

Seasonal variation of the stable C and N isotopic composition of the mesophotic black coral *Antipathella subpinnata* (Ellis & Solander, 1786)

Martina Coppari^{a,*}, Christine Ferrier-Pagès^b, Michela Castellano^a, Francesco Massa^a, Enrico Olivari^a, Giorgio Bavestrello^a, Paolo Povero^a, Marzia Bo^a

^a Dipartimento di Scienze della Terra, dell'Ambiente e della Vita, Università di Genova, Corso Europa 26, 16132, Genova, Italy

^b Centre Scientifique de Monaco, Coral Ecophysiology Team, 8 Quai Antoine 1er, 98000, Monaco, Monaco

ARTICLE INFO

Keywords:

Antipatharians
Antipathella subpinnata
Stable isotopes
Trophic ecology

ABSTRACT

Trophic relationships significantly influence ecosystem functioning, ultimately affecting populations' abundance, size structure and distribution. There is still a substantial knowledge gap on the trophic ecology of deep-sea organisms, particularly those living in the mesophotic depths (30–150 m), despite their crucial importance as ecosystem engineers. The trophic ecology of the most common mesophotic black coral species of the Mediterranean Sea, *Antipathella subpinnata* (Ellis & Solander, 1786), was studied by means of stable isotopes analysis in two seasons. The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ analysis of the tissue of *A. subpinnata*, of the sediment, as well as the main planktonic fractions (pico-nanoplankton, microplankton, and mesozooplankton) indicate a diet based on pico-nanoplankton in autumn and mesozooplankton in spring. We calculated a trophic enrichment of 1‰ in $\delta^{13}\text{C}$ and of 2.5‰ $\delta^{15}\text{N}$ between prey and predator. Such information contributes towards drawing a complete picture of the pelagic-benthic coupling in the Mediterranean mesophotic coral forests.

1. Introduction

Temperate mesophotic environments extending between 30 and 150 m depth, host dense animal-dominated communities adapted to a decreasing light gradient (Cerrano et al., 2010, 2019; Bo et al., 2011a, 2019a; Grinyó et al., 2016). These animal-dominated communities (i.e. sponges, corals, gorgonians), also called animal forests (Rossi et al., 2017a), provide structural heterogeneity and complexity, sustaining high levels of biodiversity and serving as shelter and refuge for many other associated species (Buhl-Mortensen et al., 2010; Rossi et al., 2017a). Animal forests enhance nutrient exchange and retain carbon, nitrogen and other elements (Rossi et al., 2017b), influencing pelagic-benthic coupling processes (Gili and Coma, 1998). Despite these important ecosystemic functions, mesophotic environments have been overlooked compared to shallow structured habitats (Bo et al., 2009; Gori et al., 2017). The recent use of modern sampling techniques, such as technical diving or Remotely Operated Vehicle (ROV) surveys, has facilitated the study of the spatial distribution and abundance of mesophotic organisms, however, difficulties in sampling and monitoring

species at these depths have prevented research on their ecological and biological traits.

The trophic ecology of mesophotic species, in particular, remains one of the less investigated aspects, because it is challenging to perform *in situ* incubations with their natural food sources (Ribes et al., 2003), to analyze their gut contents (Grigg, 1965; Rossi et al., 2004), or to sample and rear these fragile organisms in aquaria (Coppari et al., 2019; Orejas et al., 2019). For this purpose, the analysis of the stable carbon and nitrogen isotopic ratios of the animal tissue and potential food sources has been considered as a valid alternative to traditional methods, because it highlights not only the main food source but also the relative trophic level of an organism (DeNiro and Epstein, 1978; Post, 2002). In addition, stable isotopic analysis is also able to show the actual food assimilation integrated over a long period, corresponding to the turnover of the analyzed tissue (Hesslein et al., 1993; Carlier et al., 2007). This is particularly important for sessile organisms, whose diets can change during the year in relation to seasonal shifts in temperature and food availability (Ribes et al., 1999).

Surveys of Mediterranean mesophotic ecosystems have revealed the

* Corresponding author.

E-mail addresses: coppari.martina85@gmail.com (M. Coppari), ferrier@centrescientifique.mc (C. Ferrier-Pagès), michela.castellano@unige.it (M. Castellano), francesco.massa@unige.it (F. Massa), olivari.enrico@libero.it (E. Olivari), giorgio.bavestrello@unige.it (G. Bavestrello), povero@unige.it (P. Povero), marzia.bo@unige.it (M. Bo).

<https://doi.org/10.1016/j.ecss.2019.106520>

Received 18 December 2018; Received in revised form 27 November 2019; Accepted 2 December 2019

Available online 4 December 2019

0272-7714/© 2019 Elsevier Ltd. All rights reserved.

existence of dense forests of antipatharians, commonly known as black corals (Bo et al., 2019a). These organisms grow along the deep continental shelf and upper bathyal slope, over hard substrates, under dim-light conditions and strong currents (Wagner et al., 2012; Molodtsova and Opresko, 2017; Bo et al., 2019a). Although the bathymetric distribution of black coral species in the Mediterranean Sea extends from 50 to 2000 m, the maximal diversity and abundance are observed at mesophotic depths (Bo and Bavestrello, 2019; Bo et al., 2019a). Here, the ecosystem is dominated by *Antipathella subpinnata* (Ellis and Solander, 1786) (Hexacorallia, Myriopathidae) (Bo et al., 2018), which is considered as an ecosystem engineer species (*sensu* Jones et al., 1994) due to its arborescent and tridimensional shape and tendency to form aggregations (Bo et al., 2009, 2011a). This species has, thus, attracted attention, with studies focused on its taxonomy and phylogeny (Opresko, 2001; Bo et al., 2008, 2018), spatial and bathymetrical distribution, population structure (Bo et al., 2008; 2009), sexual (Gaino and Scoccia, 2010) and asexual reproduction (Coppari et al., 2019), associated fauna and ecological role (Bo et al., 2011b, 2019b). However, and contrarily to other Mediterranean habitat-forming coral species for which the feeding ecology has been already studied (Orejas et al., 2003; Duineveld et al., 2004; Gori et al., 2012, 2017; Grinyó et al., 2018), this aspect of *A. subpinnata* has never been investigated. This is a fundamental ecological trait, as diet greatly influences the life cycles of coastal benthic suspension feeders and profoundly interacts with the pelagic-benthic coupling processes (Coma et al., 2000).

The aim of this work is to provide a first assessment of the trophic ecology of *A. subpinnata* through the analysis of the stable isotopic ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) composition of its coenenchymal tissue and of its possible food sources. This study was performed in early spring and autumn, after the mixing of the water column (Estrada, 1996). Such mixing allows surface primary production to reach the mesophotic depth where *A. subpinnata* lives.

2. Materials and methods

2.1. Study area and environmental conditions

Organisms were sampled in the deep rocky shoal of Punta Faro ($44^\circ 17.63' \text{N}$; $9^\circ 13.27' \text{E}$) (Fig. 1), located in the Ligurian Sea, 600 m SE from the Portofino Promontory. The shoal is 200 m long and less than 100 m wide and ranges between 63 and 77 m in depth. It is dominated by a coralligenous community, mainly characterized by a dense population of *Eunicella cavolini* (Koch, 1887) (Octocorallia, Gorgoniidae) extending on the top of the elevation and a forest of *A. subpinnata*, located on the western edge. A biogenic detritic bottom populated by a dense meadow of *Lytocarpia myriophyllum* (Linnaeus, 1758) (Hydrozoa, Aglaopheniidae) surrounds the shoal. The area is under the influence of the general cyclonic circulation of the western Ligurian Sea and is mostly oligotrophic (Misic and Fabiano, 2006; Mangialajo et al., 2007; d'Ortenzio and Ribera d'Alcalà, 2009; Morabito et al., 2018). However, from late winter to spring, phytoplankton blooms can increase the chlorophyll level up to $1\text{--}2 \mu\text{g L}^{-1}$ (Ruggieri et al., 2006). Another growth phase of phytoplankton can occur in autumn, triggered by the enhanced nutrient availability brought by terrestrial river run-off, but generally with lower intensity than in winter - spring (Ruggieri et al., 2006). In addition, under windy conditions, that occur throughout the year, the presence of the Portofino Promontory's headland can occasionally supply the area with coastal waters coming from the nearby, urbanized Tigullio Gulf (Doglioli et al., 2004; Bertolino et al., 2016), enriched in particulate matter and inorganic nutrients (Ruggieri et al., 2006; Misic et al., 2011).

