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KEYWORDS

Food web – Meiofauna - Intertidal seagrass bed - Microphytobenthos - Stable isotope ratio – Nematode – Copepod

ABSTRACT

In an intertidal *Zostera noltii* Hornem. seagrass bed, food sources used by sediment meiofauna were determined seasonally by comparing stable isotope signatures ($\delta^{13}C$, $\delta^{15}N$) of sources with those of nematodes and copepods. Proportions of different carbon sources used by consumers were estimated using the SIAR mixing model on $\delta^{13}C$ values. Contrary to $\delta^{15}N$ values, food source mean $\delta^{13}C$ values encompassed a large range, from -22.1‰ (suspended particulate organic matter) to -10.0‰ (*Z. noltii* roots). $\delta^{13}C$ values of copepods (from -22.3 to -12.3‰) showed that they use many food sources (benthic and phytoplanktonic microalgae, *Z. noltii* matter). Nematode $\delta^{13}C$ values ranged from -14.6 to -11.4‰, indicating a strong role of microphytobenthos and/or *Z. noltii* matter as carbon sources. The difference of food source uses between copepods and nematodes is discussed in the light of source accessibility and availability.

INTRODUCTION

Seagrass beds are widespread in shallow coastal waters and are considered one of the most productive marine ecosystems in the world (Duarte and Chiscano 1999). Seagrass beds support a high diversity of consumers because they provide a wide range of potential food sources including seagrass leaves, roots and detrital matter, epiphytes, microphytobenthos, bacteria and allochtonous inputs of organic matter (Valentine and Duffy 2006). These food sources have different levels of digestibility (Cebrián 1999) and some of them (*e.g.* seagrass, phytoplankton) have a large seasonal pattern of production (Duarte 1989; Borowitzka et al. 2006), affecting consumer population structure and diets (Escavarage et al. 1989; Danovaro 1996; Danovaro and Gambi 2002).

Meiofauna are often characterized by high densities on seagrass leaves (*i.e*. harpacticoid copepods) (Bell et al. 1984; De Troch et al. 2001) and in surface sediments (Escavarage et al. 1989; Danovaro et al. 2002). Meiofaunal biomass is often high in seagrass sediments, from 1.0 to 2.4 g $C.m^{-2}$ (Escavarage et al. 1989; Danovaro et al. 2002), oftentimes higher than in unvegetated areas (Castel et al. 1989; Fonseca et al. 2011; Leduc and Probert 2011). Because of their short life cycle and high turnover rates (Hicks and Coull 1983; Heip et al. 1985), meiofaunal communities are thought to respond rapidly to organic matter inputs and may be closely coupled with primary production inputs (Escavarage et al. 1989). Meiofauna have high estimated secondary production rates, from 9.05 to 29.40 g $C.m^{-2}.yr^{-1}$ (Escavarage et al. 1989; Danovaro et al. 2002), and may play a key role in benthic energy flows. Meiofauna also represent a direct link between primary producers and higher trophic levels because meiofauna (harpacticoid copepods in particular) are a common prey item of fish and shrimps (Coull 1999; Hyndes and Lavery 2005).

Food sources of meiofauna are not well characterized, despite their potentially important role in the trophic dynamics of seagrass beds. This is due to the small size of these animals, rendering their sorting and study complex. At the species level harpacticoid copepods and nematodes often have specialized diets (Rieper 1982; Romeyn and Bouwman 1983; Buffan-Dubau et al. 1996; Moens and Vincx 1997; Rzeznik-Orignac et al. 2008). Nevertheless, changes of feeding strategies can be observed depending on the availability of food sources, both at community (Riera and Hubas 2003; Hyndes and Lavery 2005) and species level (Moens and Vincx 1997). Variations of food resource availability may thus influence meiofaunal community structure (Escavarage et al. 1989; Danovaro 1996; Danovaro and Gambi 2002).

Seagrass beds are ecosystems where food sources typically exhibit large variations of quality, quantity and availability throughout the year (Duarte 1989; Lebreton et al. 2009). Production of seagrass beds is usually high (Cebrián 1999) but follows strong temporal patterns of growth (Duarte 1989; Lebreton et al. 2009). Some vertebrate or invertebrate consumers directly use seagrass organic matter (Valentine and Heck, 1999) but a large part of it becomes detritus (Cebrián, 1999). Detrital organic matter constitutes a substrate for the development of bacteria (Anesio et al. 2003; Holmer et al. 2004), which may represent a food source for meiofauna (Danovaro 1996). Another food source is microphytobenthos. It often exhibits high production rates in seagrass beds (Asmus and Asmus 1985; Daehnick et al. 1992; Kaldy et al. 2002), it is available to consumers and is easily digestible (Duarte and Cebrián 1996). Microphytobenthos can also be a major carbon source for bacteria in seagrass beds (Boschker et al. 2000). Very few studies have addressed microphytobenthos fate in seagrass bed food webs and *a fortiori* the contribution of microphytobenthos as a food resource to meiofauna (Leduc et al. 2009).

Stable isotopes are commonly used to study trophodynamics in ecosystems (Fry 2006). Configurations for isotopic composition determination, requiring around 100 µg of matter or less (Carman and Fry 2002), now enable the measurement of $\delta^{13}C$ and $\delta^{15}N$ values of meiofaunal communities and the ability to determine their food resources (Riera et al. 1996; Riera and Hubas 2003). Contrary to gut content analyses, stable isotope analyses allow determination of food sources actually assimilated in the tissues of consumers over time, properly reflecting their trophodynamics depending on food source availability (Fry 2006). A limit of this technique is that stable isotope signatures sometimes overlap, making data interpretation difficult.

The aim of this study is to describe, using natural abundances of ^{13}C and ^{15}N, the food sources used by nematodes and copepods in the sediment of an intertidal *Zostera noltii* Hornem. seagrass bed in Marennes-Oléron Bay over an annual cycle. Copepod and nematode food resources are characterized by comparisons between their stable isotope ratios and those of the available food sources.

MATERIAL AND METHODS

Study area

The study was carried out in a *Zostera noltii* meadow in Marennes-Oléron Bay, a semi-enclosed system along the French Atlantic coast. This macrotidal bay (tidal range 0.9-6.5 m),

99 located between Oléron Island and the mainland, receives continental water mainly from the
100 Charente River, which flows out into the eastern side of the bay. Tidal current speeds range
101 from 0.04 to 0.27 $m.s^{-1}$ (Struski, unpublished data). The studied seagrass bed is located on the
102 western side of the bay, along Oléron Island, where the bay is more strongly influenced by
103 offshore water (Dechambenoy et al. 1977). The *Z. noltii* bed extends over 15 km along the
104 shore and is 1.5 km wide in the upper part of the flat, limited in its lower part by extensive
105 oyster farm structures (Guillaumont 1991).
106 The sampling station (45°54'32.0'' N, 1°12'50.3'' W) was located at about 250 m from
107 the upper limit of the intertidal seagrass bed. At this station, sediment is composed of silty
108 fine sand (Weber, 2003). Mean emersion time is about 5 hours per tide and the mean water
109 level is about 1.80 m during immersion. Biomass fluctuations of food resources, meiofauna
110 and macrofauna were studied in 2006 and 2007 (Lebreton et al. 2009; Lebreton unpublished
111 data).
112
113 ***Sampling and preparation of primary producers and composite food sources***
114 Seagrass roots and leaves, suspended particulate organic matter (SPOM) and surface
115 sediment fine organic matter (SSOM) were sampled seasonally from winter 2006 to summer
116 2007 (winter 2006: 02/16, spring 2006: 05/22, summer 2006: 08/09, fall 2006: 11/09, winter
117 2007: 02/21, spring 2007: 05/29, summer 2007: 07/31). Microphytobenthos was sampled
118 from winter 2007 to fall 2007 (winter 2007: 02/21, spring 2007: 05/29, summer 2007: 07/31,
119 fall 2007: 11/28), and a complementary sampling was carried out in winter 2010 (02/21).
120 Epiphytes were sampled in spring (05/29) and summer 2007 (08/02). Data from food sources
121 sampled in winter and summer 2006 and 2007 are given by Lebreton et al. (2011).
122 Below-ground and above-ground parts (roots and leaves, respectively) of *Zostera noltii*
123 were rinsed with tap water to remove detrital fragments. Detrital matter from *Z. noltii* – made
124 up of pieces of leaves or roots dark brown to black colored - was collected by sieving
125 sediment on a 500-µm sieve and then washed. Samples were freeze-dried then ground to a
126 fine and homogeneous powder using a ball mill. Microphytobenthos samples were collected
127 by scraping surficial sediment on the field and then by extracting microalgae in the laboratory
128 following the method of Riera and Richard (1996), slightly modified by Herlory et al. (2007).
129 Extracted samples were checked under a microscope for purity, then concentrated by
130 centrifugation (10 min, 1000 ×g) and freeze-dried. Microphytobenthos was mainly made up
131 of a large diversity of small pennate diatoms either epipelic, like *Amphora acutiuscula* and
132 *Navicula heterovalvata*, or epipsammic, like *Achnanthes minuscula*, *Plagiogramma*

