Marine Biology
January 2015, Volume 162 Issue 1 Pages 15-37
http://dx.doi.org/10.1007/s00227-014-2577-5
http://archimer.ifremer.fr/doc/00247/35859/

Small pelagic fish feeding patterns in relation to food resource variability: an isotopic investigation for *Sardina pilchardus* and *Engraulis encrasicolus* from the Bay of Biscay (north-east Atlantic)

Chouvelon Tiphaine [1, 2, *], Violamer L. [1], Dessier Aurelie [1], Bustamante Paco [1], Mornet Francoise [3], Pignon-Mussaud Cecilia [1], Dupuy Christine [1]

[1] Université de La Rochelle, Littoral Environnement et Sociétés (LIENSs), UMR 7266, CNRS, 2 rue Olympe de Gouges, 17000, La Rochelle 01, France
[2] IFREMER, Unité Biogéochimie et Écotoxicologie (BE), Laboratoire de Biogéochimie des Contaminants Métalliques (LBCM), Rue de l'Ile d'Yeu, 44311, Nantes 03, France
[3] IFREMER, Unité Halieutique Gascogne Sud (HGS), Station de La Rochelle, Place Gaby Coll, 17087, L'Houmeau, France

* Corresponding author : Tiphaine Chouvelon, email address : tiphaine.chouvelon@hotmail.fr

Abstract :

Small pelagic fish represent an essential link between lower and upper trophic levels in marine pelagic ecosystems and often support important fisheries. In the Bay of Biscay in the north-east Atlantic, no obvious controlling factors have yet been described that explain observed fluctuations in European sardine Sardina pilchardus and European anchovy Engraulis encrasicolus stocks, in contrast to other systems. The aim of this study was therefore to investigate to which extent these fluctuations could be trophodynamically mediated. The trophic ecology of both fish species was characterised over three contrasting periods (spring 2010 and 2011 and autumn 2011) in the area, in relation to potential variation in the abundance and composition of the mesozooplankton resource. Stable isotope analyses of carbon (δ13C) and nitrogen (δ15N) were performed on potential mesozoplanktonic prey items and in the muscle of adult fish, as well as in the liver whenever available, and mixing models were applied. In both springs, the mesozooplankton resource was abundant but qualitatively different. During this period of the year, results based on muscle isotope values in particular showed that S. pilchardus and E. encrasicolus likely do not compete strongly for food. On the medium term, E. encrasicolus always presented a greater trophic plasticity than S. pilchardus, both in terms of feeding areas and in the size of the mesozooplanktonic prey consumed. In autumn, mesozooplankton abundances were lower, and it was likely that S. pilchardus and E. encrasicolus share food resources during this period. No clear links between the variation in the mesozooplanktonic resource and the trophic segregation maintained between adults of both fish species in spring could be made. Although a certain potential exists for trophodynamically mediated fluctuations of both species under specific abiotic conditions (i.e. due to the existing trophic segregation in spring in particular), the overall results suggest that fluctuations in abundance of both fish species are probably not directly linked to their trophic ecology in the Bay of Biscay, at least at the level of adult individuals.

Introduction

Forage fish such as sardines and anchovies have a key role in marine pelagic ecosystems, representing the main pathway by which energy and nutrients are transported from lower (i.e., plankton) to upper trophic levels (i.e., marine mammals, large fish and seabirds) (Cury et al. 2000). However, the stocks of these small pelagic fish can be highly variable over time (e.g., Schwartzlose et al. 1999). These fluctuations can lead to considerable changes in the structure and function of marine ecosystems, and in turn impact fisheries (FAO 2012). Understanding the processes involved in the fluctuations of forage fish abundance therefore appears critical to maintain marine ecosystem services.

For many years, in several marine ecosystems and notably those subjected to upwelling events where sardines and anchovies cohabit (e.g., Benguela Current Ecosystem on the South African coast or Humboldt Current Ecosystem on the Peruvian coast), alternative abundance fluctuations in the populations of both species have been reported (e.g., Lluch-Belda et al. 1989; Barange et al. 2009). Several hypotheses have been proposed to explain these sardine-anchovy fluctuations. Some of these hypotheses rely on the effects of physical, atmospheric and oceanographic regime such as climatic oscillations that potentially control the survival and/or recruitment of one of the other species (e.g., Lluch-Belda et al. 1992; Chavez et al. 2003; Alheit et al. 2012). Takasuka et al. (2007) also proposed that both species display differential “optimal growth temperatures”, so that different climatic conditions can favour one species or the other during early life stages. This hypothesis extends the “optimal environmental window” theory of Cury and Roy (1989), establishing the conditions for the recruitment success of pelagic fish in upwelling areas. Other hypotheses proposed for explaining sardine-anchovy alternations include biological controlling factors such as intra-guild predation (e.g., Irigoien and De Roos 2011), or trophodynamically mediated fluctuations with the resource’s variability favouring one species or the other (e.g., Van der Lingen et al. 2006). Some studies that have investigated the diet of both species simultaneously (e.g., Louw et al. 1998; Van der Lingen et al. 2006; Espinoza et al. 2009) have effectively demonstrated that sardines and anchovies (generally adult individuals) show distinct feeding strategies, especially in terms of the size of copepod they preferentially consume. Hence, warmer or cooler oceanographic regimes would favour the

development of small or larger planktonic prey species, and thus one or other small pelagic predator. Simply determining the effects of abiotic factors influencing both the recruitment and survival of early life stages is thus not sufficient to understand fluctuations in the abundance of small pelagic fish. The knowledge of trophic interactions between species as well as fluctuations in food resource and their impact on trophic interactions also appears a crucial step.

Stable isotope analysis (SIA) of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) of the tissues of consumers and their putative prey has proven to be a powerful tool to describe the trophic ecology of marine organisms, representing an alternative or complementary tool to the traditional methods of dietary studies such as the analysis of stomach contents (Michener and Kaufman 2007). Primary producers of an ecosystem generally display different isotopic compositions (Peterson and Fry 1987; France 1995) and the enrichment in ^{13}C and ^{15}N between a source and its consumer (also called Trophic Enrichment Factor, TEF) is relatively predictable. This enrichment is less important in ^{13}C ($\leq$ 1‰) than in ^{15}N (3.4‰ on average) (De Niro and Epstein 1978, 1981; Post 2002). Hence, $\delta^{13}C$ values are generally considered as a conservative tracer of the primary producer at the base of the food web supporting consumers, and consequently a tracer of their foraging habitat (France 1995; Hobson 1999). Alternatively, $\delta^{15}N$ values are generally used as a proxy of their trophic position (Vander Zanden et al. 1997; Post 2002). Furthermore, for some years, mixing models integrating $\delta^{13}C$ and $\delta^{15}N$ values of prey and predators have proved their utility to decipher the contribution of different prey items in the diet of a predator (Parnell et al. 2010, 2013; Phillips et al. 2014). This may be particularly useful when studying the trophic links between plankton and small pelagic planktivorous fish (e.g., Costalago et al. 2012), because of the peculiar difficulty in observing direct interactions between these organisms in the open water environment, and because the small size of plankton can make stomach content analysis particularly difficult. Moreover, isotope values provide information on the food assimilated at a time scale that depends on the turnover of the tissue analysed (Tieszen et al. 1983; Hobson and Clark 1992; Sponheimer et al. 2006). For instance, carbon and nitrogen half-lives in fish tissues were shown to vary from 5-14 days in the liver to 19-21 days in the muscle of the juvenile Japanese bass *Lateolabrax japonicus* (Suzuki et al. 2005), from 3-9 days in the liver to 25-28 days in the muscle of