2.2. Sampling procedure

The main physical and biological characteristics of the water column (temperature, salinity, *in vivo* fluorescence) were recorded from August to November 2016 (hereafter autumn) and from February to May 2017 (hereafter spring) with an Idronaut 316 plus probe equipped with a Turner Cyclops 7TM fluorometer, from the surface to 65 m depth. To

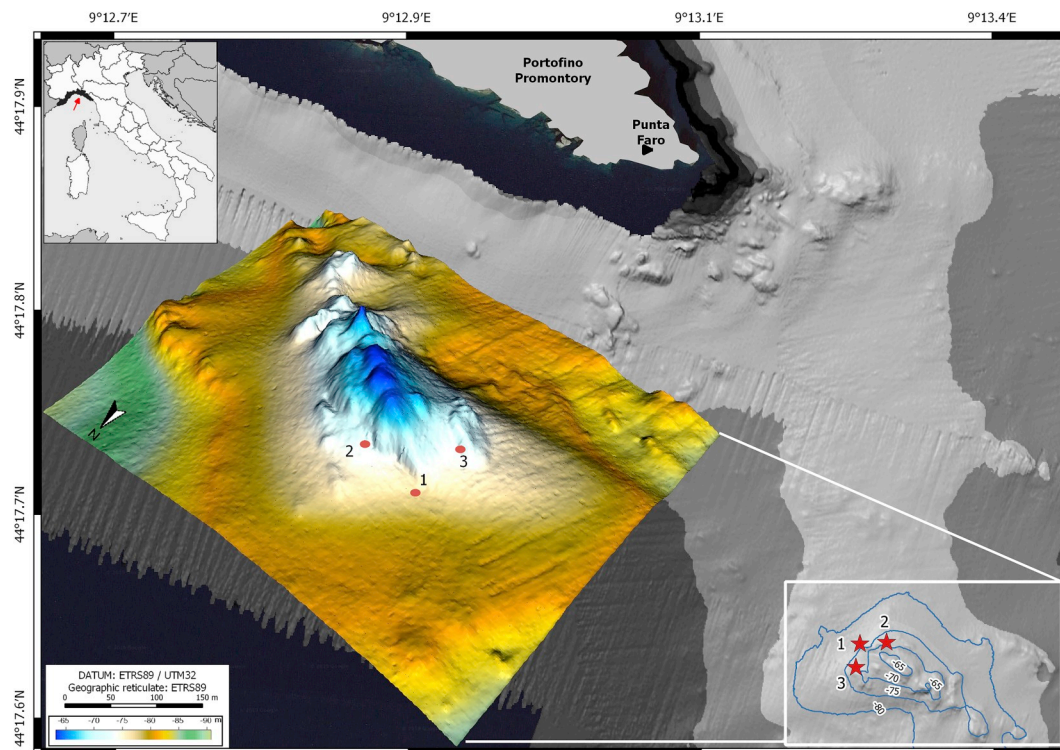


Fig. 1. Map of the study area showing the position of Punta Faro deep shoal. The stars indicate the position of the three sampling stations. The 3D map in the inset shows a vertical exaggeration of 3x.

calibrate the fluorometer, *in situ* seawater samples were collected and chlorophyll-a was determined in the laboratory (Holm-Hansen et al., 1965). In April, due to technical problems the water column was investigated only down to 27 m depth.

Three types of sampling were collected for stable isotope analysis: 1) ten pieces (10 cm long) of different colonies of *A. subpinnata* were collected, at Station 1 at a depth of 67 m, using technical SCUBA diving, in November 2016 and May 2017 for the autumn and spring sampling, respectively. Colonies were immediately frozen and kept at -80°C then broken into pieces prior to the analysis; 2) To assess the importance of resuspended POM in the diet of *A. subpinnata*, three replicates of surface sediment were also sampled at station 1 (right below the colonies; Fig. 1) in autumn and spring and kept at -80°C until analysis; 3) Pico-, nano-, micro- and mesozooplankton samples were finally collected, as described in detail below, at stations 1, 2, and 3 (Fig. 1) and kept frozen at -80°C until analysis. Station 1 was positioned right above (65 m deep) the *A. subpinnata* population, while stations 2 and 3 were positioned at the same depth, 50 m away from Station 1 (Fig. 1).

Plankton collection started three months prior to the collection of *A. subpinnata*, in order to take into account the turnover of the coral tissues for the isotopic signal. Five surveys were performed in autumn 2016, while four surveys occurred in spring 2017 (Table 1). Pico-nanoplankton (0.2–20 μm), and microplankton (20–200 μm) were collected at 65 m depth by means of a 10 L Niskin bottle. For all fractions, water was pre-filtered through a 200 μm mesh filter to remove the biggest particles. For pico- and nanoplankton, two replicates of 2 L of water were filtered through a 20 μm mesh and retrieved on pre-combusted glass fiber filters (Whatman GF/F). For microplankton, 10 L of water were filtered on a 20 μm mesh, resuspended in pre-filtered sterilized seawater and ultimately filtered through pre-combusted GF/F filters. Filters were kept at -80°C until analysis. For mesozooplankton (0.2–20 mm), two replicates were collected at the three stations by vertical hauls from 65 m to the surface. Two other replicates were collected right above the black coral forest in the depth range between 65 and 50 m by means of a WP2 closing plankton net with a 200 μm mesh size. Filtered water volumes were calculated based on the area of the net mouth and the length of the released wire (Sameoto et al., 2000). One replicate (from which three sub-samples were collected) was kept at -80°C for the stable isotope analysis, after filtration through a 1 mm mesh to eliminate organisms larger than *A. subpinnata* polyps (about 0.7–0.9 mm in transverse diameter) (Bo et al., 2018). The second replicate was fixed with 4% formalin to perform taxonomic studies, in order to identify the dominant feeding strategy of zooplankton in each sampling period (e.g. filter feeders or predators). All samples were sorted into different taxa and identified under a stereomicroscope (Zeiss) in order to separate filter feeders from predators, while copepods were identified to species or genus level (Camatti and Ferrari, 2010).

2.3. Stable isotopic analysis

Filters for the $\delta^{13}\text{C}$ analyses were treated with 100 μL HCl (10%), and rinsed with distilled water while filters for $\delta^{15}\text{N}$ were left untreated and analyzed separately. They were all freeze-dried until subsequent analyses. Mesozooplankton samples were first divided into two fractions and treated with or without HCl (and rinsing), before being freeze-dried and ground to a powder using an agate mortar and pestle. Sediment samples were first ground to a powder, divided into sub-samples of 50 mg, treated with or without HCl, and freeze-dried until subsequent analysis. Coral samples were first freeze-dried, and then ground gently to separate the tissue from the axial skeleton, before being treated with or without HCl for the isotopic analyses.

Stable isotope analyses were performed with a Dual pumped SerCon H 20–20 Isotope Ratio Mass Spectrometry (IRMS) coupled to Thermo EA1110 elemental analyzer. International reference materials (IAEA-600 and IAEA-CH6, International Atomic Energy Agency) were used for calibration of results to Vienna PeeDee Belemnite (VPDB) and air, and

Table 1

$\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ mean value and standard deviation (SD) of the different food sources and of the tissue of *Antipathella subpinnata*. * indicates that these three samples were sub-replicates.