staurophorum and *Plagiogrammopsis vanheurckii*, as observed by Bogaczewicz-Adamczak (unpublished data). Epiphytes samples were composed of two species of diatoms (*Cocconeis scutellum* and *C. placentula*) (Lebreton et al. 2009). These diatoms were separated from leaves by agitation following the procedure described by Lebreton et al. (2011).

Stable isotope analyses were also carried out on composite food sources - *i. e.* SSOM and SPOM – which composition results from a mix of alive or detrital primary producers. For SSOM analyses, surface sediment (top first cm) was sieved wet on a 315-µm sieve to remove large detritus and macrofauna. Sediment was freeze-dried, ground using a mortar and pestle then acidified to remove carbonates using 1 $mol.L^{-1}$ HCl. HCl was added drop-by-drop until cessation of bubbling. Samples were then dried at 60°C using a dry bath under air flow. Dried samples were re-homogenized into ultrapure water using an ultrasonic bath. Sediment samples were then freeze-dried again and re-grinded. SPOM from surface water was sampled close to the seagrass bed (45°55'50.4'' N, 1°10'12.0'' W) at mid-tide, biweekly to monthly. A volume of seawater from 50 to 60 mL was pre-filtered on a 200-µm sieve to eliminate large zooplankton and detrital particles. Then water was filtered on precombusted Whatman GF/F fiber glass filters (0.7 µm porosity) under moderate vacuum. Filters were freeze dried then acidified using HCl fumes to remove carbonates. All samples were stored at –20°C before analysis.

Sampling and preparation of meiofauna for isotope analyses

Sampling was carried out following the seasonal cycle (spring 2007: 05/05, summer 2007: 09/10, fall 2007: 11/28, winter 2008: 03/18). Meiofauna were collected by scraping about 0.25 m^2 of surficial sediment. A surface sediment layer, upper first cm of sediment, and a subsurface sediment layer, from 1 to 4 cm deep, were sampled separately in order to study both meiofauna communities. Surface sediment layer was light brown colored whereas subsurface sediment layer was dark brown to black colored, which was the evidence of anoxic conditions in this last layer. Surface sediment layer was scrapped using a spatula until the required depth (1 cm) was reached. Depth was measured using steel rulers pushed in sediment and regularly disposed on the scrapped area. The same procedure was applied to sample the subsurface sediment layer on the previously scrapped area. In the laboratory, sediment samples were sieved on a 1 mm-mesh sieve to eliminate fresh and detrital *Zostera noltii* matter, macrofauna and shells. Sieved sediment was then stored at 18°C and meiofauna were extracted within 24 hours following field sampling.

Some conditions are necessary to determine the isotopic composition of meiofauna. Large numbers of individuals must be extracted from the sediment to get enough material for analyses, and the extracted population must be representative of the whole community. Moreover, meiofauna must be extracted alive and kept in filtered seawater to allow evacuation of gut contents. Some authors used methods based on downward migration of nematodes under permanent light, taking advantage of the negatively phototactic behavior of nematodes (Riera et al. 1996; Rzeznik-Orignac et al. 2008). However these methods do not allow a complete and representative extraction of the community (Rzeznik-Orignac et al. 2004), particularly for copepods, that may have some influence on isotopic signatures. Other protocols are based on successive elutriation and centrifugation procedures using the colloidal silica Ludox™ HS 40 (Heip et al. 1985; Rzeznik-Orignac et al. 2004) or $MgSO_4$, adjusted with distilled water to the meiofauna density of 1.130 (Somerfield et al. 2005). These methods give more representative samples of meiofauna (nematodes and copepods) but are performed on fixed or frozen samples (Giere 2009).

We used a similar Ludox-based protocol but slightly modified as to keep meiofauna alive. The usual procedure involves a thorough rinse of the sediment with distilled water to remove the interstitial seawater containing some ions, such as Ca^{2+} and Mg^{2+}, which turn the Ludox™ solution to a gel (de Jonge 1979). This procedure gave low recovery of live meiofauna. We therefore tested extractions with solutions of higher osmolarity, *i.e.* NaCl solutions at 20 g L^{-1} and at 30 g L^{-1}, both for the Ludox™ dilution and the sediment rinse. Both protocols gave a recovery of fully alive meiofauna. Since the NaCl 20 g L^{-1} solution method gave clean samples (*i.e.* absence of detrital matter, diatoms…), it was used throughout this study.

Fifty ml of sediment were mixed with 200 ml of a 20 g L^{-1} NaCl solution, vigorously shaken then centrifuged at 2500 ×g during 1 min at 10°C. The supernatant was discarded and this step was repeated once. Meiofauna were extracted by addition of 200 ml of Ludox™ HS 40 adjusted to a density of 1.130 with a 20 g L^{-1} NaCl solution. Samples were vigorously shaken then centrifuged at 2500 ×g during 4 min at 10°C. The supernatant, containing meiofauna, was collected and the resultant pellet was processed once again following the same procedure. Burgess (2001) observed very high extraction efficiencies of meiofauna taxa using this technique after one extraction (nematodes: 97.4%, copepods: 96.0%). Repeating the procedure yielded very few individuals, and increased mortality (Lebreton pers obs). Supernatants with meiofauna were thoroughly rinsed on a 40-µm sieve, firstly with a solution of NaCl at 20 g L^{-1} in order to wash the Ludox™ HS 40 from samples, then with filtered sea water. Sample rinsing was always performed just after supernatant collection in order to

quickly remove meiofauna from Ludox™ HS 40. Both supernatants from same samples were pooled then their quality (state of meiofauna, absence of detrital matter) was checked under binocular. Meiofauna were kept alive in Petri dishes during 12 hours at 18°C in filtered seawater to allow evacuation of gut contents (Buffan-Dubau et al. 1996; Riera et al. 1996), before storage at -20°C without preliminary sorting.

After thawing, sorting of samples was done in a Dollfus counting chamber in which nematodes and copepods were picked with fine forceps (Dumont #55). They were then washed in distilled water (Milli Q), counted and brought together in a tin capsule for stable isotope analysis, which had been previously weighed (Microbalance Sartorius ME5, ± 0.001 mg) and filled with 300 µl of ultrapure water. Water was then evaporated under vacuum in a desiccator containing P_2O_5 and capsules were precisely weighted (± 0.001 mg). When possible, three replicates of 300 nematodes or copepods (about 100 µg DW) were done per season and per sediment layer (surface or subsurface). Due to lack of material, only one sample per season was analyzed for subsurface copepod communities.