the juvenile sand goby *Pomatoschistus minutus* (Guelinckx et al. 2007), and from 10-20 days in the liver to 49-107 days in the muscle of the flat fish *Paralichthys dentatus* (Buchheister and Latour 2010).

The Bay of Biscay is a very large bay located in the north-east Atlantic Ocean. It supports a rich fauna including many protected species, e.g., marine mammals, seabirds, sharks and rays, and is subjected to numerous anthropogenic activities including important fisheries (Lorance et al. 2009; OSPAR 2010). In particular, European sardine (*Sardina pilchardus*) and European anchovy (*Engraulis encrasicolus*) fisheries are of major importance in the area (ICES 2010a). No quota currently exists for sardine despite an observed decrease in their catches in this area (OSPAR 2010). Conversely, a decrease in anchovy stocks during the 2000s led to the closing of its fishery in 2005. The moratorium ended in 2010 and finally resulted in the establishment of quotas for this species (ICES 2010a, b). In the Bay of Biscay, strong fluctuations in the abundance of small pelagic fish such as sardines and anchovies have been observed for several years (ICES 2010a). However, in contrast to upwelling areas where alternative abundance fluctuations have been demonstrated and/or linked to climatic events or biological controlling factors (see above), no clear relationships between both fish species have yet been shown in the Bay of Biscay ecosystem. Sardine and anchovy have always demonstrated both alternation and co-occurrence in spring-survey data (ICES 2010b) and no obvious controlling factors have been identified to-date explaining general fluctuations in the abundances of small pelagic fish in the area. Besides, an ecological network analysis of the Bay of Biscay continental food web provided evidence that bottom-up processes play a significant role in the population dynamics of upper-trophic levels and in the global structuring of this marine ecosystem (Lassalle et al. 2011).

In a previous study in the area, Chouvelon et al (2014) examined the trophic ecology of adults of the two fish species by SIA during a single specific period (spring 2010). The authors highlighted a trophic segregation between species during the study period. This may support the hypothesis that fluctuations of both fish species' abundances could be, at least in part, trophodynamically mediated, if the food environment on the medium to long-term would tend to favour one species or the other, as a

function of their respective dietary preferences (Van der Lingen et al. 2006). However, no link could be made with food resource composition and availability in this previous study (Chouvelon et al. 2014), because only one period of sampling and a single tissue (muscle tissue, i.e., medium to long-term integrator of the food assimilated) were considered. Demonstration of such a link could highlight a strong dependency of one or both fish species to resource composition and availability, and/or reveal a relative trophic plasticity in one or both species relative to food resource variability. This may finally help to understand to which extent fluctuations and/or alternations of both species may be strongly trophodynamically mediated or not in the area.

In this general context, the aim of this study was twofold: 1) investigating intra- (seasonal) and inter-annual variations in the trophic ecology of adult sardines and anchovies from the Bay of Biscay; 2) linking potential temporal variation in the diet of both fish species with variations in the mesozooplankton resource, to depict potential differential feeding strategies in both fish species in relation to resource variability. Several studies have highlighted that zooplankton (and notably copepods belonging to the mesozooplankton community) are by far the most important dietary component for sardines and anchovies compared to phytoplankton (e.g., Van der Lingen et al. 2006; Espinoza et al. 2009; Nikolioudakis et al. 2012;). As such, we focused on mesozooplanktonic prey as the major food resource for both fish species in the present study. Three different periods of sampling with contrasting abiotic conditions were considered, with one of these periods referring to those investigated by Chouvelon et al. (2014). SIA was undertaken on identified mesozooplanktonic prey and predators and mixing models used to estimate consumption patterns. The results obtained provide some understanding as to what extent potential trophodynamic differences and/or dependence on food resource variability (composition and availability) can influence fluctuations and/or alternations of both fish species abundances in the highly productive Bay of Biscay area.

Materials and Methods

155 Sample collection

156

157 Mesozooplankton and fish samples were collected in the spring of 2010 and 2011 and autumn of 2011,
158 during sea surveys conducted by the French Research Institute for the Exploitation of the Sea
159 (IFREMER) on the continental shelf to the shelf-edge of the Bay of Biscay: PELGAS 2010 and
160 PELGAS 2011 surveys (25th April – 5th June 2010 and 26th April – 4th June 2011, respectively), and
161 EVHOE 2011 survey (18th October – 30th November 2011). As noted above, isotope values of samples
162 from the PELGAS 2010 survey were presented in a previous study in the area (Chouvelon et al.,
163 2014), as well as the methodological aspects related to the study of trophic relationships between
164 mesozooplankton and planktivorous fish through SIA. Isotope results of the spring 2010 survey are
165 thus only used here for direct comparison with the two other periods examined (i.e., spring and
166 autumn 2011), and further links with the variation in resource abundance between the three periods.

167 These seasons were selected for sampling for various reasons regarding the objectives of the
168 study. First, it was hypothesized that food resource abundance and composition would greatly differ
169 between spring and autumn, i.e., between seasons presenting different environmental conditions in
170 temperate areas such as the Bay of Biscay (Villate et al. 1997; Valdés and Moral 1998, Zarauz et al.
171 2007). Moreover, survey data indicated that both springtime periods were different in terms of
172 temperature and salinity patterns in particular, potentially leading to different food resource
173 availability as well (i.e., warmer sea surface temperatures during the spring 2011 campaign, in
174 comparison with the spring 2010 campaign; IFREMER survey data, see also
175 www.previmer.org/observations). Finally, sampling seasons were chosen with regard to the main
176 spawning period of both fish species, potentially driving different feeding strategies in the study
177 fishes. Indeed, for the Bay of Biscay anchovy, the peak spawning period has been reported to be in
178 spring (i.e., May-June; Motos 1996), and the onset of spawning is concurrent with the sharp seasonal
179 increase in surface temperature (ICES 2010b). Even though feeding migrations would occur after
180 spawning (i.e., in summer and autumn), with fat content increasing during these seasons (Dubreuil and
181 Petitgas 2009), anchovy continue to feed during the spawning season (Plounevez and Champalbert

1999), with the duration of the spawning season depending on energy intake during this period (ICES, 2010b). For the Atlanto-Iberian and Biscay sardine, the main spawning period is between October and June and thus partly overlaps with those of anchovy in the Bay of Biscay (ICES 2010b). As for anchovy, fat content peaks in early autumn (i.e., beginning of the spawning season), although sardines also feed throughout the year (ICES 2010b).