DATE	FOOD SOURCE	n	$\delta^{15}\text{N}$ (‰)		$\delta^{13}\text{C}$ (‰)	
			mean	SD	mean	SD
01/08/2016	Pico-nanoplankton	3	3.13	0.30	−23.74	0.19
	Microplankton	3	6.79	0.53	−24.03	0.09
	Mesozooplankton (65–0 m)	3	3.40	0.42	−21.32	0.80
23/08/2016	Mesozooplankton (65–50 m)	3*	3.47	0.04	−21.78	0.37
	Pico-nanoplankton	3	4.81	1.21	−23.54	0.34
	Microplankton	3	8.26	0.72	−24.31	0.62
09/09/2016	Mesozooplankton (65–0 m)	3	3.33	0.38	−20.90	0.01
	Mesozooplankton (65–50 m)	3*	3.88	0.07	−21.62	0.24
	Pico-nanoplankton	3	3.02	1.25	−23.67	0.28
04/10/2016	Microplankton	3	6.81	0.34	−24.19	0.39
	Mesozooplankton (65–0 m)	3	2.98	0.25	−21.96	0.17
	Mesozooplankton (65–50 m)	3*	3.47	0.58	−22.43	0.31
30/11/2016	Pico-nanoplankton	3	1.49	0.12	−23.45	0.16
	Microplankton	3	3.34	1.04	−24.73	0.11
	Mesozooplankton (65–0 m)	3	2.65	0.14	−21.89	0.18
15/02/2017	Mesozooplankton (65–50 m)	3*	3.91	0.10	−21.73	0.13
	Pico-nanoplankton	3	2.30	0.70	−22.52	0.16
	Microplankton	3	5.34	0.18	−23.56	0.28
10/03/2017	Mesozooplankton (65–0 m)	3	4.67	0.11	−20.07	0.09
	Mesozooplankton (65–50 m)	3*	4.36	0.06	−20.25	0.12
	Sediment	3	3.36	0.79	−1.56	3.19
12/04/2017	<i>A. subpinnata</i>	10	6.05	0.31	−21.35	0.37
	Pico-nanoplankton	1($\delta^{15}\text{N}$) 3($\delta^{13}\text{C}$)	1.69	/	−23.92	0.14
	Microplankton	3	6.31	0.49	−24.33	0.28
10/05/2017	Mesozooplankton (65–0 m)	3	4.15	2.10	−22.03	0.62
	Mesozooplankton (65–50 m)	3*	3.10	0.22	−22.33	0.26
	Pico-nanoplankton	3	6.00	1.83	−23.17	0.38
12/04/2017	Microplankton	3	6.68	1.61	−23.34	1.07
	Mesozooplankton (65–0 m)	3	4.00	0.08	−21.72	0.12
	Mesozooplankton (65–50 m)	3*	4.22	0.06	−21.90	0.29
10/05/2017	Pico-nanoplankton	3	4.73	0.32	−23.77	0.58
	Microplankton	3	10.34	2.17	−24.31	0.23
	Mesozooplankton (65–0 m)	3	5.12	0.08	−22.76	0.08
10/05/2017	Mesozooplankton (65–50 m)	3*	5.40	0.11	−22.40	0.14
	Pico-nanoplankton	3	7.78	0.37	−24.48	0.21
	Microplankton	3	10.35	2.04	−25.08	0.47
10/05/2017	Mesozooplankton (65–0 m)	2	5.01	0.08	−22.47	0.02
	Mesozooplankton (65–50 m)	2	6.02	0.21	−22.08	0.13
	Sediment	3	2.08	1.61	0.20	0.43
10/05/2017	<i>A. subpinnata</i>	10	6.11	0.18	−21.89	0.21

two different analytical quality control samples were also analyzed with each batch. Precision was determined by repeated analysis of the reference materials with quality controls that were better than $\pm 0.20\%$ and $\pm 0.15\%$ for measured $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values, respectively. Data are expressed in the standard δ unit notation:

$$\delta X = [(R_{\text{sample}}/R_{\text{reference}}) - 1] \times 10^3$$

where $R = {}^{13}\text{C}: {}^{12}\text{C}$ for carbon and ${}^{15}\text{N}: {}^{14}\text{N}$ for nitrogen, and reported relative to VPDB for carbon and to atmospheric N_2 for nitrogen.

2.4. Statistical analysis

In order to test for significant differences in the isotopic signature of all food sources and *A. subpinnata* tissue, a one-way ANOVA was performed on the dataset, separately for autumn and spring. Normality and homogeneity of variance were verified with Shapiro-Wilk Test and Bartlett Test, respectively, and no transformation of the data was

applied. When the assumptions of normality and homogeneity of variance were not met, a Wilcoxon test was used to check for isotopic signature differences among the food sources and the black coral tissue. A one-way ANOVA was also used to evaluate significant seasonal differences in the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of the tissue of *A. subpinnata*. A two-way ANOVA was used to test the increase in $\delta^{15}\text{N}$ of the food sources among seasons. Statistical analyses were performed with R (<http://www.r-project.org>).

3. Results

3.1. Environmental characterization

The main physical and biological characteristics (temperature, salinity, chlorophyll-a) of the sampling area in 2016–2017 are summarized in Fig. 2.

The water column experienced major changes between summer and

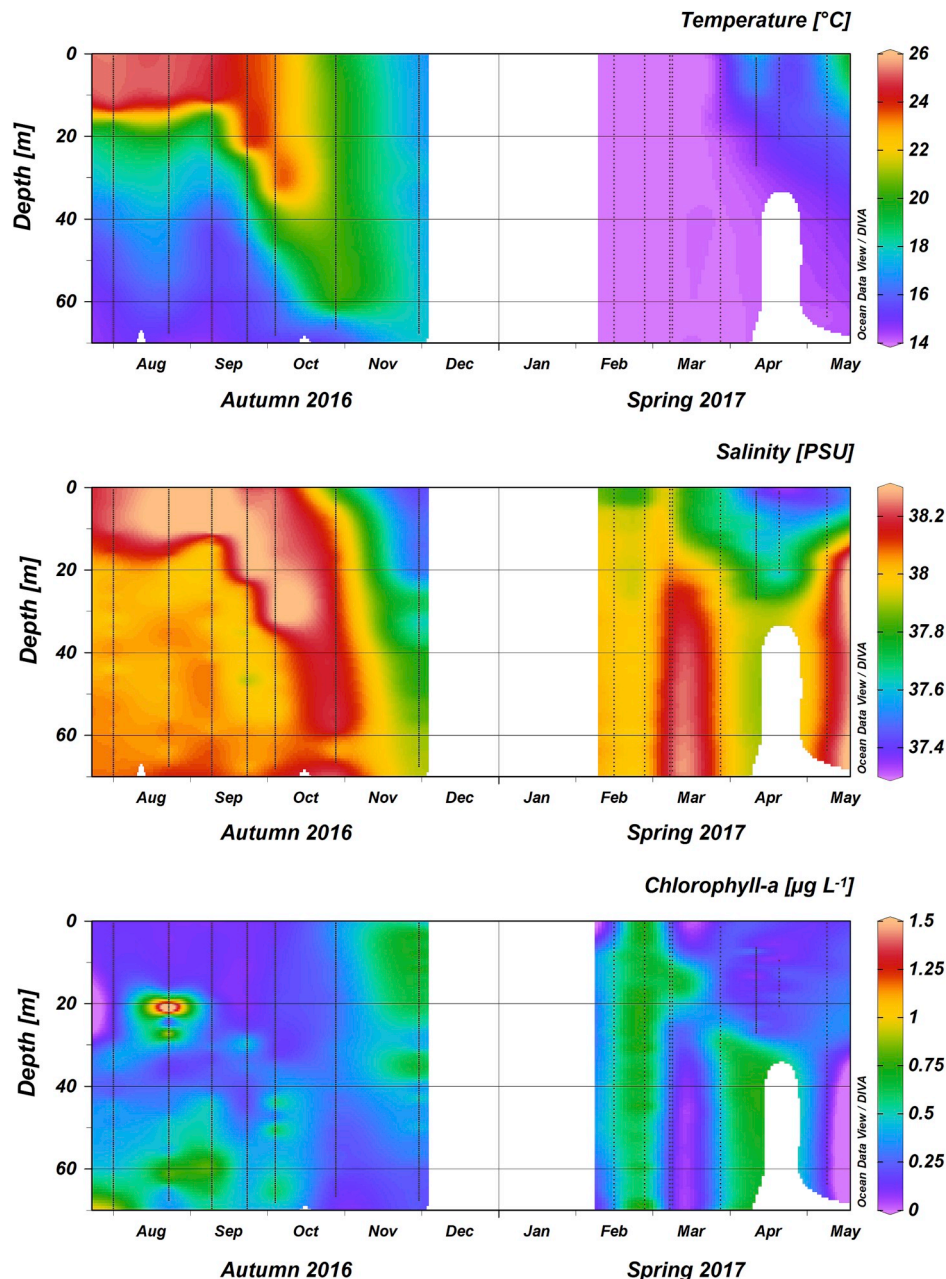


Fig. 2. Environmental characteristics of the study area during the two sampling periods. (Temperature, °C; Salinity, PSU; Chlorophyll-a, $\mu\text{g L}^{-1}$).