Stable isotope ratios and data analyses

Samples were analyzed using an elemental analyzer (Flash EA 1112, Thermo Scientific, Milan, Italy) coupled to an isotope ratio mass spectrometer (Delta V Advantage with a Conflo IV interface, Thermo Scientific, Bremen, Germany). Results are expressed in the δ unit notation as deviations from standards (Vienna Pee Dee Belemnite for $\delta^{13}C$ and N_2 in air for $\delta^{15}N$) following the formula: $\delta^{13}C$ or $\delta^{15}N = [(R_{sample}/R_{standard})-1] \times 10^3$, where R is $^{13}C/^{12}C$ or $^{15}N/^{14}N$. Calibration was done using reference materials (USGS-24, IAEA-CH6, IAEA-600 for carbon; IAEA-N1, -N2, -N3, -600 for nitrogen). Analytical precision based on analyses of acetanilide (Thermo Scientific) used as laboratory internal standard was < 0.06‰ and < 0.1‰ for carbon and nitrogen, respectively.

Comparisons between stable isotope values were conducted using non-parametric procedures, which are more powerful than parametric statistics for small sized samples (replicate numbers almost always < 10) (Zar 2011). Kruskal-Wallis tests were used: 1. to compare stable isotope signatures of the different food sources (all seasons merged), 2. to study seasonal variations of food source and consumer signatures among seasons. Kruskal-Wallis tests were followed by multiple comparisons of means by using the pgirmess package (Giraudoux 2011) of the R software (R Development Core Team 2008). Mann-Whitney-Wilcoxon tests were applied: 1. to compare stable isotope signatures of nematodes between surface and subsurface sediment samples, 2. to compare nematode and copepod signatures.

Seasonal composition of SSOM was defined by using the mixing model developed in the R package SIAR (Parnell et al 2010). Only $\delta^{13}C$ values were used for computations and no trophic enrichment was weighted into computations. Only SPOM, microphytobenthos and *Z. noltii* detrital matter were taken into account because *Z. noltii* (leaves and roots) and epiphytes were considered as not being part of SSOM (Lebreton et al 2009, 2011). Models were run for 500,000 iterations and the first 50,000 iterations were discarded. Credibility intervals (CI) of 0.95, 0.75 and 0.25 were computed and displayed on figures. Only the lowest and highest limits of 0.95 CI were detailed in the manuscript. Credibility intervals are used in Bayesian statistics to define the domain of *a posteriori* probability distribution used for interval estimation (*e. g*. if the 0.95 CI of a contribution value ranges from A to B, it means that there is a 95% chance that the contribution value lies between A and B) (Edwards et al. 1963).

Isotopic ratios of consumers and food sources were compared considering a trophic enrichment of 0.3‰ for $\delta^{13}C$ values and of 2.3‰ for $\delta^{15}N$ values (Vander Zanden and Rasmussen 2001). Contributions of carbon food sources for nematodes and copepods were estimated by running SIAR mixing model on $\delta^{13}C$ values. SIAR does not accept missing data. $\delta^{15}N$ values were not included in the model due the lack of few $\delta^{15}N$ data of some food sources. In order to reduce the number of food sources in calculations, *Z. noltti* leaves and roots were considered as a single group called *Z. noltii* fresh matter. SSOM was not taken into account because its composition was primarily based on three other food sources already included: *Z. noltii* detrital matter, microphytobenthos and pelagic algae (Lebreton et al 2011), signatures of which were assumed to be close to those of SPOM. Due to the lack of epiphyte $\delta^{13}C$ value in winter, the winter value used in the model was the overall mean value of the three other seasons. Trophic enrichment used for computations was 0.3±1.3‰ (mean ± standard deviation) (Vander Zanden and Rasmussen 2001). Running model parameters were the same as for SSOM.

RESULTS

Stable isotope signatures of primary producers

Mean $\delta^{13}C$ values ranged from -15.5‰ (microphytobenthos, winter 2007) to -9.3‰ (roots, spring 2006) (Table 1). Microphytobenthos was characterized by the most depleted mean annual $\delta^{13}C$ value (-14.1‰), followed by *Zostera noltii* detrital matter (-12.7‰), epiphytes (-11.6‰), *Z. noltii* leaves (-10.3‰) and roots (-10.0‰). $\delta^{13}C$ values of *Z. noltii* (roots, leaves, detrital matter) showed no clear seasonal pattern (Table 1). Due to analytical issues, $\delta^{15}N$

267 values of microphytobenthos samples could not be measured in winter 2007. Mean $\delta^{15}N$
268 values ranged from 4.9 (leaves, winter 2006) to 11.0‰ (roots, fall 2006) (Table 1). No clear
269 seasonal pattern was observed among $\delta^{15}N$ values of primary producers.
270
271 ***Stable isotope signatures of composite food sources***
272 Mean $\delta^{13}C$ values of SPOM ranged from -23.5‰ (spring 2006) to -21.1‰ (spring 2007)
273 (Table 1). SPOM presented significantly lower $\delta^{13}C$ values than SSOM, microphytobenthos,
274 epiphytes, *Z. noltii* roots, leaves and detrital matter (Kruskal-Wallis test, $P < 0.001$). Due to
275 analytical issues, $\delta^{15}N$ values of SPOM samples could not be determined.
276 Stable isotope signatures of SSOM ranged from -19.1‰ (spring 2007) to -17.4‰ (winter
277 2007) and from 5.0 (spring 2006) to 6.8‰ (summer 2006) for $\delta^{13}C$ and $\delta^{15}N$, respectively
278 (Table 1). Stable isotope composition of SSOM showed no clear seasonal pattern (Table 1).
279 SSOM was significantly more depleted in ^{15}N than Z. *noltii* roots, leaves and detrital matter
280 (Kruskal-Wallis tests, $P < 0.001$).
281 Mixing model estimations of contributions showed that SSOM was primarily composed
282 of settled SPOM and secondarily of microphytobenthos and *Z. noltii* detrital matter (Fig. 1).
283 All seasons put together, 0.95 CI ranged in fact from 15 to 81%, from 0 to 60% and from 0 to
284 54% for SPOM, microphytobenthos and *Z. noltii* detrital matter, respectively. Contribution of
285 SPOM to SSOM was particularly high in fall, with 0.95 CI of SPOM ranging from 44 to 81%.
286
287 ***Copepods: stable isotope signatures and mixing model estimations of contributions***
288 Copepod abundance was very low in subsurface (1-4 cm) sediment layer (Lebreton
289 unpubl data). As a result, none or only one sample of copepods was collected per season in
290 this sediment layer. When they were determined, isotopic signatures of subsurface samples
291 were always in the range of those of surface (0-1 cm) samples. Surface and subsurface
292 communities have thus been considered as coming from the same community thereafter.
293 Copepods presented a wide range of $\delta^{13}C$ values (-22.3 to -12.3‰). Range of $\delta^{15}N$ values
294 was smaller, with values from 6.6 to 8.9‰ (Fig. 2). No significant seasonal variations of
295 copepod signatures were observed (Table 2). For $\delta^{13}C$ signatures, this absence of difference is
296 probably related with the large standard deviations observed at most seasons. The difference
297 between the average of food sources $\delta^{15}N$ values (SSOM, microphytobenthos and *Z. noltii*
298 detrital matter) and copepods $\delta^{15}N$ values was 1.1‰ on average (range from 0.0 to 2.2‰).
299 In summer, mixing model gives higher upper and lower limits of 0.95 CI (from 16 to
300 49%) for SPOM relative to epiphytes and *Z. noltii* fresh and detrital matter, for which 0.95 CI

ranged from 0 to 36% (Fig. 3). At other seasons, ranges of food source contributions are large and relatively equal: SPOM: 0.95 CI from 1 to 45%, epiphytes: 0.95 CI from 0 to 37%, *Z. noltii* detrital matter: 0.95 CI from 0 to 39%, *Z. noltii* fresh matter: 0.95 CI from 0 to 37%. Contributions of microphytobenthos to copepod carbon sources are equal all year long (0.95 CI from 0 to 41%).