Mesozooplankton were collected by vertical trawls of 200µm mesh-size WP2 nets, from 100m depth (or bottom depth for inshore stations) to the surface. 10 to 16 stations were selected depending on the survey (Fig. 1). During PELGAS (spring) surveys, the stations followed transects used for the hydroacoustic assessment of small pelagic fish biomass. They were thus distributed from the north to the south of the Bay of Biscay, and from the coastline (C) to the continental slope (Sl) including stations over the continental shelf (Sh) (Fig. 1A). During PELGAS 2011, one oceanic (O) station was also considered (Fig. 1B). During EVHOE 2011 survey, stations followed randomly distributed fishing trawls, although as in PELGAS the stations were selected in order to cover all the Bay of Biscay area (Fig. 1C). After collection, mesozooplankton samples were concentrated on a 200µm mesh and preserved in 70% ethanol for further taxonomic identification and stable isotope analysis.

Adult sardines and anchovies were collected by pelagic trawls during PELGAS surveys (76*70 trawl with vertical opening of ~ 25 m, or 57*52 trawl with vertical opening 15-20 m), and by bottom trawls during EVHOE survey (large vertical opening (GOV) trawl 36/47). This is due to a difference in the main initial objectives of the surveys (i.e., assessing abundance and distribution of pelagic fish in the Bay of Biscay using acoustic method during PELGAS; assessing abundance and distribution of demersal and benthic resources using bottom trawl during EVHOE). During each survey and for each fish species, individuals were collected in 7 to 8 trawls over the continental shelf (Fig. 1). In some trawls, both species occurred at the same time however this does not indicate that they come from the same shoal given the duration of each trawl (between 30 to 60 minutes). Fish were immediately frozen at -20°C until further dissection and analyses back to the laboratory.

208 Taxonomic determination of mesozooplankton and preparation for analysis

209

210 Taxonomic identification of mesozooplankton was carried out at the laboratory with a Leica M3Z
211 stereo microscope (mag. x65 to x160), to genus and to species level whenever possible. For each
212 spring station, identified taxa contributing at least 5% of the total abundance of the sample both in
213 number (individuals. m^{-3}) and in biomass (mg. m^{-3}) (i.e., 'dominant taxa'), and likely to be part of the
214 diet of sardines and anchovies (i.e., species that may be found in stomach contents of anchovies from
215 the Bay of Biscay area as reported by Plounevez and Champalbert (1999)) were sorted for further SIA.
216 For each autumn station, as the diversity was lower, only identified taxa contributing at least to 10% of
217 the total abundance of the sample both in number and in biomass were subsequently sorted for SIA.
218 As such, one to four 'dominant taxa' were analysed for stable isotope ratios within each of the stations
219 sampled over the three periods. Details for the calculation of the relative abundance of each identified
220 taxa in number and in biomass can be found in Chouvelon et al. (2014).

221 Depending on their size, 20 to 350 individuals belonging to each of the 'dominant taxa' were
222 taken out from ethanol and carefully washed with distilled water in order to completely remove the
223 ethanol, detritus and phytoplankton. Sorted and washed organisms were finally frozen at -80°C for 48h
224 to be freeze-dried (24h). A pool of individuals for each species sorted by station was then packed into
225 2 tin capsules for stable isotope analysis (i.e., half of sorted organisms within each capsule) and the
226 mean value of the two capsules was used in further data analyses (Chouvelon et al. 2014).

227 For each fish species and for each survey, 30 to 40 adult individuals of similar size classes
228 (average total length ± standard deviation (SD) of 18.1 ± 2.2 cm and 13.9 ± 1.6 cm for sardines and
229 anchovies, respectively) were defrosted and dissected at the laboratory to obtain portions of dorsal
230 white muscle as well as the liver (Pinnegar and Polunin 1999). Specifically, the average total length
231 for sardines was of 17.3 ± 2.6 cm, 18.7 ± 0.7 cm and 18.4 ± 2.6 cm for individuals collected in spring
232 2010, spring 2011 and autumn 2011, respectively. The average total length for anchovies was of 14.6
233 ± 1.8 cm, 13.3 ± 1.1 cm and 13.7 ± 1.5 cm for individuals collected in spring 2010, spring 2011 and

autumn 2011, respectively. Within each species and at each season, these sizes corresponded to mature individuals and allowed comparison of morphologically similar fishes (i.e., adult individuals) at the three seasons investigated. Also, sardines were larger than anchovies, because the size at maturity is higher for sardines (i.e., about 14 cm length, 1 to 2 years old individuals) than for anchovies (i.e., about 10 cm length, one year old) (ICES 2010b). Muscle and liver samples were individually stored frozen at -20°C in plastic bags prior to a 72h freeze-drying period. White muscle and liver samples were ground manually or with a planetary ball mill (Retsch PM 200), and were treated with cyclohexane in order to remove naturally ^{13}C-depleted lipids (De Niro and Epstein 1977). Lipid-free samples were finally dried in an oven at 45°C for 48h and packed in tin capsules for SIA.

Stable isotope analysis

The natural abundance of carbon and nitrogen stable isotopes in plankton and fish was determined with a Thermo Scientific Delta V Advantage mass spectrometer coupled to a Thermo Scientific Flash EA1112 elemental analyser. Results are expressed as isotope ratios δX (‰) relative to international standards (Pee Dee Belemnite for carbon and atmospheric N2 for nitrogen), according to the formula:

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

where X = ^{13}C or ^{15}N and R = $^{13}C/^{12}C$ or $^{15}N/^{14}N$ (Peterson and Fry 1987). Replicate measurements of internal laboratory standards (acetanilide) indicated a precision of 0.2‰ for both $\delta^{13}C$ and $\delta^{15}N$ values.

Data treatment and statistical analyses

Chouvelon et al. (2014) demonstrated a significant effect of preservation (ethanol 70% *vs*. freezing at -20°C) and of lipid content on mesozooplankton $\delta^{13}C$ and $\delta^{15}N$ values. In the present study, for

consistency of the treatment applied to prey and predators both in terms of preservation and of lipid correction, we thus applied the same corrections as proposed by Chouvelon et al. (2014) for further analysis of the diet of sardine and anchovy through SIA. Briefly, this consisted in correcting $\delta^{13}C$ and $\delta^{15}N$ values of all mesozooplanktonic organisms preserved in 70% ethanol for the effect of ethanol, and only $\delta^{13}C$ values of mesozooplanktonic organisms were corrected for the effect of lipid content (Chouvelon et al. 2014). The corrected values were then used in further statistical analyses and mixing models.