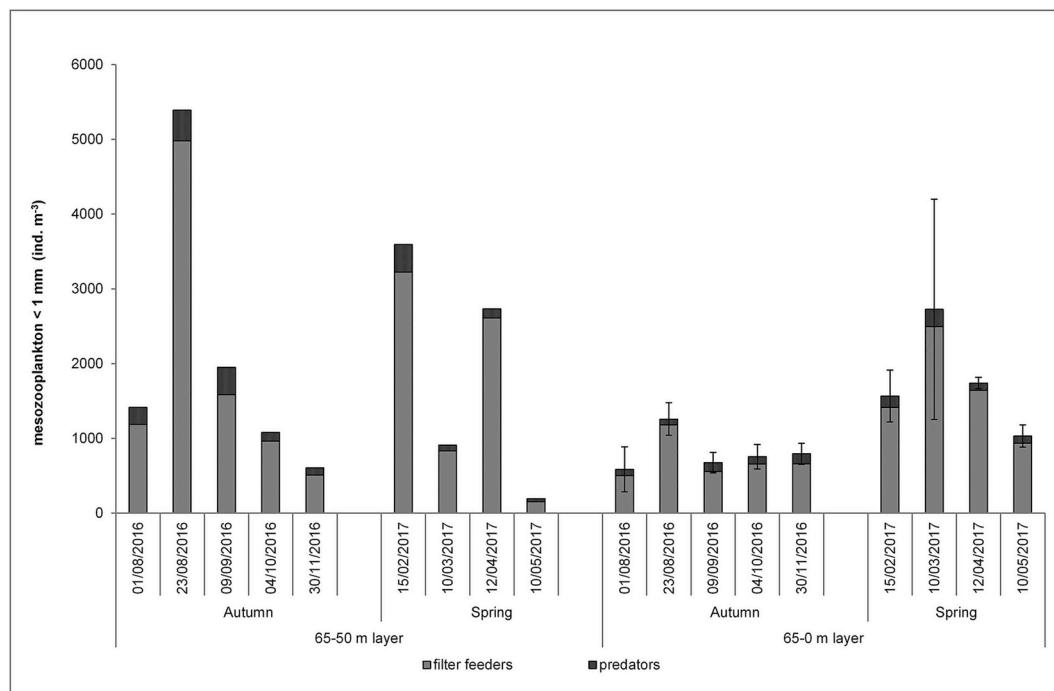


Fig. 3. Small mesozooplankton abundance (ind. <1 mm³) in the 65-50 m and 65-0 m layers.

autumn 2016: the summer thermohaline stratification with high temperatures and low salinity (Fig. 2) was stable until the end of September. During this period, phytoplankton biomass remained low above the thermocline, but slightly higher below 50 m (Deep Chlorophyll Maxima Fig. 2). The only exception was a subsurface maximum layer (>1 µg L⁻¹) at 20–30 m depth on 23rd August. In October, there was a progressive homogenization of the water column down to 60 m (temperature >20 °C) due to the seasonal thermal cycle and the increasing action of autumn storms and waves. In November, when black coral sampling occurred, the water column temperature was nearly homogeneous (between 17 and 17.8 °C), however, strong precipitation and flow of continental water bodies during the whole month, significantly decreased salinity in the surface layer. At this period, phytoplankton biomass increased in the upper layer, following inflows of nutrient-rich freshwater.

In February 2017, the water column showed isothermal conditions, with stable temperature and salinity, as well as an equal distribution of phytoplankton biomass throughout the water column. Chlorophyll-a concentrations were low, except for a slight increase at the end of the month. Between March and May, when black coral sampling occurred, temperature started to increase, particularly in surface waters, while phytoplankton biomass was low, except for April in deep waters.

The small mesozooplankton (<1 mm) was mainly composed of filter feeders: copepods were predominant (most abundant genera: *Clausocalanus*, *Oithona* and *Paracalanus*), followed by appendicularians

(*Oikopleura* and *Fritillaria* genera), and pteropods, and, in summer-autumn 2016, cladocerans (Fig. 3). Considering all samples and seasons, the percentages of predator species (mainly *Candacia* spp., *Corycaeus* spp., *Oncaea* spp., Cnidaria and Chetognata) varied from 4.4% to 20.4% in the 65–50 m samples and from 5.6% to 17.4% in the 65–0 m samples. In autumn, the mesozooplankton abundance varied from 607 ind. m⁻³ to 5390 ind. m⁻³ in the 65–50 m layer, and from 586 ± 301 ind. m⁻³ to 1259 ± 216 ind. m⁻³ in the 65-0 m layer. In spring, the density of mesozooplankton varied from 193 ind. m⁻³ to 3594 ind. m⁻³ in the 65–50 m layer and from 1032 ± 148 ind. m⁻³ to 2726 ± 1472 ind. m⁻³ considering the entire water column (65–0 m layer). While the abundance in the 65-50 m layer was highly variable at both sampling seasons, the abundance in the entire water column followed the seasonal development of mesozooplankton, with low values in summer and much higher values in spring (Licandro and Ibanez, 2000).

3.2. Stable isotopic composition

The results of the isotopic analyses of *A. subpinnata* and possible food sources in the two seasons are summarized in Table 1. The δ¹³C values of *A. subpinnata* were significantly different between autumn (−21.35 ± 0.37) and spring (−21.89 ± 0.21) (ANOVA, F = 19.36; p = <0.01). δ¹⁵N values, however, were stable across seasons (ANOVA, F = 0.383; p = 0.542), between 6.05 ± 0.31 and 6.11 ± 0.18 in autumn and spring, respectively.

Table 2

Table summarizing the results of the Wilcoxon test and one-way ANOVA between *Antipathella subpinnata*'s tissue and the different food sources. n.s. means 'not significant'.

Food source	Antipathella subpinnata			
	Autumn		Spring	
	δ ¹⁵ N	δ ¹³ C	δ ¹⁵ N	δ ¹³ C
Pico-nanoplankton	W = 161 p < 0.01	W = 180 p < 0.01	W = 60 p = n.s.	W = 144 p < 0.01
Microplankton	W = 64 p = n.s.	W = 180 p < 0.01	W = 24 p < 0.01	W = 143 p < 0.01
Mesoplankton (65–0 m)	W = 180 p < 0.01	W = 105 p = n.s.	W = 132 p < 0.01	W = 90 p = n.s.
Mesoplankton (65–50 m)	W = 168 p < 0.01	W = 108 p = n.s.	W = 147 p < 0.01	F = 7.58 p = 0.01

The $\delta^{13}\text{C}$ values of pico-, nano- and microplankton significantly differed from *A. subpinnata*'s tissue in both seasons (Table 2). In contrast, the $\delta^{13}\text{C}$ values of the mesozooplankton (65–50 m and 65–0) did not significantly differ from the coral tissue in either season (Table 2). The $\delta^{15}\text{N}$ signature of pico-nanoplankton was significantly different from the coral tissue in autumn but not in spring. An opposite result was observed for microplankton. All fractions of mesozooplankton were significantly different from the coral's tissue at both seasons. Even though not significant, an increasing trend in the $\delta^{15}\text{N}$ signature was observed in spring for the plankton components (2-way ANOVA, food source $F = 7.66$; $p = <0.01$; season $F = 7.59$; $p = 0.01$; food source \times season $F = 0.76$; $p = 0.55$) (Fig. 4).

Carbon and nitrogen signatures of the different samples were simultaneously plotted in Fig. 5 to highlight the preferentially exploited food sources of *A. subpinnata* in each season. Such plots are used in complex food chains to determine the food source of a specific consumer assuming a mean trophic enrichment of 1‰ in $\delta^{13}\text{C}$ (Rau et al., 1990) and 2.5–3.4‰ in $\delta^{15}\text{N}$ (Minagawa and Wada, 1984; Gollety et al., 2010) for the consumer. In other words, the potential types of prey ingested by *A. subpinnata* are those, which are depleted by 1‰ in $\delta^{13}\text{C}$ and by 2.5–3.5‰ in $\delta^{15}\text{N}$ compared to the tissue of the black coral. Application of the above model to *A. subpinnata* in Fig. 5 shows that the expected mean isotopic ratios for the preferentially exploited food resources (represented by a red square in Fig. 5) were close to the ratios of pico-nanoplankton in autumn and mesozooplankton in spring and rather close to mesozooplankton in autumn (Fig. 5). Microplankton, whose signature is always higher than the signature of *A. subpinnata* could not be considered as a potential food source. Also, resuspended sediment, whose signature was very low compared to the potential food source (Table 1), could not be considered either (and the signature was too low to be clearly represented in Fig. 5).