Nematodes: stable isotope signatures and mixing model estimations of contributions

Stable isotope signatures of nematodes from surface and subsurface samples were similar at all sampling dates (Table 3). Thus, they have been considered as a single community thereafter. Isotopic signatures ranged from -14.7 to -11.4‰ for $\delta^{13}C$ and from 8.5 to 11.6‰ for $\delta^{15}N$. Considering the trophic enrichment, the theoretical signatures of nematode's food sources are close to those of *Z. noltii* detrital matter and of microphytobenthos (Fig. 2). The difference between the average of food sources $\delta^{15}N$ values (microphytobenthos and *Z. noltii* detrital matter) and nematodes $\delta^{15}N$ values is equal to 2.7‰ in average and ranged from 1.4 to 4.4‰. Nematode $\delta^{13}C$ values were more depleted in summer (-13.6 ± 0.6‰) than in fall and winter (-12.2 ± 0.5‰ and -12.5 ± 0.2‰, respectively; Kruskal-Wallis tests, $P < 0.001$) (Table 2). $\delta^{15}N$ values were higher in winter than in summer (10.7 ± 0.7‰ *vs.* 8.9 ± 0.3‰, Kruskal-Wallis tests, $P < 0.001$). Mixing model computations show relatively equal contributions of microphytobenthos, epiphytes and fresh and detrital *Z. noltii* organic matter as carbon sources for nematodes (0.95 CI ranging from 0 to 46%, Fig. 3). SPOM contributions are low, with 0.95 CI ranging from 1 to 24%.

Copepods and nematodes showed similar signatures for $\delta^{13}C$ in fall and winter (Mann-Whitney-Wilcoxon tests, fall: $P = 0.095$, winter: $P = 0.095$) and for $\delta^{15}N$ in fall (Mann-Whitney-Wilcoxon test, $P = 0.071$). Copepods presented lower $\delta^{13}C$ values than nematodes in spring and summer (Mann-Whitney-Wilcoxon tests, spring: $P = 0.010$, summer: $P = 0.009$), with $\delta^{13}C$ values 2.1‰ less enriched than nematodes. $\delta^{15}N$ values of copepods were lower than these of nematodes in winter, spring and summer (Mann-Whitney-Wilcoxon tests, winter: $P = 0.024$, spring: $P = 0.010$, summer: $P = 0.010$), with values 3.0 ‰ less enriched than nematodes.

DISCUSSION

Stable isotope signatures of primary producers

$\delta^{13}C$ values well discriminate the potential food sources, except between *Z. noltii* leaves and roots and between microphytobenthos and *Zostera noltii* detrital matter, mainly because of the large seasonal variations of microphytobenthos $\delta^{13}C$ signatures. These food source signatures are well within the range of previous observations already made in the same seagrass bed (Kang et al. 1999) and in others (Boschker et al. 2000; Leduc et al. 2006; Kharlamenko et al. 2008; Schaal et al. 2008). Microphytobenthos signatures are also similar to those observed for this food source in adjacent bare mudflats (Riera and Richard 1996; Rzeznik-Orignac et al. 2008). The ^{13}C-depletion observed between live and detrital *Z. noltii* matter is probably due to the higher levels of lignin, which is a ^{13}C-depleted component, in detrital material (Benner et al. 1987).

Origin and composition of composite food sources

SPOM values (from –23.5 to –21.1‰) are ranging from those of marine (-19.1‰) and estuarine phytoplankton (-23.5‰) observed by Riera and Richard (1996) in Marennes-Oléron Bay. SPOM was thus composed of a mix of both these primary producers, possibly with a different influence of the former or the latter depending on seasons.

SSOM $\delta^{13}C$ values were intermediate between those of SPOM and those of two benthic sources: microphytobenthos and *Zostera noltii* detrital matter. Mixing model contributions thus suggest that SSOM is clearly composed of a high amount of settled SPOM. SSOM is also composed of microphytobenthos and *Z. noltii* detrital matter. Because stable isotope signatures of these sources are close, mixing model results do not allow determining contributions of benthic sources (*i. e.* microphytobenthos and *Z. noltii* detrital matter) to SSOM. Nevertheless, Lebreton et al. (2011) ruled out the hypothesis of a high content of *Z. noltii* fine particles (<315 µm) in SSOM, because very low amounts of seagrass fatty acid markers were observed in SSOM. This weak contribution of *Z. noltii* matter was unexpected considering the high amount of large (>500 µm) *Z. noltii* detrital particles in the sediment (Lebreton et al. 2009). This high amount was however measured on 15 cm deep cores, so *Z. noltii* detrital matter is likely stored as large particles in sediment or buried deeper than the first top centimeter sediment layer.

This weak influence of *Z. noltii* to SSOM composition (Lebreton et al. 2009) thus reinforces the role of microalgae, either benthic or pelagic when settled, in the functioning of this intertidal seagrass bed. Food source contributions to SSOM originating from mixing model computations are therefore probably a few overestimated for *Z. noltii* and a few underestimated for pelagic and benthic microalgae. These algae were primarily originating

from phytoplankton and secondarily from microphytobenthos. Contribution of settled pelagic microalgae was particularly high in fall, which was confirmed with relatively high amount of flagellate fatty acid markers, particularly 22:6(*n*-3), observed in SSOM (Lebreton unpublished data). Flagellate densities are generally low in sediment (Pascal et al. 2009) and high in water column (Galois et al. 1996), indicating that flagellate organic matter from SSOM was mainly issued from SPOM. The *Z. noltii* seagrass bed thus traps high quantities of SPOM into sediment because seagrass canopy weakens hydrodynamics (Koch et al. 2006), making settled SPOM available to benthic consumers.

Food sources used by benthic copepod communities

Mixing model estimations of contributions and the very large range of copepod $\delta^{13}C$ values suggest that these consumers can use many food sources: SPOM (*i. e.* settled pelagic microalgae), microphytobenthos, epiphytes and *Zostera noltii* matter. The small differences of $\delta^{15}N$ values between these food sources and copepods (average equal to 1.1‰) suggest that they are primary consumers.

The very low $\delta^{13}C$ value (-22.3‰) noticed in spring and the high contribution of SPOM to copepod carbon sources observed in summer indicate an increased feeding on a mixture of marine/estuarine phytoplankton, likely due its higher availability during blooms occurring at these periods (Galois et al. 1996). Higher amounts of SPOM in SSOM during fall (see previous section) did not clearly affect copepod isotopic composition, demonstrating that copepod food sources were not directly depending on SSOM composition and suggesting that copepods probably perform some selection on food sources in SSOM. Copepod community is in fact probably composed of different feeding mode individuals, like surface-dwelling consumers, which directly graze settled phytoplankton cells at sediment surface, and endobenthic individuals, which performed filter-feeding, like Canuellidae (De Troch et al. 2003; Hicks and Coull 1983).

Copepods also rely on benthic ^{13}C-enriched food resources (*i. e.* microphytobenthos, epiphytes and/or *Z. noltii* matter). The high contribution of epiphytes as a carbon source for copepods is most probably an artifact of the mixing model in relation with epiphyte $\delta^{13}C$ values, which are intermediate between those of microphytobenthos and of *Z. noltii* matter. Epiphytes are absent in winter, due to the absence of seagrass leaves, and epiphyte biomass is very low during other seasons in Marennes-Oléron seagrass bed, with biomass at least 1000 times less than those of microphytobenthos or *Z. noltii* detrital matter (Lebreton et al, 2009). Thus, even if epiphytes could be eaten by benthic copepods, they most probably represent a

very weak carbon source. The second main contribution for copepods is thus microphytobenthos and/or *Z. noltii* matter. The relatively close stable isotope signatures between these food sources do not allow clearly determining which of these food sources, or if both of them, are used by copepods.