All statistical analyses were conducted with R (R Development Team 2011). Normality of all data was tested using Shapiro-Wilk's tests, i.e., for further use of parametric or non-parametric statistics. A Student t test or a Mann-Whitney-Wilcoxon test was thus applied when comparing two series of samples, e.g., for testing significant difference between both species. Similarly, an ANOVA (followed by post-hoc Tukey tests) or a Kruskal-Wallis test (followed by a multiple comparison test with Holm's adjustment method) was applied when comparing more than two series of samples, e.g., for testing significant difference between periods.

In order to link potential variations in the trophic ecology of both fish species inferred from SIA with the variability of the mesozooplankton resource, data on mesozooplankton abundances presented in the present study mainly concerns the taxa contributing to more than 5% of the total abundance both in number and in biomass, in at least one station for one of the periods considered ('dominant taxa'). These taxa were effectively those analysed for SIA and considered in mixing models (see following section). The representativeness of these 'dominant taxa' relative to the whole mesozooplankton community was previously checked by analysing the correlation between total mesozooplankton abundance and total abundance of these 'dominant taxa' through a Spearman correlation coefficient test.

Isotopic mixing models

To account for numerous potential prey items in the diets of sardines and anchovies, the wide variability in the $\delta^{13}C$ and $\delta^{15}N$ values of mesozooplancton, and for the uncertainty of TEFs (i.e., the difference (Δ) in $\delta^{13}C$ and $\delta^{15}N$ between the predator's tissue analysed and its diet), Bayesian isotopic mixing models were used (available as an open source R package SIAR; Parnell et al. 2010). In mixing models that are mathematically underdetermined (with more unknowns than equations and no unique solution) where the number of sources exceeds n+1 (Phillips and Gregg 2003), one possible approach to encompass this common problem and to simplify the analysis is to combine some sources (Phillips et al. 2005). In the present study, potential prey items, that is all entities 'taxa-station' (e.g., '*Temora* sp.-C2', 'Medium undetermined Calanoid-Sh3') analysed for isotopes were thus grouped before running SIAR. As in Chouvelon et al. (2014), this grouping was performed through a Hierarchical Cluster Analysis (HCA) for each period considered. HCA were based on $\delta^{13}C$ and $\delta^{15}N$ values, average size (total length) of organisms (see Table 1), and geographical coordinates of each entity 'taxa-station' analysed for isotope ratios. The groups defined by HCA were then used in mixing modelling (Table 2).

To the best of our knowledge, precise TEFs are still unknown for mesozooplankton-feeders such as sardines and anchovies. Post (2002) suggested that TEFs of 0.4 ± 1.3‰ and 3.4 ± 1‰ for $\delta^{13}C$ and $\delta^{15}N$, respectively, could be widely applicable within a food web. Nevertheless, there is increasing evidence in the literature that TEFs may be highly variable as a function of the consumer's taxa, or as a function of the type and the quality of the consumer's food (e.g., Vanderklift and Ponsard 2003; Caut et al. 2009). Recent studies have also shown that even considering uncertainty around TEFs or discrimination factors, Bayesian models outputs may be very sensitive to the chosen TEFs (e.g., Bond and Diamond 2011). To apply sensitivity analyses on the results obtained, four mixing models by species and by tissue were thus run using different values of TEFs found in the literature, for both $\delta^{13}C$ and $\delta^{15}N$ (Post 2002, for general values; Pinnegar and Polunin 1999; Trueman et al. 2005; and Sweeting et al. 2007a, b for fish muscle or liver in particular; see Table 4 for the detailed TEFs used). The variability around $\delta^{13}C$ and $\delta^{15}N$ values of each source taken into account in the mixing models corresponded to the standard deviation around the mean of each source group (i.e., SD given in Table

2). For each period considered, for each tissue and for each species, an average value of the estimated contribution of each group of mesozooplankonic prey was finally calculated from the four mixing models applied (Table 4).

Results

General abundance and distribution patterns in the mesozooplankton community

Over the three study periods and considering all the stations selected for taxonomical identification, total abundance of mesozooplankton (in number) was the highest in spring 2010 and varied between 541 to 7417 ind. m^{-3} (mean ± SD: 3316 ± 2609 ind. m^{-3}, CV = 79% for the 13 stations covered at this period in the Bay of Biscay area). In spring 2011, total abundances were slightly lower on average but varied among a similar range of values, i.e., from 305 to 8433 ind. m^{-3} (1935 ± 2108 ind. m^{-3}, CV = 109% for the 16 stations covered). Total abundances finally displayed the lowest values in autumn 2011, varying from 53 to 3366 ind. m^{-3} (758 ± 1042 ind. m^{-3}, CV = 138% for the 10 stations covered). Within the whole mesozooplankton community, the percentage of copepods relative to the total abundance of mesozooplanktonic organisms varied from 21 to 99% in spring 2010, and from 49 to 96% in spring 2011. The values were the highest in autumn 2011, varying from 72 to 97% (Fig. 2).

The correlation between the total abundance of mesozooplankton (in number) and the abundance of taxa contributing to more than 5% of the total abundance (i.e., 'dominant taxa') was highly significant ($r_{Spearman}$ = 0.984, $p < 0.0001$, n = 39 stations – i.e., all stations covered during the three periods investigated). This indicated these 'dominant taxa' of the wider mesozooplankton community. In spring of 2010 and 2011, coastal stations were mainly characterised by small to medium-sized organisms such as *Acartia* sp., *Temora* sp. or Appendicularia (Fig. 3A). The large copepod *C. helgolandicus* (especially abundant in spring 2010) or the smaller *Oithona* sp. were more abundant in stations from the shelf and/or from the slope, or in the station O4 from the oceanic area

337 sampled in spring 2011. In autumn 2011, the copepods *Oncaea* sp. and *Temora* sp. were the most
338 abundant in stations located near the coast and/or on the shelf, while large Decapod larvae were
339 abundant in 2 out of the 10 stations analysed. Total abundances in stations located on or near the slope
340 were very low at this period (Fig. 3).