4. Discussion

This study investigates for the first time the feeding ecology of *A. subpinnata*, one of the most representative habitat-forming black coral species of the Mediterranean mesophotic zone. Such species is an important component of the Mediterranean animal forests, which connect the bottom substrate and the water column (benthic-pelagic coupling) through trophic interactions and biogeochemical cycling (Cerrano et al., 2019). Animal forests, indeed, contribute to a large fraction of the benthic secondary production in coastal Mediterranean areas (reviewed in Munari et al., 2013; Rossi et al., 2017b) and they can significantly affect the composition of the water column in terms of plankton concentration and composition, either through mucus/nutrient release or suspension feeding. In turn, the relative abundance of plankton and other particles in seawater affect the growth rate and

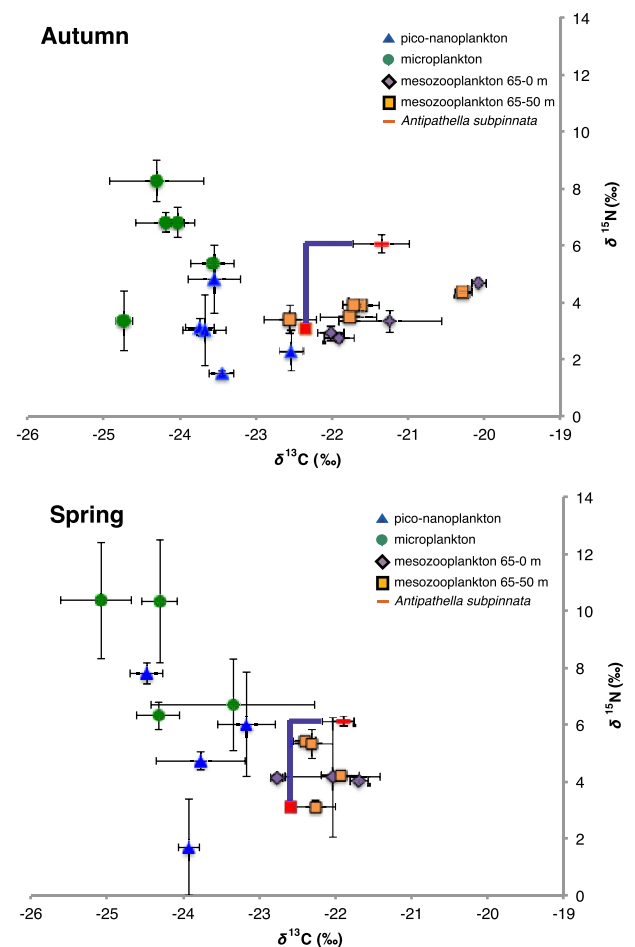


Fig. 5. $\delta^{13}\text{C}$ vs. $\delta^{15}\text{N}$ biplot showing stable isotopic composition of *Antipathella subpinnata* and analyzed food sources (pico-nanoplankton, microplankton and mesozooplankton) in autumn and spring. Sediment isotopic composition is not reported in this figure. The red square indicates the expected isotopic signature of the preferred food sources; the blue line indicates the enrichment factors between the preferred food sources and the black coral tissue. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

community structure of the suspension feeders (reviewed in Coma et al., 2000). Despite the importance of benthic habitats for ecosystem functioning, and for their role in carbon and nutrient budgets, processes

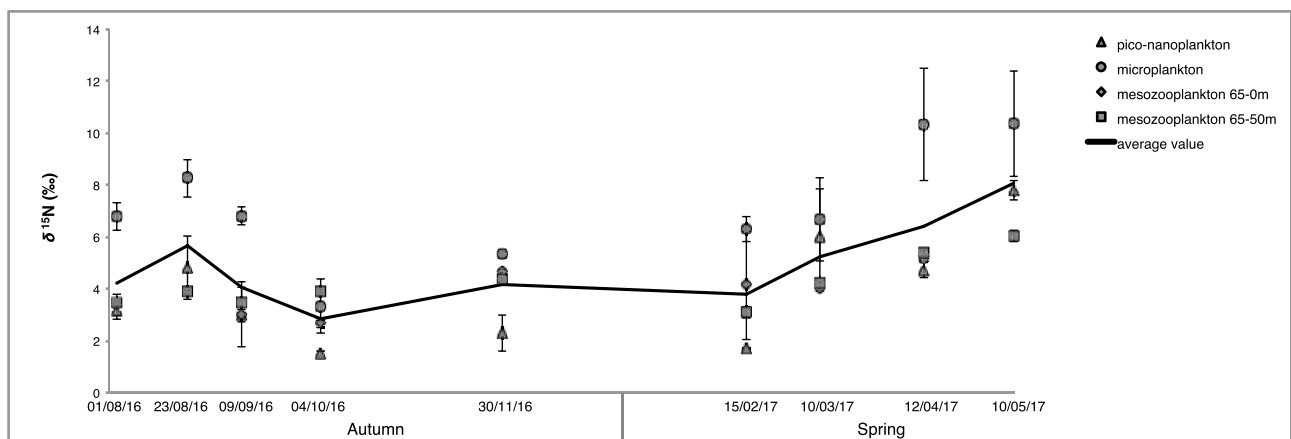


Fig. 4. Trend of variation of the $\delta^{15}\text{N}$ in all the food sources during the sampling period.

affecting the benthic-pelagic coupling are still not well understood, especially in mesophotic environments, such as those of Mediterranean black corals.

The deep shoal of Punta Faro shows the typical features of a Mediterranean oligotrophic mesophotic environment (d'Ortenzio and Ribera d'Alcalà, 2009; Mazzocchi et al., 2014), with low concentrations of chlorophyll and mesozooplankton. Mesozooplankton is mainly dominated by filter feeders such as copepods, in agreement with previous observations (Bănuș et al., 2013; Mazzocchi et al., 2014; Morabito et al., 2018), while microplankton, although not directly observed, is often dominated by diatoms and dinoflagellates during both spring and autumn (Gomez and Gorski, 2003; Ruggieri et al., 2006; Bănuș et al., 2013). In addition, slight temporal changes in the plankton isotopic signature are recorded (Fig. 4), which can be due to cascading effects (Conese et al., 2019) or to changes in the inorganic nutrient sources used by phytoplankton (Kerhervé et al., 2001).

The diet of *A. subpinnata* was estimated considering a mean trophic enrichment of 1‰ in $\delta^{13}\text{C}$ (Rau et al., 1990) and of 2.5–3‰ in $\delta^{15}\text{N}$ (Minagawa and Wada, 1984; Gollety et al., 2010) for the consumer as a result of the assimilation of food. Indeed, while carbon isotope composition of consumers is similar to the $\delta^{13}\text{C}$ of the diet (DeNiro and Epstein, 1978), the enrichment factor in $\delta^{15}\text{N}$ between two consecutive trophic levels is higher, due to the excretion of the light ^{14}N and the retention of the heavy ^{15}N inside the consumer tissue (Minagawa and Wada, 1984; McCutchan et al., 2003).

Overall, this model application indicated that *A. subpinnata* feeds consistently on pico-nanoplankton, but opportunistically captures larger preys when available. The high contribution of mesozooplankton to the black coral diet at both seasons is certainly due to the mixing of the water column (Fig. 2), with nitrate-enriched waters inducing phytoplankton blooms and the consecutive rise of zooplankton abundance (Mayot et al., 2017). Structurally, *A. subpinnata*'s polyps should also be able to feed on microplankton, since the size is intermediate between nano- and mesoplankton. However, the $\delta^{15}\text{N}$ of microplankton is higher compared to the $\delta^{15}\text{N}$ of *A. subpinnata*, and the same can be observed for the $\delta^{13}\text{C}$. Altogether, the isotopic data suggest that microplankton is not contributing to a large percentage of *A. subpinnata*'s diet. This is either due to low concentrations in seawater, which could not be clearly detected in the analysis, or to the fact that microplankton is often dominated by diatoms, which are not preyed by the black coral. These results are in agreement with the gut content analysis and underwater observations performed on shallow-water tropical black corals, which have highlighted a predatory activity on zooplankton for species characterized by large polyps (Grigg, 1965; Warner, 1981; Goldberg and Taylor, 1989; Tazioli et al., 2007) and a filter-feeding strategy based on mucous nets for species with small-sized polyps (Warner, 1977; Lewis, 1978; Goldberg and Taylor, 1989; Tazioli et al., 2007).