This large range of food source intakes (SPOM, microphytobenthos and *Z. noltii* matter) and their seasonal variations well correspond with the opportunistic trophic behavior of benthic copepods, which are known to feed on microalgae (Hicks and Coull 1983; De Troch et al. 2006) and also on detritus (Rieper-Kirchner 1990). In seagrass beds, the studies conducted by Leduc et al. (2009) and Hyndes and Lavery (2005) on copepods from sediment also showed that they are opportunistic feeders, feeding either on macroalgae or seagrass detrital matter, SPOM, microphytobenthos or bacteria, depending on resource availability.

Food resources of nematodes: microphytobenthos and seagrass matter

Except in fall, mean $\delta^{15}N$ values of nematodes were higher than those of copepods. Taking an average trophic-level increase in $\delta^{15}N$ values of 2.5‰ for first level consumers and of 3.4‰ for higher level consumers (Vander Zanden and Rasmussen 2001; Moens et al. 2005), the difference between $\delta^{15}N$ values of food sources and of nematodes (ranging from 1.4 to 4.4‰) spans one or two trophic levels. This large range - with higher values than for copepods - could be explained by two reasons: 1. Nematode may rely on higher quantities of *Z. noltii* detrital matter than copepods. Detritivores are thought to derive their nutrition from bacteria associated with detrital particles (*e.g. Z. noltii* detrital matter) and not from the non-living plant substrate, particularly for nitrogen (Findlay and Tenore 1982). This mediation increases $\delta^{15}N$ values in bacteria (Dijkstra et al. 2008) and then in nematodes due to trophic isotopic enrichment. 2. Nematodes are characterized by different feeding modes: selective or non-selective deposit-feeders, epistrate-feeders (*i.e.* protists and microalgae consumers) or omnivore-carnivores (Wieser 1953; Romeyn and Bouwman 1983). These feeding modes place them at different trophic levels and $\delta^{15}N$ values of nematodes are related to these trophic levels (Carman & Fry, 2002; Moens et al. 2005; Rzeznick et al. 2008). Thus, the high range observed for $\delta^{15}N$ values is probably representative of this range of feeding behaviors.

Mixing model results suggest relatively similar contributions of microphytobenthos, epiphytes and *Zostera noltii* fresh and detrital matter to nematode carbon sources at all sampling dates. The ^{13}C-enriched composition of nematodes and the low contributions of SPOM computed with SIAR demonstrate a lower use of SPOM by nematodes in comparison with copepods. As already detailed for copepods, we suggest that the high contribution of

435 epiphytes as nematode carbon source is an artifact of the mixing model (see previous section).
436 Moreover, accessibility of epiphytes to nematodes is probably very low, due to the
437 exclusively benthic behavior of these consumers. Contributions of other food sources are thus
438 probably slightly underestimated.
439 In the seagrass bed, nematodes thus mostly rely on microphytobenthos and *Zostera noltii*
440 matter. The close isotopic composition between microphytobenthos, fresh and detrital *Z. noltii*
441 matter does not clearly allow determining which one or if all these food resources are used by
442 nematodes. Nevertheless, none of the known marine nematode feeding modes has ever been
443 demonstrated to be capable of directly grazing living macrophyte tissue (Wieser, 1953;
444 Romeyn and Bouwman, 1983; Moens and Vincx, 1997), suggesting that *Z. noltii* matter is
445 only eaten as detrital by nematodes. When detrital, *Z. noltii* organic matter is probably
446 assimilated through bacteria mediation (Findlay and Tenore, 1982). Nematofauna community
447 presents a large diversity of feeding modes (Wieser, 1953; Romeyn and Bouwman, 1983;
448 Moens and Vincx, 1997). The range of nematode $\delta^{13}C$ values thus includes herbivores and
449 bacterivores, ^{13}C-depleted, but also carnivores, ^{13}C-enriched. These ^{13}C-enriched values can
450 explain the high contributions of fresh *Z. noltii* matter computed through SIAR, which may be
451 overestimated.
452 In another intertidal seagrass bed, Leduc et al. (2009) observed that nematodes also rely
453 mainly on microphytobenthos and on seagrass detrital matter. Improving upon this previous
454 study, our work follows seasonal variations of nematode food sources: on a temporal view,
455 nematode signatures are generally closer to those of microphytobenthos in summer and to
456 those of *Zostera noltii* matter from fall to winter. This seasonal change is probably related
457 with the higher quantities of *Z. noltii* detrital matter in sediment from fall to winter (Lebreton
458 et al. 2009), making this resource more available to nematodes. These variations probably
459 cause changes of food resources at two scales for nematodes. At the individual scale, the
460 nematodes characterized by opportunistic feeding behavior probably rely on availability of
461 sources (Riera and Hubas 2003). At the community scale, there is also probably an evolution
462 of the structure of the nematode community, depending on nematode trophic behaviors
463 (Escavarage et al. 1989; Danovaro 1996; Danovaro and Gambi 2002).
464
465 ***Role of SPOM as meiofauna food source***
466 Even though nematodes weakly use SPOM, this food resource appeared to be largely
467 consumed by copepods. This difference between nematodes and copepods is probably related
468 with availability of food sources to these consumers in relation with their location in the

sediment. Nematodes are able to migrate through sediment layers (Heip et al. 1985) whereas copepods are mostly located in first top millimeters in muddy sediment (Hicks and Coull 1983; Buffan-Dubau and Castel 1996) due to their sensitivity to anoxic conditions (Hicks and Coull 1983). As a result, nematodes may have access to the whole range of sediment food resources (*i. e. Z. noltii* detrital matter, microphytobenthos, trapped SPOM). Exploitation of food sources by nematodes probably reflects the biomass of available resources in both surface and subsurface sediment layers. On the contrary, copepods may mostly access to food resources from the very top sediment layer, which may contain higher quantities of trapped SPOM than underneath sediment layers. This assumption is confirmed by meiofauna densities in this seagrass bed: 12 10^6 individuals m^{-2} and 2.3 10^6 individuals m^{-2} in surface and subsurface sediment layers, respectively, for nematodes and 1.2 10^6 individuals m^{-2} and 8.2 10^4 individuals m^{-2} in surface and subsurface sediment layers, respectively, for copepods (Lebreton unpubl data).

SPOM availability for the copepod community is also influenced by the location (intertidal *vs*. subtidal) and topography of the seagrass bed. Harpacticoid copepods sampled with nets in a Mediterranean subtidal seagrass bed were mainly relying on SPOM and of benthic organic matter (*e.g.* epiphytes) (Vizzini et al. 2002; Vizzini and Mazzola 2003). In addition, Leduc et al (2009) suggested that seagrass bed architecture (*e.g.* large *vs*. thin leaves of *Posidonia vs. Zostera*, respectively) probably has a strong influence on particle trapping, explaining why SPOM has more influence in *Posidonia* beds. On Marennes-Oléron *Zostera noltii* seagrass bed, influence of marine and estuarine SPOM is probably weakened due to the intertidal location of the seagrass bed and thinness of leaves. Phytoplankton is nevertheless an important carbon source for copepods, probably thanks to the high quality of pelagic microalgae as a food resource and to the influence of marine offshore water at this location (Dechambenoy et al. 1977).