341 Finally, the proportion of small organisms (see Table 1 for size ranges) was relatively stable
342 throughout the three periods considered, varying between 51 to 55% relative to the whole
343 mesozooplankton community (i.e., community now represented by the 'dominant taxa') (Fig. 4A).
344 The proportion of medium-sized organisms was higher in both spring and autumn 2011 (44 and 43%,
345 respectively) than in spring 2010 (34%), whereas the proportion of large organisms such as
346 *C. helgolandicus* was the highest in spring 2010 (16% *vs*. 4% and 2% in spring and autumn 2011,
347 respectively) (Fig. 4A). Abundances of organisms were the highest in stations from the coast and from
348 the shelf both in spring and autumn 2011 (Figs. 3 and 4). However in spring 2011, a non-negligible
349 part of the total abundance of mesozooplankton also belonged to stations from the slope (i.e., 21%), as
350 well as in spring 2010 where high abundances of organisms were found in the more oceanic stations
351 (i.e., 32%) (Figs. 3 and 4).

352

353 Definition of prey groups and variability of mesozooplankton $\delta^{13}C$ and $\delta^{15}N$ values

354

355 The HCA defined eight, nine and six groups of prey for spring 2010, spring 2011 and autumn 2011,
356 respectively (Table 2). As such, the groups reflected a certain ecological significance for further
357 interpretation of the results of isotopic models, both in terms of sizes of organisms and in terms of
358 their sampling location. Isotope values of the different groups were relatively distinct from each other
359 (Table 2). Average $\delta^{15}N$ values varied from 4.2 ± 0.6 (group 1) to 7.2 ± 0.9‰ (group 8) in spring
360 2010, from 4.6 ± 0.3 (group 1) to 8.9 ± 0.6‰ (group 6) in spring 2011, and from 2.4 ± 0.2 (group 1) to
361 7.0 ± 0.4‰ (group 4) in autumn 2011. Average $\delta^{13}C$ values varied from -22.2 ± 0.1 (group 6) to -19.4
362 ± 0.3‰ (group 2) in spring 2010, from -21.8 ± 0.4 (group 1) to -19.0 ± 0.6‰ (group 7) in spring 2011,

and from -20.9 ± 0.0 (group 4) to -20.2 ± 0.2‰ (group 5) in autumn 2011 (Table 2). Groups with large bodied organisms generally displayed higher $\delta^{15}N$ values than those containing small to medium-sized organisms within a same area. Also, within a same range of sizes, organisms collected in coastal waters generally displayed higher $\delta^{15}N$ values than those collected in more oceanic waters (Table 2). For instance, in spring 2010, large organisms from the shelf to the slope in the northern part (group 5) showed an average $\delta^{15}N$ value of 7.1 ± 0.9‰. On the contrary, the average $\delta^{15}N$ value of small to medium-sized organisms from the slope in the northern part (group 1) was of 4.2 ± 0.6‰, and in the same area, small to medium-sized organisms from the coast to the shelf in the northern part (group 4) displayed an average $\delta^{15}N$ value of 7.0 ± 0.6‰. In spring 2011, the same pattern of differences could be observed between these three types of groups collected in the northern part (corresponding to group 3, groups 1 and 2 considered together, and group 6, respectively). This was also the case of groups from the southern area. Large organisms from the shelf to the slope (group 7) showed an average $\delta^{15}N$ value of 7.6 ± 0.7‰, while those of small to medium-sized organisms from shelf to the slope (group 4) was of 5.6 ± 0.5‰. Small to medium-sized organisms from the coast to the shelf (group 8) displayed an average $\delta^{15}N$ value of 7.2 ± 0.3‰ (Table 2).

Fish muscle $\delta^{13}C$ and $\delta^{15}N$ values and isotopic mixing models

Within each of the three periods considered, *S. pilchardus* and *E. encrasicolus* differed significantly for both muscle $\delta^{13}C$ and $\delta^{15}N$ values (all p-values < 0.05; Table 3). *E. encrasicolus* always had lower $\delta^{13}C$ and $\delta^{15}N$ values on average than *S. pilchardus* (Table 3, Fig. 5). In *S. pilchardus*, individuals sampled in autumn 2011 displayed significantly lower muscle $\delta^{13}C$ values than individuals sampled in both spring 2010 and 2011, while $\delta^{15}N$ values were not significantly different between periods (p-values > 0.05, Table 3). In contrast in *E. encrasicolus*, muscle $\delta^{13}C$ values were not significantly different between individuals collected at the three periods, but individuals collected in autumn 2011

388 showed significantly higher $\delta^{15}N$ values than those sampled in springs 2010 and 2011 (Table 3, Fig.
389 5).

390 In spring 2010, 3 groups out of the 8 previously defined mainly contributed to the diet of
391 *S. pilchardus*, whatever the TEF used: group 8 corresponding to small to medium-sized organisms
392 from the coast to the shelf in the central to southern part (average mean contribution ± SD = 43.7 ±
393 5.9%), group 4 corresponding to small to medium-sized organisms from the coast to the shelf in the
394 northern part (28.9 ± 9.6%), and in lower proportion group 5 corresponding to large organisms from
395 the shelf to the slope in the northern part (14.7 ± 9.5%; Table 4). The same three groups presented the
396 highest estimated contribution in the diet of *E. encrasicolus* as well (22.3 ± 7.7%, 19.3 ± 7.7% and
397 17.6 ± 10.0% for groups 8, 4 and 5, respectively). However in the latter species, two other groups also
398 contributed significantly to its diet (i.e., average contribution close to or ≥ 10%): namely group 6
399 corresponding to large organisms from the slope in the central to southern part (13.1 ± 11.8%), and
400 group 2 containing medium-sized organisms from the coast to the shelf in the central to northern part
401 (11.3 ± 10.8%; Table 4).

402 In spring 2011, 4 groups out of the 9 defined mainly contributed to the diet of *S. pilchardus* and
403 *E. encrasicolus* (i.e., average contribution ≥ 10% in both species): group 6 containing small to
404 medium-sized organisms from the coast in the northern part (40.8 ± 16.0% and 11.2 ± 8.1% in *S.*
405 *pilchardus* and *E. encrasicolus*, respectively), group 3 corresponding to large organisms from the shelf
406 to the slope in the northern part (17.4 ± 13.8% and 29.6 ± 24.8% in *S. pilchardus* and *E. encrasicolus*,
407 respectively), group 8 corresponding to small to medium-sized organisms from the coast to the shelf in
408 the southern part (12.4 ± 13.0% and 11.7 ± 14.7% in *S. pilchardus* and *E. encrasicolus*, respectively),
409 and finally group 7 including large organisms from the shelf to the slope in the southern part (12.1 ±
410 12.1% and 9.5 ± 9.8% in *S. pilchardus* and *E. encrasicolus*, respectively). In total, these four groups
411 (i.e., groups 3, 6, 7 and 8) contributed on average to 82.7% and 62.0% to the diet of *S. pilchardus* and
412 *E. encrasicolus*, respectively (Table 4). However, group 6 presented the highest contribution in *S.*
413 *pilchardus* (40.8 ± 16.0%) whatever the TEF used, the group 3 was the most significant group in the
414 diet of *E. encrasicolus* (29.6 ± 24.8%) in 3 out of the 4 models performed (Table 4).