Based on similar studies, various differences emerged between the diet of *A. subpinnata* and that of other black corals, particularly *Leopathes glaberrima* (Esper, 1788) and *Bathypathes arctica* (Lütken, 1871), studied at 520 m and 1000 m, respectively (Sherwood et al., 2008; Carlier et al., 2009). *A. subpinnata*'s tissue is ^{13}C and ^{15}N depleted compared to these bathyal species and its isotopic signature does not match with that of resuspended sediment suggesting that detrital POM from the seafloor is not the preferential food source of the target species, at least in the study area. This difference might be explained by the mesophotic depth of the population of *A. subpinnata* investigated in this study (~70 m). The shallower distribution of this species allows for higher availability of the labile and fresh POM to the bottom, and a rapid consumption by the coral. However, the consumption of POM may not only be depth-dependent, but might also relate to the environmental silting levels. The Ligurian black coral population, located along the western side of the rocky elevation, indirectly receives water outflow from the Tigullio Gulf (Bertolino et al., 2016), suggesting moderate water turbulence and a low silted environment, which both seem to be the preferential conditions for the dense forests of this species (Bo et al.,

2009). Far less abundant aggregations, characterized by smaller colonies, are also observed in highly silted environments (Bo et al., 2012), suggesting that in such habitats the exploitation of resuspended POM could be more predominant and may have an influence on the population structure. An environment-dependent relationship might explain also the isotopic signature showed by the shallow-water species *Rhipidipathes* sp., more similar to that of deep-water species (William and Grottoli, 2010).

Previous studies also highlighted that the $\delta^{15}\text{N}$ enrichment between trophic levels could shift between 2.5 and 3.4‰ in certain taxa (DeNiro and Epstein, 1981; Vander Zanden and Rasmussen, 2001). Our study confirms the lowest value of $\delta^{15}\text{N}$ enrichment for *A. subpinnata* (2.5‰). This smaller enrichment was already reported for black corals by Sherwood et al. (2008) and was related to the reduced input of food in bathyal environments (Iken et al., 2001). In the case of *A. subpinnata*, we hypothesized, on one hand, that this low $\delta^{15}\text{N}$ enrichment could be related to the capability of this species to capture the pico-nanoplankton fraction directly, limiting the $\delta^{15}\text{N}$ enrichment along the trophic web. On the other hand, it is plausible that, based on the limited size of mesozooplankton catchable by *A. subpinnata* (<1 mm, due to the size of the polyp), this prey fraction is mainly composed by filter feeders which feed at a lower trophic level, thus contributing only marginally to the increase of $\delta^{15}\text{N}$.

The results obtained here can be compared with those found so far for other Mediterranean mesophotic anthozoans. Isotopic analyses carried out on gorgonians living below 50 m depth, support a heterotrophic diet for these species targeting zooplankton and POM (Gori et al., 2017; Grinyó et al., 2018), while *A. subpinnata*, relies more on fresh rather than detrital material. This mirrors the feeding habits of the mesophotic Mediterranean gorgonian *Paramuricea macrospina* (Koch, 1882) (Grinyó et al., 2018).

This study highlights for the first time the contribution of the most abundant Mediterranean black coral species to the Mediterranean pelagic-benthic coupling processes. By focusing on the mesophotic zone, this study helps filling the knowledge gap on trophic food webs in this bathymetric range, which has been relatively overlooked (Gori et al., 2011). Overall, it shows that *A. subpinnata* takes advantage of all nutrient sources available, transforming this planktonic food into a highly valuable benthic biomass. Further studies should now investigate the growth rate and reproductive outputs of this species, to assess its contribution to the benthic secondary production of mesophotic ecosystems.

Authors' contribution

MCo collected and analyzed the data and wrote the manuscript, CFP analyzed the data and wrote the manuscript, MCa and FM collected and analyzed the data and wrote the manuscript, EO collected the data, GB and PP wrote the manuscript and MB conceived the idea and wrote the manuscript.

Declaration of competing interest

The authors declare that they have no competing interests.

Acknowledgement

Authors would like to thank the staff of the diving center "Il Grande Blu", Gabriele Costa and Francesco Enrichetti for their help during sampling procedures; the Company of Biologist travel grant, which was used by the corresponding author for a working period at the Centre Scientifique de Monaco.

This work was funded by BIOMOUNT project MIUR-SIR (RBSI14HC90, Biodiversity patterns of the Tyrrhenian Seamounts).