The influence of SPOM has been previously observed for suspension feeders in this seagrass bed (Lebreton et al 2011), underlining the role of SPOM in this intertidal ecosystem. Like suspension feeders, benthic copepods mediate a bentho-pelagic coupling by consuming pelagic organic matter, increasing the flux of organic matter from pelagos to benthos. Moreover, by using this organic matter, copepods increase its quality through its transformation into animal tissue and make it more available for strictly benthic consumers.

Role of benthic food sources: microphytobenthos and Zostera noltii matter

As previously observed in other seagrass beds, microalgae are an important food resource for meiofauna; particularly microphytobenthos for nematodes (Vizzini et al. 2002; Vizzini and Mazzola 2003; Leduc et al. 2009). This trophic scheme, in which meiofauna largely use microphytobenthos, is very close to those observed in salt marshes (Riera et al. 1996; Moens et al. 2002), where high production of detrital matter from vascular plants occurs. This demonstrates that meiofauna can favor microphytobenthos even if high amounts of organic matter are present (Moens et al. 2002). Few studies about meiofauna in seagrass beds take microphytobenthos into account (Hyndes and Lavery 2005; Leduc et al. 2009) so the role of this food resource remains poorly understood in this habitat.

Another food source exploited by meiofauna is *Zostera noltii* detrital matter. The use of detrital matter is generally mediated by bacteria which colonize it (Findlay and Tenore 1982; Holmer et al. 2004). Bacteria have $\delta^{13}C$ similar to their substrate (Boschker et al. 2000) but higher $\delta^{15}N$ values (Dijkstra et al, 2008). The role of bacterial mediation in this transfer between *Z. noltii* detrital matter and meiofauna could not be clearly determined in this study, mainly because it has been carried out at the community level. About copepods, differences between $\delta^{15}N$ values of food sources and these of copepods remains weak in this ecosystem. This suggests that bacterial mediation is low even for *Z. noltii* detrital matter, or that fresh *Z. noltii* could be eaten by copepods which is unlikely. About nematodes, the large range of $\delta^{15}N$ values does not allow determining if the higher $\delta^{15}N$ values of these consumers are related to a bacterial activity or to their different feeding modes. Bacteria role remains unclear since numerous studies on detritus based-food webs have determined that bacteria mediation can be important (Findlay and Tenore, 1982; Anesio et al. 2003, Holmer et al. 2004) while others showed that bacteria mainly use microalgae as a substrate instead of seagrass matter (Boschker et al. 2000). This suggests that the knowledge about bacteria mediation in trophic fluxes needs to be improved.

The use of microphytobenthos and *Z. noltii* detrital matter by meiofauna is probably related with different properties of these resources: quality and availability for microphytobenthos and quantity for *Z. noltii* detrital matter. Microphytobenthos presents high nutritional quality and assimilation rates (Cebrián 1999). Moreover, microphytobenthos has generally constant biomass all year long (Lebreton et al. 2009), particularly in surficial sediment. It is thus constantly available for consumers. Microphytobenthos is also probably more available for copepods than *Zostera noltii* detrital matter, which quantities are low in surficial sediment (Lebreton et al. 2011). High biomass of *Z. noltii* detrital matter is stored in deep sediment layers, that nematodes can reach thanks to their migration ability. The

utilization of this food source is probably more related with quantitative than with qualitative issues because phanerogams are known to have low nutritional quality, particularly detrital matter, in comparison with microphytobenthos (Cebrián 1999).

Nevertheless, the close signatures between microphytobenthos and seagrass detrital matter makes that the role of each of these two resources needs to be clarified. The overlap of signatures between different food sources is a common problem in stable isotope studies. The combination of those results with other trophic markers studies (*i.e.* sulfur stable isotopes, fatty acid analyses, labeled food source experiments) could be an interesting way to dispel some uncertainties (Leduc et al. 2006; Leduc et al. 2009).

CONCLUSION

Three food sources are mostly used by meiofauna in the seagrass bed: SPOM, by copepods, microphytobenthos and *Z. noltii* detrital matter, both by copepods and nematodes. Food sources used by meiofauna appeared related to their accessibility (*i.e.* water column or sediment location of food sources) and availability (*i.e.* seasonality of inputs). In this study, carbon sources used by meiofauna were determined at the community scale. Thus, changes of community stable isotope signatures can be related with a trophic plasticity of dominant species but can also reflect changes of community structure. This shows limits of community scale studies and suggests that studies about carbon sources used by meiofauna should now focus on species or trophic groups.

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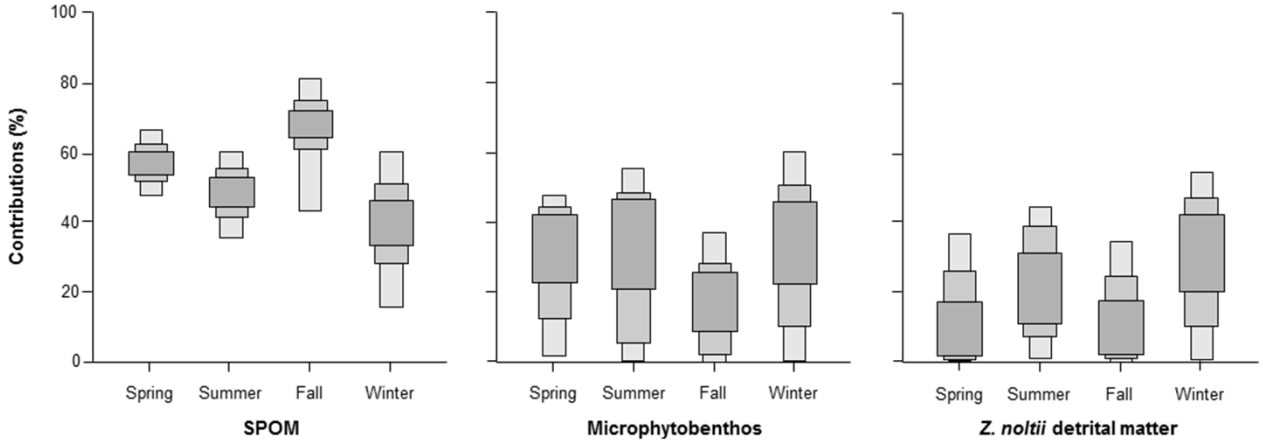
761
762

