6	*Molgolaimus*	5.2
Pomponema	3.7			*Metasphaerolaimus*	3.2	*Leptolaimus*	5.1
Diplopeltula	3.0			*Diplopeltula*	3.2	*Tricoma*	4.9
Elzalia	3.0					*Halalaimus*	4.6
Sphaerolaimus	3.0					*Campylaimus*	4.1
						Diplopeltula	4.0
						Sphaerolaimus	3.3
						Leptolaimoides	3.1

Autumn 2013

BC500	%	BC1500	%	BC1750	%
Desmoscolex	15.2	*Sabatieria*	9.1	*Halalaimus*	9.5
Molgolaimus	8.8	*Halalaimus*	7.9	*Amphimonhystrella*	7.3
Tricoma	6.8	*Molgolaimus*	5.6	*Sabatieria*	6.2
Acantholaimus	7.3	*Desmoscolex*	5.1	*Tricoma*	5.6
Dichromadora	5.7	*Leptolaimus*	5.0	*Acantholaimus*	4.8
Leptolaimus	4.9	*Tricoma*	4.6	*Daptonema*	4.2
Elzalia	4.9	*Diplopeltula*	4.3	*Paramesacanthion*	4.2
Halalaimus	4.8	*Sphaerolaimus*	3.5	*Diplopeltula*	4.0
Cervonema	4.4	*Pselionema*	3.2	*Molgolaimus*	3.8
Greeffiella	4.3	*Campylaimus*	3.1	*Marylynnia*	3.4
Paramesacanthion	3.4			*Gnomoxyala*	3.2

The nematode communities seemed to be less heterogeneous in the spring periods than in the autumn ones, causing the stations sampled during the former to be less disperse in the nMDS plot (Fig. A2), likely in relation with the high surface productivity during the two spring seasons (see figure 3.8, **Chapter 3**).

The SIMPER analysis showed a maximum dissimilarity between BC500 and BC1750 assemblages (~60%), mainly through the presence of *Molgolaimus* (10% and 8.5%) and *Cervonema* (6% and 8.5%) in 2012, and by *Halalaimus* (10.8%) and *Daptonema* (5.9%), and *Desmoscolex* (13%) and *Amphymonhystrella* (6.7%) in spring and autumn 2013, respectively.

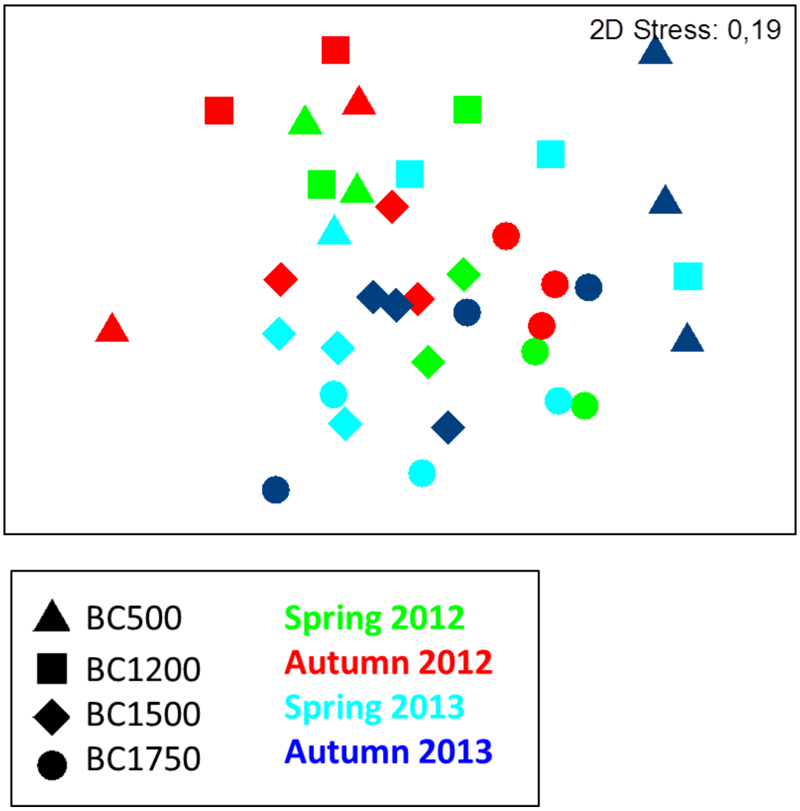

Figure A2. Non-metric MDS (nMDS) plot based on standarized nematode genera relative abundance data and Bray Curtis similarity resemblance for each sampling period (spring and autumn 2012 and 2012) and station (BC500, BC1200, BC1500 and BC1750).

This is certainly a work still in progress. Potential environmental drivers, but also other nematode descriptors (e.g., density, biomass, functional diversity, gender/life-stage ratios) and their variability along the vertical sediment profile have to be further assessed. Based on the present preliminary findings, we could state that the meiofaunal communities from Blanes Canyon showed temporal trends during the study period, which were more evident at the upper region. However, the differences between stations seemed to be much higher than the temporal variability. Therefore, we suggest that the influence of the topographic heterogeneity of the canyon along the bathymetric gradient, combined with a differential influence of the hydrodynamic disturbances, seemed to over impose on the seasonally driving effects of food availability on the nematode assemblages at Blanes Canyon.