References

- Bertolino, M., Betti, F., Bo, M., Cattaneo-Vietti, R., Pansini, M., Romero, J., Bavestrello, G., 2016. Changes and stability of a Mediterranean hard bottom benthic community over 25 years. *J. Mar. Biol. Assoc. U. K.* 96, 341–350.
- Bo, M., Bavestrello, G., 2019. Mediterranean black coral communities. In: Orejas, C., Jiménez, C. (Eds.), *Mediterranean Cold-Water Corals: Past, Present and Future*. Springer, ISBN 978-3-319-91607-1.
- Bo, M., Tazioli, S., Spano, N., Bavestrello, G., 2008. *Antipathella subpinnata* (antipatharia, Myriopathidae) in Italian seas. *Ital. J. Zool.* 75, 185–195.
- Bo, M., Bavestrello, G., Canese, S., Giusti, M., Salvati, E., Angiolillo, M., Greco, S., 2009. Characteristics of a black coral meadow in the twilight zone of the central Sea, Mediterranean Sea. *Mar. Ecol. Prog. Ser.* 397, 53–61.
- Bo, M., Bertolino, M., Borghini, M., Castellano, M., Covazzi Harriague, A., Camillo, C.G., Gasparini, G., Misic, C., Povero, P., Pusceddu, P., Schroeder, K., Bavestrello, G., 2011a. Characteristics of the mesophotic megabenthic assemblages of the vercelli seamount (north Tyrrhenian Sea). *PLoS One* 6 (2), e16357.
- Bo, M., Di Camillo, C.G., Puce, S., Canese, S., Giusti, M., Angiolillo, M., Bavestrello, G., 2011b. A tubulariid hydroid associated with anthozoan corals in the Mediterranean Sea. *Ital. J. Zool.* 78, 487–496.
- Bo, M., Canese, S., Spaggiari, C., Pusceddu, A., Bertolino, M., Angiolillo, M., Giusti, M., Loreto, M.F., Salvati, E., Greco, S., Bavestrello, G., 2012. Deep coral oases in the south Tyrrhenian Sea. *PLoS One* 7 (11), e49870.
- Bo, M., Barucca, M., Biscotti, M.A., Brugler, M.R., Canapa, A., Canese, S., Lo Iacono, C., Bavestrello, G., 2018. Phylogenetic relationships of Mediterranean black corals (Cnidaria: anthozoa: Hexacorallia) and implications for classification within the order Antipatharia. *Invertebr. Syst.* 32, 1102–1110.
- Bo, M., Montgomery, A.D., Opresko, D.M., Wagner, D., Bavestrello, G., 2019a. Antipatharians of the mesophotic zone: four case studies. In: Loya, Y., Puglise, K.A., Cridge, T.C.L. (Eds.), *Mesophotic Coral Ecosystems*. Springer, ISBN 978-3-319-92734-3.
- Bo, M., Bavestrello, G., Di Muzio, G., Canese, S., Betti, F., 2019b. First record of a symbiotic relationship between a polyclad and a black coral with description of *Anthoplana antipathellae* gen. et sp. nov. (Acotylea, Notoplanidae). *Mar. Biodivers.* 1–22.
- 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 margin. *Mar. Ecol.* 31, 21–50.
- Bănar, D., Carloti, F., Barani, A., Grégori, G., Neffati, N., Harmelin-Vivien, M., 2013. Seasonal variation of stable isotope ratios of size-fractionated zooplankton in the Bay of Marseille (NW Mediterranean Sea). *J. Plankton Res.* 36, 145–156.
- Camatti, E., Ferrari, I., 2010. Manuali e linee guida 56/2010. ISPRA. In: Totti, Cecilia (Ed.), *Metodologie di studio del plancton marino*. Giorgio Socal, Isabella Buttino, Marina Cabrini, Olga Mangoni, Antonella Penna, ISBN 978-88-448-0427-5, pp. 489–506.
- Carlier, A., Riera, P., Amouroux, J.M., Bodiou, J.Y., Grémare, A., 2007. Benthic trophic network in the Bay of Banyuls-sur-Mer (northwest Mediterranean, France): an assessment based on stable carbon and nitrogen isotopes analysis. *Estuar. Coast Shelf Sci.* 72, 1–15.
- Carlier, A., Le Guilloux, E., Olu, K., Sarrazin, J., Mastrototaro, F., Taviani, M., Clavier, J., 2009. Trophic relationships in a deep Mediterranean cold-water coral bank (santa maria di Leuca, ionian sea). *Mar. Ecol. Prog. Ser.* 397, 125–137.
- Cerrano, C., Danovaro, R., Gambi, C., Pusceddu, A., Riva, A., Schiaparelli, S., 2010. Gold coral (*Savalia savaglia*) and gorgonian forests enhance benthic biodiversity and ecosystem functioning in the mesophotic zone. *Biodivers. Conserv.* 19, 153–167.
- Cerrano, C., Bastari, A., Calcinai, B., Di Camillo, C., Pica, D., Puce, S., Valisano, L., Torsani, F., 2019. Temperate mesophotic ecosystems: gaps and perspectives of an emerging conservation challenge for the Mediterranean Sea. *The European Zoological Journal* 86, 370–388.
- Coma, R., Ribes, M., Gili, J.M., Zabala, M., 2000. Seasonality in coastal benthic ecosystems. *Trends Ecol. Evol.* 15, 448–453.
- Conese, I., Fanelli, E., Miserochci, S., Langone, L., 2019. Food web structure and trophodynamics of deep-sea plankton from the Bari Canyon and adjacent slope (Southern Adriatic, central Mediterranean Sea). *Prog. Oceanogr.* 175, 92–104.
- Coppari, M., Mestice, F., Betti, F., Bavestrello, G., Castellano, L., Bo, M., 2019. Fragmentation, re-attachment ability and growth rate of the Mediterranean black coral *Antipathella subpinnata*. *Coral Reefs* 38, 1–14.
- DeNiro, M.J., Epstein, S., 1978. Influence of diet on the distribution of carbon isotopes in animals. *Geochim. Cosmochim. Acta* 42, 495–506.
- DeNiro, M.J., Epstein, S., 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochim. Cosmochim. Acta* 45, 341–351.
- Doglioli, A.M., Magaldi, M.G., Vezzulli, L., Tucci, S., 2004. Development of a numerical model to study the dispersion of wastes coming from a marine fish farm in the Ligurian Sea (Western Mediterranean). *Aquaculture* 231, 215–235.
- Duineveld, G.D., Lavaleye, M.S.S., Berghuis, E.M., 2004. Particle flux and food supply to a seamount cold-water coral community (Galicia Bank, NW Spain). *Mar. Ecol. Prog. Ser.* 277, 13–23.
- d'Ortenzio, F., Ribera d'Alcalà, M., 2009. On the trophic regimes of the Mediterranean Sea: a satellite analysis. *Bioscience* 6, 139–148.
- Estrada, M., 1996. Primary production in the northwestern Mediterranean. *Scienza Marina* 60, 55–64.
- Gaino, E., Scoccia, F., 2010. Gamete spawning in *Antipathella subpinnata* (Anthozoa, Antipatharia): a structural and ultrastructural investigation. *Zoomorphology* 129, 213–219.
- Gili, J.M., Coma, R., 1998. Benthic suspension feeders: their paramount role in littoral marine food webs. *Trends Ecol. Evol.* 13, 316–321.
- Goldberg, W.M., Taylor, G.T., 1989. Cellular structure and ultrastructure of the black coral *Antipathes aperta*: 1. Organization of the tentacular epidermis and nervous system. *J. Morphol.* 202, 239–253.
- Golléty, C., Riera, P., Davout, D., 2010. Complexity of the food web structure of the *Ascophyllum nodosum* zone evidenced by a $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ study. *J. Sea Res.* 64, 304–312.
- Gómez, F., Gorsky, G., 2003. Annual microplankton cycles in villefranche bay, Ligurian Sea, NW mediterranean. *J. Plankton Res.* 25, 323–339.
- Gori, A., Rossi, S., Berganzo, E., Pretus, J.L., Dale, M.R.T., Gili, J.M., 2011. Spatial distribution patterns of the gorgonians *Eunicella singularis*, *Paramuricea clavata* and *Leptogorgia sarmentosa* (cape of creus, northwestern Mediterranean Sea). *Mar. Biol.* 158, 143–158.
- Gori, A., Viladrich, N., Gili, J.M., Kotta, M., Cucio, C., Magni, L., Bramanti, L., Rossi, S., 2012. Reproductive cycle and trophic ecology in deep versus shallow populations of the Mediterranean gorgonian *Eunicella singularis* (Cap de Creus, northwestern Mediterranean Sea). *Coral Reefs* 31, 823–837.
- Gori, A., Bavestrello, G., Grinyó, J., Dominguez-Carrió, C., Ambroso, S., Bo, M., 2017. Animal forests in deep coastal bottoms and continental shelves of the Mediterranean Sea. In: Rossi, S., Bramanti, L., Gori, A., Orejas Saco del Valle, C. (Eds.), *Marine Animal Forests. The Ecology of Benthic Biodiversity Hotspots*, vol. 1. Springer International Publishing, ISBN 978-3-319-21011-7, pp. 207–233. Chp. 7.
- Grigg, R.W., 1965. Ecological studies of black coral in Hawaii. *Pac. Sci.* 19, 244–260.
- Grinyó, J., Gori, A., Ambroso, S., Purroy, A., Calatayud, C., Dominguez-Carrió, C., Coppari, M., Lo Iacono, C., López-González, P., Gili, J.M., 2016. Diversity, distribution and population size structure of deep Mediterranean gorgonian assemblages (Menorca Channel, Western Mediterranean Sea). *Prog. Oceanogr.* 145, 42–56.
- Grinyó, J., Viladrich, N., Díaz, D., Muñoz, A., Mallol, S., Salazar, J., Castillo, R., Gili, J.M., Gori, A., 2018. Reproduction, energy storage and metabolic requirements in a mesophotic population of the gorgonian *Paramuricea macrospina*. *PLoS One*. <https://doi.org/10.1371/journal.pone.0203308>.
- Hesslein, R.H., Hallard, K.A., Ramlal, P., 1993. Replacement of sulfur, carbon, and nitrogen in tissue of growing broad whitefish (*Coregonus nasus*) in response to a change in diet traced by $\delta^{34}\text{S}$, $\delta^{13}\text{C}$, $\delta^{15}\text{N}$. *Can. J. Fish. Aquat. Sci.* 50, 2071–2076.
- Holm-Hansen, O., Lorenzen, C.J., Holmes, R.W., Strickland, J.D.H., 1965. Fluorometric determination of chlorophyll. *ICES. Journal of Marine Science* 30, 3–15.
- Iken, K., Brey, T., Wand, U., Voigt, J., Junghans, P., 2001. Food web structure of the benthic community at the Porcupine Abyssal Plain (NE Atlantic): a stable isotope analysis. *Prog. Oceanogr.* 50, 383–405.
- Jones, C.G., Lawton, J.H., Shachak, M., 1994. Organisms as ecosystem engineers. *Oikos* 69, 373–386.
- Kerhervé, P., Minagawa, M., Heussner, S., Monaco, A., 2001. Stable isotopes ($^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$) in settling organic matter of the northwestern Mediterranean Sea: biogeochemical implications. *Oceanol. Acta* 24, 77–85.
- Lewis, J.B., 1978. Feeding mechanisms in black corals (Antipatharia). *J. Zool.* 186, 393–396.
- Licandro, P., Ibanez, F., 2000. Changes of zooplankton communities in the Gulf of Tigullio (Ligurian Sea, western mediterranean) from 1985 to 1995. Influence of hydrodynamic factors. *J. Plankton Res.* 22, 2225–2253.
- Mangialajo, L., Ruggieri, N., Asnaghi, V., Chiantore, M., Povero, P., Cattaneo-Vietti, R., 2007. Ecological status in the Ligurian Sea: the effect of coastline urbanization and the importance of proper reference sites. *Mar. Pollut. Bull.* 55, 30–41.
- Mayot, N., d'Ortenzio, F., Taillandier, V., Prieur, L., De Fommervault, O.P., Claustre, H., Bosse, A., Testor, P., Conan, P., 2017. Physical and biogeochemical controls of the phytoplankton blooms in North Western Mediterranean Sea: a multiplatform approach over a complete annual cycle (2012–2013 DEWEX experiment). *J. Geophys. Res.: Oceans* 122, 9999–10019.
- Mazzocchi, M.G., Siokou, I., Tirelli, V., de Puelles, M.F., Örek, Y.A., de Olazabal, A., Gubanova, A., Kress, N., Protapapa, M., Solidoro, C., Tagliatela, S., Terbiyik Kurt, T., 2014. Regional and seasonal characteristics of epipelagic mesozooplankton in the Mediterranean Sea based on an artificial neural network analysis. *J. Mar. Syst.* 135, 64–80.
- McCutchan Jr., J.H., Lewis Jr., W.M., Kendall, C., McGrath, C.C., 2003. Variation in trophic shift for stable isotope ratios of carbon, nitrogen, and sulfur. *Oikos* 102, 378–390.
- Minagawa, M., Wada, E., 1984. Stepwise enrichment of ^{15}N along food chains: further evidence and the relation between $\delta^{15}\text{N}$ and animal age. *Geochim. Cosmochim. Acta* 48, 1135–1140.
- Misic, C., Fabiano, M., 2006. Ecto-enzymatic activity and its relationship to chlorophyll-a and bacteria in the Gulf of genoa (Ligurian Sea, NW mediterranean). *J. Mar. Syst.* 60, 193–206.
- Misic, C., Castellano, M., Harriague, A.C., 2011. Organic matter features, degradation and remineralisation at two coastal sites in the Ligurian Sea (NW Mediterranean) differently influenced by anthropogenic forcing. *Mar. Environ. Res.* 72, 67–74.
- Molodtsova, T.N., Opresko, D.M., 2017. Black corals (anthozoa: antipatharia) of the claron-clinterton fracture zone. *Mar. Biodivers.* 47, 349–365.
- Morabito, G., Mazzocchi, M.G., Salmasso, N., Zingone, A., Bergami, C., Flaim, G., Accorroni, S., Basset, A., Bastianini, M., Belmonte, G., Bernardi Aubry, F., Bertani, I., Bresciani, M., Buzzi, F., Cabrini, M., Camatti, E., Caroppo, C., Cataletto, B., Castellano, M., Del Negro, P., de Olazabal, A., Di Capua, I., Elia, A.C., Fornasaro, D., Giallaini, M., Grilli, F., Leoni, B., Lipizer, M., Longobardi, L., Ludovisi, A., Luglie, A., Manca, M., Margiotta, M., Mariani, M.A., Marini, M., Marzocchi, M., Obertegger, U., Oggioni, A., Padedda, B.M., Pansera, M., Piscia, R., Povero, P., Pulina, S., Romagnoli, T., Rosati, I., Rossetti, G., Rubino, F., Sarno, D., Satta, C.T., Sechi, N.,