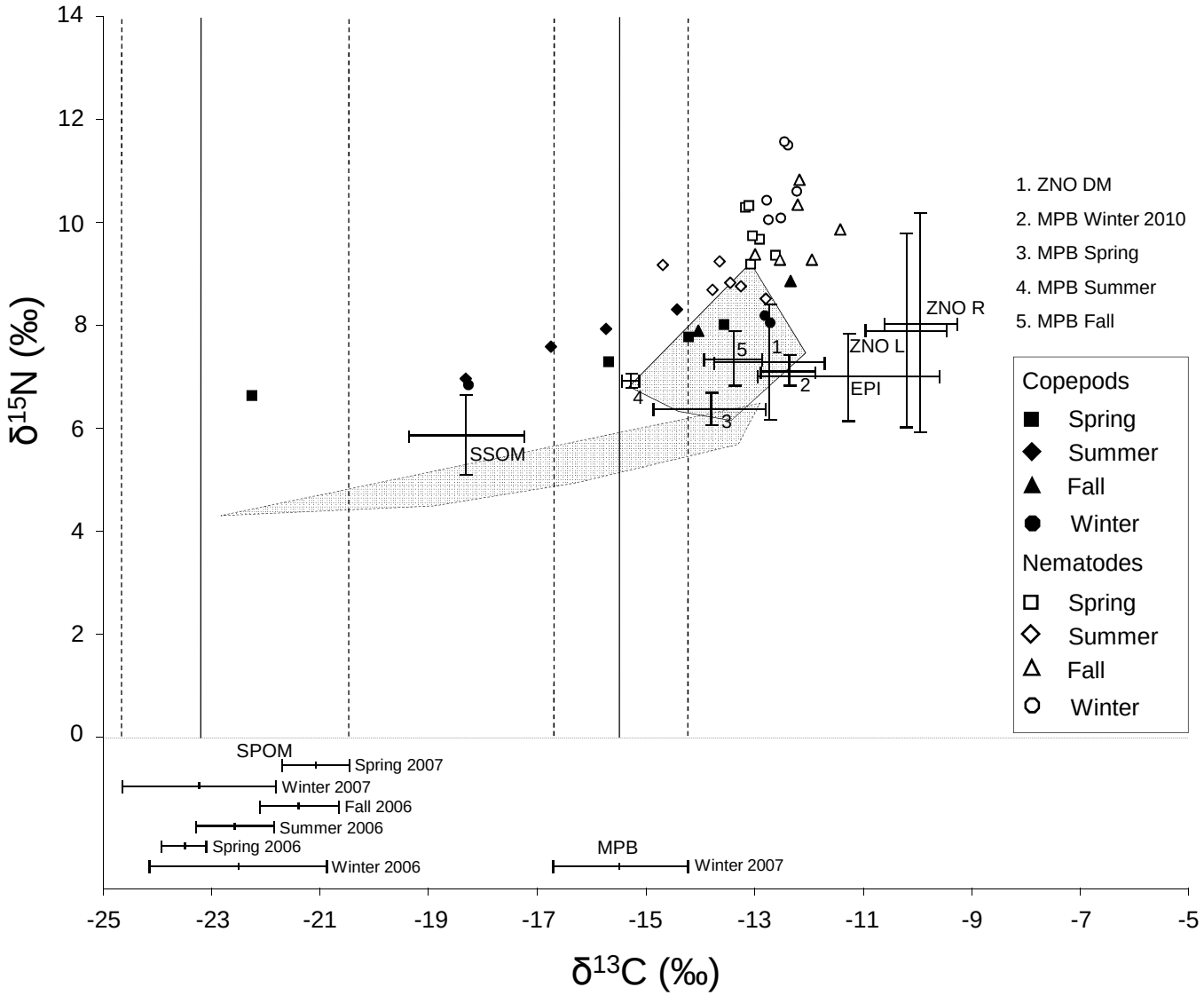
FIGURE CAPTIONS

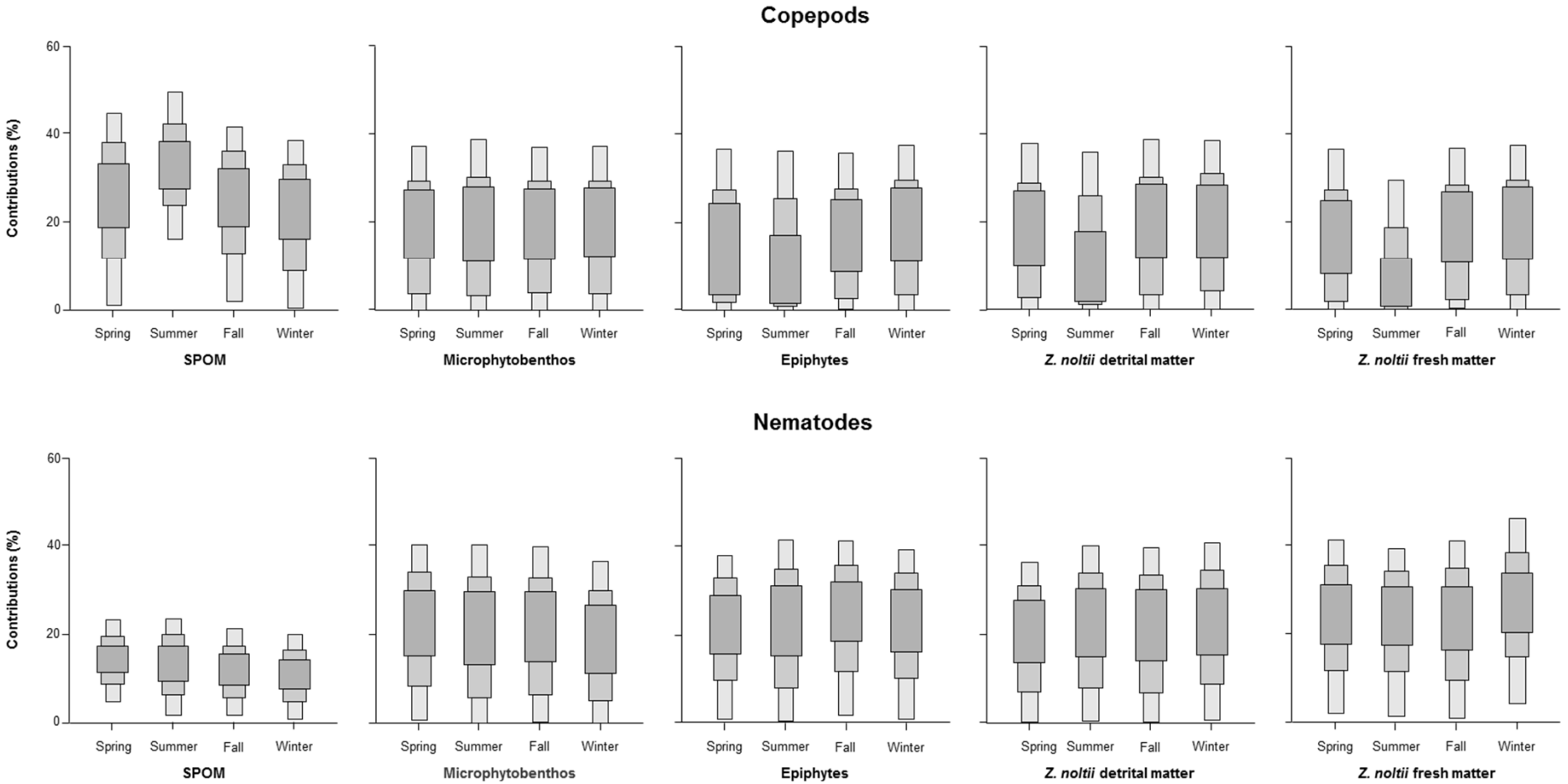
Fig. 1 Seasonal comparison of contributions (%) of SPOM (suspended particulate organic matter), microphytobenthos and *Z. noltii* detrital matter to surface sediment fine organic matter resulting from the mixing model SIAR. 0.95, 0.75, 0.25 credibility intervals are in dark grey, light gray and white, respectively

Fig. 2 Plots of $\delta^{13}C$ and $\delta^{15}N$ values of copepods (‰), nematodes (‰) and of potential food sources (‰, mean ± SD) in the Marennes-Oléron seagrass bed. For copepods and nematodes, each point corresponds to one sample, *i.e.* about 300 individuals. Grey polygons symbolize ranges of nematodes (full line) and copepods (dotted line) theoretical food source signatures taking into account the trophic enrichment (see material and methods). To make comparisons between food source and consumer signatures easier, seasonal data of food sources which show no seasonal variations (*i.e.* SSOM, *Zostera noltii* detrital matter, see table 1) or which are outside of the grey polygons (*i.e.* epiphytes, *Z. noltii* roots and leaves) are aggregated. No $\delta^{15}N$ values were available for SPOM and MPB. For SPOM, full line represents annual mean of $\delta^{13}C$ values and dotted lines represent lowest and highest limits of SD. For MPB sampled in winter 2007, full line represents mean and dotted lines represent SD. Food sources: ZNO R: *Z. noltii* roots; ZNO DM: *Z. noltii* detrital matter; MPB: Microphytobenthos; SSOM: Surface sediment organic matter; SPOM: Suspended particulate organic matter