Mixing models performed on $\delta^{13}C$ and $\delta^{15}N$ values in the muscle of the fish sampled in autumn 2011 highlighted the major contribution of 3 of the 6 groups defined in the diet of both species. In total, group 4 (corresponding to small to medium-sized organisms from the coast to the shelf in the northern part) and group 5 (containing small to medium-sized organisms from the coast to the shelf in the central part) both contributed on average 76.5% and 69.7% to the diet of *S. pilchardus* and *E. encrasicolus*, respectively (Table 4). Group 6 including small to medium-sized organisms from the coast to the slope in the southern part was the third contributor to the diet of both species, with an average contribution of 12.6 ± 7.2% and 14.0 ± 6.8% in *S. pilchardus* and *E. encrasicolus*, respectively (Table 4).

Fish liver $\delta^{13}C$ and $\delta^{15}N$ values and isotopic mixing models

In both spring and autumn 2011, *S. pilchardus* and *E. encrasicolus* differed significantly in liver $\delta^{13}C$ values (both p-values < 0.05). *E. encrasicolus* always displayed lower $\delta^{13}C$ values on average than *S. pilchardus* (Table 3, Fig. 5). However, liver $\delta^{15}N$ values did not differ significantly between both species at both periods. In *S. pilchardus*, individuals sampled in autumn 2011 showed significantly lower $\delta^{13}C$ values and higher $\delta^{15}N$ values than those sampled in spring 2011. In *E. encrasicolus*, individuals collected in autumn 2011 had higher average $\delta^{15}N$ values than those sampled in spring 2011, but $\delta^{13}C$ values did not differ between seasons (Table 3, Fig. 5).

Interestingly in both species, mixing models performed on liver $\delta^{13}C$ and $\delta^{15}N$ values of the fish sampled in spring 2011 showed an average contribution of all the defined prey groups ≥ 5% (Table 4). Four to 5 groups out of the 9 defined presented an average contribution ≥10% in both species, with group 6 (containing small to medium-sized organisms from the coast in the northern part), group 3 (including large organisms from the shelf to the slope in the northern part) and group 9 (corresponding to small to medium-sized organisms from the oceanic area) being common major groups (given here in the increasing order of contribution) for both fish species. Other major groups contributing to their

short-term diet were group 4 (including small to medium-sized organisms from the shelf to the slope in the central to southern part) and group 8 (corresponding to small to medium-sized organisms from the coast to the shelf in the northern part) in *S. pilchardus*, and the group 1 in *E. encrasicolus* (containing small to medium-sized organisms from the slope in the northern part) (Table 4).

In autumn 2011, the results of the mixing models based on liver tissues were quite similar to those obtained with models performed on muscle $\delta^{13}C$ and $\delta^{15}N$ values. The same 3 groups out of the 6 defined contributed significantly to the diet of both species (i.e., groups 4, 5 and 6). Group 4 (corresponding to small to medium-sized organisms from the coast to the shelf in the northern part) contributed more than 50% on average to the diet of both species (53.7 ± 20.6% and 53.0 ± 28.3% in *S. pilchardus* and *E. encrasicolus*, respectively; Table 4). Group 2 including small to medium-sized organisms from the slope in the northern part also contributed 10.7 ± 5.3 % on average to the short-term diet of *E. encrasicolus*.

Discussion

Spatial, temporal and size-related variability of mesozooplankton abundances and isotope values over time

With all stations taken into account within a given period, the average total abundances of mesozooplankton showed a general decreasing trend over the three periods considered with spring 2010 > spring 2011 > autumn 2011. In all cases, copepods dominated the mesozooplankton community, with the exception of some coastal stations (e.g., C4) that sometimes displayed a relatively high percentage of meroplankton or other taxa (e.g., Appendicularia, Cladocerans), especially in spring. These general patterns in the composition of the mesozooplankton community analysed here are consistent with the current knowledge on this compartment concerning European shelf seas (Williams et al. 1994), and more specifically concerning the Bay of Biscay area (Villate et

al. 1997; Valdés and Moral 1998; Plounevez and Champalbert 1999; Albaina and Irigoien 2004). When focusing on abundances and distribution of the 'dominant taxa', which were well correlated with total mesozooplankton abundances, the abundances were generally higher in coastal stations and notably in autumn. This is quite common for neritic areas at this latitude: i.e., maximum densities are generally observed in late spring extending into summer, a secondary peak of high biomass occurs in autumn, and values are minimum in winter. In contrast, oceanic areas generally present a single annual peak in spring, there is no autumn peak or it is very weak, and generally low summer values are observed (Valdés and Moral 1998). In the Bay of Biscay and especially in spring, Plounevez and Champalbert (1999) and Dupuy et al. (2011) effectively reported higher zooplankton biomass in neritic stations and notably those located in the water plume of the Gironde estuary, relative to more oceanic stations. However in our study, abundances were also quite high in stations from the slope relative to coastal stations in spring 2010, with high densities of the copepod *C. helgolandicus* in particular when compared to spring 2011 (Fig. 3).

Spatio-temporal variation in mesozooplankton abundance and composition, especially inter-annual variations (i.e., between 2 consecutive springs) can be directly related to spatial and year-to-year variations in water temperature and salinity (Villate et al. 1997, Zarauz et al. 2007). Moreover in the Bay of Biscay, the plumes of the Gironde and the Loire rivers considerably influence the hydrological structure and the primary production on the continental shelf, all along the year (Planque et al. 2004; Puillat et al. 2004, 2006; Loyer et al. 2006; Dupuy et al. 2011). Slope currents occuring on the shelf-break (Koutsikopoulos and Le Cann 1996) can also favour primary production in these waters due to nutrients inputs (e.g., Holligan and Groom 1986). For instance, Albaina and Irigoien (2004) related peaks of mesozooplankton abundance and distinct mesozooplankton assemblages with the plume of the Gironde river (i.e., nutrients discharge) and the frontal structure associated with the shelf-break (i.e., internal wave generation) in the area. In our study, inter-annual variations in mesozooplankton abundances and composition between both springtime periods can be directly linked to temperature and salinity patterns observed during the sampling campaigns as well, and consequently to a temporal lag between both years in the ecological processes occurring in this area in