- Stanca, E., Tirelli, V., Totti, C., Pugnetti, A., 2018. Plankton dynamics across the freshwater, transitional and marine research sites of the LTER-Italy Network. Patterns, fluctuations, drivers. *Sci. Total Environ.* 627, 373–387.
- Munari, C., Serafin, G., Mistri, M., 2013. Structure, growth and secondary production of two Tyrrhenian populations of the white gorgonian *Eunicella singularis* (Esper 1791). *Estuarine, Coastal and Shelf Science* 119, 162–166.
- Orejas, C., Gili, J.M., Arntz, W., 2003. Role of small-plankton communities in the diet of two Antarctic octocorals (*Primnois antarctica* and *Primnoella* sp.). *Mar. Ecol. Prog. Ser.* 250, 105–116.
- Orejas, C., Taviani, M., Ambroso, S., Andreou, V., Bilan, M., Bo, M., Brooke, S., Buhl-Mortensen, P., Cordes, C., Dominguez-Carrió, C., Ferrier-Pagès, C., Godinho, A., Gori, A., Grinyó, J., Gutiérrez-Zarate, C., Henninge, S., Jiménez, C., Larsson, A.I., Lartaud, F., Lunden, J., Maier, C., Maier, S.R., Movilla, J., Murray, F., Peru, E., Purser, A., Rakka, M., Reynaud, S., Murray Roberts, J., Siles, P., Strömberg, S.M., Thomsen, L., van Oevelen, D., Veiga, A., Carreiro-Silva, M., 2019. 38 cold-water coral in aquaria: advances and challenges. A focus on the mediterranean. *Mediterranean Cold-Water Corals: Past, Present and Future*. Springer, Cham, pp. 435–471.
- Post, D.M., 2002. Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology* 83, 703–718.
- Rau, G.H., Teysse, J.L., Rassoulzadegan, F., Fowler, S.W., 1990. $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ variations among size-fractionated marine particles: implications for their origin and trophic relationships. *Mar. Ecol. Prog. Ser.* 59, 33–38.
- Ribes, M., Coma, R., Gili, J.M., 1999. Heterogeneous feeding in benthic suspension feeders: the natural diet and grazing rate of the temperate gorgonian *Paramuricea clavata* (Cnidaria: octocorallia) over a year cycle. *Mar. Ecol. Prog. Ser.* 183, 125–137.
- Ribes, M., Coma, R., Rossi, S., 2003. Natural feeding of the temperate asymbiotic octocoral-gorgonian *Leptogorgia sarmentosa* (Cnidaria: octocorallia). *Mar. Ecol. Prog. Ser.* 254, 141–150.
- Rossi, S., Ribes, M., Coma, R., Gili, J.M., 2004. Temporal variability in zooplankton prey capture rate of the passive suspension feeder *Leptogorgia sarmentosa* (Cnidaria: octocorallia), a case study. *Mar. Biol.* 144, 89–99.
- Rossi, S., Bramanti, L., Gori, A., Orejas, C., 2017a. An overview of the animal forests of the world. In: Rossi, S., Bramanti, L., Gori, A., Orejas, C. (Eds.), *Marine Animal Forests: the Ecology of Benthic Biodiversity Hotspots*. Springer Germany, pp. 1–26.
- Rossi, S., Bramanti, L., Gori, A., Orejas, C., 2017b. Benthic-pelagic coupling: new perspectives in marine animal forests. In: Rossi, S., Bramanti, L., Gori, A., Orejas, C. (Eds.), *Marine Animal Forests: the Ecology of Benthic Biodiversity Hotspots*. Springer Germany, pp. 1–26.
- Ruggieri, N., Castellano, M., Misić, C., Gasparini, G., Cattaneo-Vietti, R., Povero, P., 2006. Seasonal and interannual dynamics of a coastal ecosystem (Portofino, Ligurian Sea) in relation to meteorological constraints. *Geophys. Res. Abstr.* 8, EGU06-A-07774.
- Sameoto, D., Wiebe, P., Runge, J., Postel, L., Dunn, J., Miller, C., Coombs, S., 2000. Collecting zooplankton. p. 55–81. In: Harris, R.P., Wiebe, P.H., Lenz, J., Skioldal, H. R., Huntley, M. (Eds.), *ICES Zooplankton Methodology Manual*. Academic Press, San Diego, California.
- Sherwood, O.A., Jamieson, R.E., Edinger, E.N., Wareham, V.E., 2008. S, C and N isotopic composition of cold-water corals from the Newfoundland and Labrador continental slope: examination of trophic, depth and spatial effects. *Deep Sea Research Part I* 55, 1392–1402.
- Tazioli, S., Bo, M., Boyer, M., Rotinsulu, H., Bavestrello, G., 2007. Ecology of some common antipatharians from the marine park of bunaken (north sulawesi, Indonesia). *Zool. Stud.* 46, 227–241.
- Vander Zanden, M.J., Rasmussen, J.B., 2001. Variation in $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ trophic fractionation: implication for aquatic food web studies. *Limnol. Oceanogr.* 46, 2061–2066.
- Wagner, D., Luck, D.G., Toonen, R.J., 2012. The biology and ecology of black corals (Cnidaria: anthozoa: hexacorallia: antipatharia). *Adv. Mar. Biol.* 63, 67.
- Warner, G.F., 1977. On the shapes of passive suspension feeders. In: Keegan, B.F., PO, Ceidigh, Boaden, P.J.S. (Eds.), *Biology of Benthic Organisms*. Pergamon, Oxford, pp. 567–576.
- Warner, G.F., 1981. Species descriptions and ecological observations of black corals (Antipatharia) from Trinidad. *Bull. Mar. Sci.* 31, 147–163.
- Williams, B., Grotoli, A.G., 2010. Stable nitrogen and carbon isotope ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) variability in shallow tropical Pacific soft coral and black coral taxa and implications for paleoceanographic reconstruction. *Geochem. Cosmochim. Acta* 74, 5280–5288.