Fig. 3 Seasonal comparison of contributions (%) of the potential food sources (SPOM: suspended particulate organic matter, microphytobenthos, epiphytes, *Z. noltii* detrital matter and fresh matter) as carbon sources for nematodes and copepods resulting from the mixing model SIAR. 0.95, 0.75, 0.25 credibility intervals are in dark grey, light gray and white, respectively


Contributions (%)
100
80
60
40
20
0
Spring
Summer
Fall
Winter
SPOM
Spring
Summer
Fall
Winter
Microphytobenthos
Spring
Summer
Fall
Winter
Z. noltii detrital matter


14
12
10
8
6
4
2
0
δ15N (‰)
-25
-23
-21
-19
-17
-15
-13
-11
-9
-7
-5
δ13C (‰)
1. ZNO DM
2. MPB Winter 2010
3. MPB Spring
4. MPB Summer
5. MPB Fall
Copepods
Spring
Summer
Fall
Winter
Nematodes
Spring
Summer
Fall
Winter
ZNO R
ZNO L
EPI
SSOM
SPOM
Spring 2007
Winter 2007
Fall 2006
Summer 2006
Spring 2006
Winter 2006
MPB
Winter 2007


Copepods
Contributions (%)
60
40
20
0
Spring
Summer
Fall
Winter
SPOM
Microphytobenthos
Epiphytes
Z. noltii detrital matter
Z. noltii fresh matter
Nematodes
Contributions (%)
60
40
20
0
Spring
Summer
Fall
Winter
SPOM
Microphytobenthos
Epiphytes
Z. noltii detrital matter
Z. noltii fresh matter

	Winter	Spring	Summer	Fall	P values	Comparisons of means
$\delta^{13}C$						
Z. noltii leaves	2006: -9.9 ± 0.1, N = 3 2007: -9.7 ± 0.5, N = 3	2006: -10.7 ± 0.2, N = 3 2007: -11.2 ± 0.2, N = 3	2006: -9.4 ± 0.2, N = 6 2007: -11.1 ± 0.2, N = 3	2006: -10.4 ± 0.6, N = 3	0.003	Summer 2006 > Spring 2007 Summer 2006 > Summer 2007
Z. noltii roots	2006: -10.2 ± 0.1, N = 3 2007: -9.8 ± 0.1, N = 3	2006: -9.3 ± 0.2, N = 3 2007: -11.1 ± 0.4, N = 3	2006: -9.3 ± 0.4, N = 6 2007: -10.1 ± 0.1, N = 3	2006: -10.4 ± 0.5, N = 3	0.005	Summer 2006 > Spring 2007
Z. noltii detrital matter	2006: -12.8 ± 1.3, N = 3	2006: -12.3 ± 1.7, N = 3	2006: -13.2 ± 0.8, N = 3 2007: -12.5 ± 0.7, N = 3	2006: -12.8 ± 0.8, N = 3	0.894	=
Epiphytes		2007: -10.4 ± 1.5, N = 5	2007: -12.8 ± <0.1, N = 3		0.025	Spring 07 > Summer 07
Microphytobenthos	2007: -15.5 ± 1.2, N = 6 2010: -12.4 ± 0.5, N = 4	2007: -13.8 ± 1.0, N = 5	2007: -15.3 ± 0.2, N = 4	2007: -13.4 ± 0.5, N = 4	0.002	Winter 2007 < Winter 2010 Summer 2007 < Winter 2010
SSOM	2007: -17.4 ± 1.9, N = 4	2006: -18.2 ± 0.3, N = 3 2007: -19.1 ± 0.1, N = 3	2006: -18.1 ± 1.0, N = 3 2007: -18.6 ± 0.1, N = 2	2006: -18.9 ± 0.1, N = 3	0.067	=
SPOM	2006: -22.5 ± 1.6, N = 14 2007: -23.2 ± 1.5, N = 5	2006: -23.5 ± 0.4, N = 12 2007: -21.1 ± 0.6, N = 7	2006: -22.6 ± 0.7, N = 18	2006: -21.4 ± 0.7, N = 11	> 0.001	Spring 2006 < Fall 2006 Spring 2006 < Spring 2007
$\delta^{15}N$						
Z. noltii leaves	2006: 4.9 ± 0.3, N = 3 2007: 6.5 ± 0.5, N = 3	2006: 8.2 ± 0.5, N = 3 2007: 7.0 ± 0.4, N = 3	2006: 8.7 ± 2.0, N = 3 2007: 9.4 ± 0.4, N = 3	2006: 9.9 ± 0.8, N = 3	0.011	Winter 06 < Fall 06
Z. noltii roots	2006: 6.4 ± 0.2, N = 3 2007: 7.6 ± 0.3, N = 3	2006: 7.4 ± 1.5, N = 3 2007: 6.5 ± 0.7, N = 3	2006: 6.9 ± 0.4, N = 3 2007: 10.5 ± 1.8, N = 3	2006: 11.0 ± 2.4, N = 3	0.017	=
Z. noltii detrital matter	2006: 6.9 ± 0.1, N = 3	2006: 6.9 ± 1.1, N = 3	2006: 7.1 ± 0.3, N = 3 2007: 8.5 ± 0.7, N = 3	2006: 7.0 ± 2.1, N = 3	0.213	=
Epiphytes		2007: 7.8, N = 1	2007: 6.2 ± 0.2, N = 3			
Microphytobenthos	2010: 7.1 ± 0.3, N = 4	2007: 6.4 ± 0.3, N = 5	2007: 6.9 ± 0.1, N = 4	2007: 7.4 ± 0.5, N = 4	0.016	Spring 2007 < Fall 2007
SSOM	2007: 5.6 ± 0.4, N = 3	2006: 5.0 ± 0.1, N = 3 2007: 6.0 ± 0.1, N = 3	2006: 6.8 ± 1.3, N = 3 2007: 5.9 ± 0.2, N = 2	2006: 6.0 ± 0.2, N = 3	0.038	=

Table 1 Stable isotope ratios (‰, mean ± SD, number of samples) of primary producers and composite food sources per season and summary of Kruskal-Wallis tests between sampling seasons. Values from winter and summer 2006 and 2007 are issued from Lebreton et al. (2011)

	Spring 2007	Summer 2007	Fall 2007	Winter 2008	P values	Comparisons of means
$\delta^{13}C$						
Nematodes	-13.0 ± 0.2, N = 6	-13.6 ± 0.6, N = 6	-12.2 ± 0.5, N = 6	-12.5 ± 0.2, N = 6	0.001	Summer < Fall Summer < Winter
Copepods	-16.4 ± 4.0, N = 4	-16.4 ± 1.4, N = 5	-14.4 ± 2.3, N = 3	-14.6 ± 3.2, N = 3	0.490	=
$\delta^{15}N$						
Nematodes	9.8 ± 0.5, N = 6	8.9 ± 0.3, N = 6	9.8 ± 0.7, N = 6	10.7 ± 0.7, N = 6	0.001	Summer < Winter
Copepods	7.4 ± 0.6, N = 4	7.7 ± 0.6, N = 4	8.4 ± 0.7, N = 2	7.7 ± 0.7, N = 3	0.507	=

Table 2 Stable isotope ratios (‰, mean ± SD, number of samples) of nematodes and copepods and summary of Kruskal-Wallis tests at the different seasons from spring 2007 to winter 2008

	$\delta^{13}C$	$\delta^{15}N$
Spring 2007	0.400	0.200
Summer 2007	1.000	0.533
Fall 2007	0.400	0.100
Winter 2008	0.400	0.700

Table 3 Summary of P values of Mann-Whitney-Wilcoxon tests between stable isotope signatures of nematodes ($\delta^{13}C$, $\delta^{15}N$) from top and bottom communities