spring (i.e., water stratification, planktonic blooms). Indeed, during the survey in spring 2010, sea surface temperatures were low, especially in the northern part of the area (from 12 to 14.5°C), and river discharges were low too (IFREMER survey data; previsions for sea surface physico-chemical parameters by date in the Bay of Biscay may be also found at www.previmer.org/observations). Surface temperatures increased and stratification strengthened only during the second half of the sampling campaign in spring 2010. On the contrary, during the spring 2011 survey, sea surface temperatures over the Bay of Biscay area were high (above the average on the time series PELGAS) and relatively homogeneous over the whole Bay of Biscay area (from 15.5 to 17°C on average). River discharges were as low as in 2010, but temperature depth profiles showed a strong stratification of the water column (IFREMER survey data). Furthermore, there was evidence that a spring bloom had occurred before the survey in 2011. Between both surveys in springs 2010 and 2011, abiotic conditions were thus totally different. Furthermore, the Bay of Biscay is known to face late winter phytoplankton blooms, mainly constituted of diatoms, and this within both the Gironde and Loire rivers plumes (Herbland et al. 1998; Labry et al. 2001; Gohin et al. 2003; Dupuy et al. 2011). This results in early phosphorus limitation in spring that subsequently favours the development of small autotrophic unicellular species on which microzooplankton feeds (Sautour et al. 2000; Dupuy et al. 2011). Interestingly in spring 2010, while temperatures were particularly low and the spring bloom had not already occurred, large organisms such as the copepod *C. helgolandicus* were more abundant than in 2011, and notably in stations from the slope. Coastal zones effectively generally show a larger ratio of small organisms (Sourisseau and Carlotti 2006; Irigoien et al. 2009), and neritic species of copepods are generally smaller in body size than offshore species (Williams et al. 1994). Moreover, *C. helgolandicus* preferentially feeds on diatoms (Irigoien et al. 2000), such as those that can develop in late winter phytoplankton blooms. Differences in hydrological characteristics (e.g., temperature, salinity and water stratification), as well as associated ecological processes described in the literature for the Bay of Biscay area (e.g., different phytoplankton blooms between winter and spring) may thus explain the mesozooplankton variability especially found between both consecutive spring surveys studied here (i.e., late winter conditions in spring 2010 *vs*. advanced spring conditions in spring 2011).

Alternatively, even though mesozooplankton varied greatly over the three periods considered in terms of abundances and composition, patterns of isotopic values within this planktonic compartment were similar from one period to another. There was some inter-specific variability of isotope values linked to the size of organisms, as described previously in Chouvelon et al. (2014). Larger organisms displayed higher $\delta^{15}N$ values than smaller organisms in a given area, reflecting an *a priori* higher trophic level of larger organisms in the planktonic food web. The only exception consisted in particularly low $\delta^{15}N$ values measured in large Decapod larvae analysed as a whole in autumn 2011. In arthropods, crude exoskeleton chitin is effectively depleted in ^{15}N but not in ^{13}C (Schimmelmann and De Niro 1986). As described in Chouvelon et al. (2014), there was also an intra-taxa variability of isotope values linked to spatial patterns in the area, especially concerning $\delta^{15}N$ values that were more variable than $\delta^{13}C$ values between mesozooplanktonic groups of prey. The temporal variability of plankton isotopic signatures, which could have constrained the use of mixing models on liver and muscle data from planktonic prey sampled at only one period (those of the survey) was thus negligible, at least at the scale of the Bay of Biscay ecosystem. In fact, spatial differences in $\delta^{15}N$ values in particular are more likely linked to processes occurring at the dissolved inorganic nitrogen (DIN) level (for a complete review on this subject see Sherwood and Rose 2005; Montoya 2007; and references therein). Many processes can effectively lead to enriched-^{15}N values of the available DIN pool, and the following general conclusions can be drawn: (1) when DIN demand is higher than the supply of nutrients, primary producers may be faced with a ^{15}N-enriched nitrogen source (e.g., “recycled” or-ammonium-enriched, especially if it comes from higher trophic levels), which is then reflected in the local food chain. Alternatively, during upwelling events for instance (in areas subject to this), the physical supply of “new” nutrients overwhelms the biological uptake rate and favours ^{15}N-depleted nitrogen sources (at least non-enriched) for producers of this environment. Moreover, high primary production (blooms) during spring on the continental shelf reduces nutrient quantities, thus favouring ^{15}N-enrichment of the available DIN. Even if short-lived, this effect may be lasting for benthic consumers in particular due to the sinking of particles to the bottom; (2) rivers may be a vector of ^{15}N-enriched organic matter into coastal waters as well (Fry 1988; McClelland et al. 1997; Vizzini and Mazzola 2006). All these processes can be involved in the Bay of Biscay, however

549 the derived-spatial patterns of $\delta^{15}N$ values from the base of the food chain (i.e., investigated at the
550 mesozooplankton level here) were thus similar from one period to another.

551

552 Linking resource variability and feeding patterns of sardines and anchovies over time

553

554 During the three study periods, *S. pilchardus* and *E. encrasicolus* were well segregated by both their
555 $\delta^{13}C$ and $\delta^{15}N$ values as measured in the muscle of individuals. Moreover, mixing models applied on
556 this tissue (medium-term integrator of the food consumed) emphasised different feeding strategies of
557 the two fish species. In both spring periods surveyed (2010 and 2011), *E. encrasicolus* showed a
558 greater trophic plasticity than *S. pilchardus*, both in terms of feeding areas and in terms of sizes of
559 prey organisms among the mesozooplankton resource (i.e. zooplankton > 200 μm). Indeed, almost all
560 the defined groups of mesozooplankton prey presented an average contribution $\geq 5\%$ in
561 *E. encrasicolus*, while only some of the defined groups presented such a contribution to the diet of *S.*
562 *pilchardus* in both spring periods. In terms of feeding areas, groups 8 and 6 (in Spring 2010 and
563 Spring 2011, respectively) containing organisms from the coast to the slope effectively showed the
564 highest contribution to the diet of sardines (i.e., 43.7 ± 5.9% and 40.8 ± 16.0%, respectively). It
565 suggests that sardines are more limited to coastal areas and the mesozooplanktonic species of these
566 waters for feeding than anchovies. Besides, these groups showed the highest $\delta^{15}N$ values at both
567 periods, which is in accordance with the highest $\delta^{15}N$ values measured in muscle tissue of
568 *S. pilchardus* at the two periods, and which also suggests that the feeding pattern of pelagic fish is
569 constrained spatially. Indeed, in terms of sizes of prey, significantly lower $\delta^{13}C$ and $\delta^{15}N$ values
570 measured in the muscle of anchovies collected in both springs 2010 and 2011 could have been related,
571 at first sight, to the consumption of lower trophic level organisms in anchovies. However, the spatial
572 variability of $\delta^{13}C$ and $\delta^{15}N$ values from the base of the different food webs in the area (Chouvelon et
573 al., 2012), and also shown here with isotope values of mesozooplanktonic species, rather supports the
574 hypothesis of more offshore feeding habits for anchovies than the hypothesis of a lower trophic level.

Anchovies would effectively be able to capture larger particles than sardines (Louw et al. 1998; Van der Lingen et al. 2006), thanks to differences in gill-raker morphology between both species and the existence of a larger branchial apparatus in anchovies (James and Findlay 1989). In several cases, anchovies have thus been found to feed at a slightly higher trophic level than sardines (e.g., Stergiou and Karpouzi, 2002), and specifically in the Bay of Biscay (i.e., data from Ecopath modelling; Lassalle et al., 2011). Moreover, this morphological difference would lead anchovies to be opportunistic and efficient planktivores (James and Probyn 1989) on prey species from the mesozooplankton compartment at least, and would confirm that *E. encrasicolus* is not specialist feeder in the Bay of Biscay area, as already reported for the North and Baltic Seas (Raab et al. 2011). Such particulate-feeding in anchovies allows for a rapid and efficient intake of prey minimising metabolic costs, and is thus the main feeding mode in this species (James and Probyn 1989; Van der Lingen 1994). In contrast, filter-feeding on smaller zooplanktonic prey and/or phytoplankton would be the major feeding mode in sardines (Van der Lingen 1994; Garrido et al. 2007). However, most dietary carbon and/or nitrogen is obtained from zooplanktonic prey (and not phytoplankton) in adult sardines in general (Van der Lingen 1994; Bode et al. 2004; Nikolioudakis et al. 2011; Costalago et al. 2012), and the contribution of phytoplankton to sardine diet can vary greatly at small spatial scales and seasonally (Garrido et al. 2008).

Medium-term feeding preferences of sardines and anchovies differed within both spring periods studied here. Alternatively, their diets were relatively similar during the autumn period following our mixing model results, whereas average isotope values were significantly different (although associated standard deviations were large). This may be due to the fact that the isotopic mixing models used here consider individual fish values (i.e., consumers), and not mean values ± standard deviation as for prey (Parnell et al. 2010). As such, mixing models based on muscle tissues highlighted a preference of both species for small to medium-sized organisms from neritic waters (i.e., from the coast to the shelf) in central and northern parts of the Bay of Biscay, which notably corresponds to the autumn/winter-feeding grounds described for anchovies in this area (ICES 2010b). In fact, it appeared that the more abundant and diversified the mesozooplankton resource is in terms of

prey sizes available (i.e., with spring 2010 > spring 2011 > autumn 2011), the more sardines are specialised on fewer prey groups compared to anchovies (Table 2). Indeed, 25% of the groups of prey (i.e., 3 out of the 8 defined) contributed on average to 87.3% to the medium-term diet of *S. pilchardus* in spring 2010, while 45% of the groups contributed to 82.4% to its diet in spring 2011, and 50% of the groups contributed to 89.1% to its diet in autumn 2011. In autumn 2011, the same groups contributed to 83.7% to the medium-term diet of *E. encrasicolus*. Thus, when the mesozooplankton resource is abundant and diversified (i.e., in both springs compared to the autumn period), and while potential competition could be high because of some spawning overlap between the two species (ICES 2010b), it is likely that the high degree of specialisation shown by sardines limits competition with anchovies (and with other small pelagic fish in general) in spring. On the contrary, trophic overlap could occur in autumn, when the resource is less abundant and diversified, leading to potential competition for food between both fish species. Moreover during this period, it has been reported that the fat content of both species peaks (ICES 2010b), indicating a common period of need for reserve storages before the beginning of the spawning season (i.e., for sardine), or before winter (for anchovy). However, both species are able to feed throughout the year and notably during the spawning season, which may limit the competition for resource in autumn as well.

In spring 2010, major contributing groups of prey to the medium-term diet of both fish species were mostly constituted of small to medium-sized organisms from neritic waters, despite a wider range of prey sizes and of feeding areas for *E. encrasicolus* as noticed above. In contrast, in spring 2011, 2 out of the 4 major groups of prey for both species (i.e., contributing more than 10% to the medium-term diet of both species) contained large organisms from the shelf to slope areas. Interestingly, this was not in accordance with the reported differences in abundance and diversity of mesozooplanktonic prey between the two consecutive springs, both in terms of sizes available (i.e., abundance of larger prey in spring 2010 > spring 2011) and in terms of mesozooplankton distribution in the area (i.e., abundant species were more fairly distributed between coastal and shelf to slope areas in spring 2010, whereas abundances were slightly higher in coastal areas in spring 2011). Furthermore, if the mesozooplankton community showed variation from one spring to another, this did not visibly

impact the medium-term feeding strategies of both species, which remained the same (i.e., general segregation). Therefore, our results do not highlight any obvious link between variation observed in the mesozooplankton resource and the trophic ecology of both fish species depicted through SIA, at least concerning both spring periods studied. In autumn 2011, the spread of isotope values for anchovies was relatively large. Chouvelon et al. (2012) already reported such a wide range of $\delta^{15}N$ values in anchovies sampled in the autumns of 2009 and 2010 in the Bay of Biscay, in comparison to individuals sampled in springs 2009 and 2010 and in comparison with sardines sampled at the same periods. As a potential explanation, the authors argued for two different hypotheses. The first one is related to the high mobility of most small pelagic fish species (e.g., Nøttestad et al., 1999). Indeed, we cannot exclude here a potential mixing of individuals and/or part of the population that have fed in different areas presenting different baseline signatures in $\delta^{15}N$ in the Bay of Biscay, particularly in autumn when food supply is less abundant in neritic waters. The second hypothesis refers to a possible greater trophic plasticity of anchovies so as to avoid competition with sardines at this period of the year, as an adjustment on behalf of the species facing variations in the food supply (e.g., Lefebvre et al. 2009). In autumn, abundances of mesozooplankton may effectively stay at levels that anyway sustain energetic needs of both species and other plankton-feeders. For instance, Plounevez and Champalbert (1999) already suggested that feeding efficiency in *E. encrasicolus* would be more related to zooplankton specific composition than to zooplankton abundance, even if the results of our study cannot confirm or invalidate this hypothesis. Marquis et al. (2011) also reported that small pelagic fish only represent 30% of the total predation on the mesozooplanktonic compartment in coastal stations in the Bay of Biscay (from spring data), and 60 and 65% at the mid-shelf and the slope stations, respectively. These authors suggested that a large fraction of the mesozooplankton production would be then available for other planktivorous organisms such as suprabenthic zooplankton (euphausiids and mysids) or macrozooplankton (medusae or large tunicates) in the Bay of Biscay (Marquis et al. 2011). Finally, this could also explain why the variations observed in the mesozooplanktonic community in the present study do not fully correlate with the trophic ecology of adult anchovies and sardines, depicted here through SIA over the three periods investigated.

The lack of relationships between variations in the mesozooplankton resource and the trophic ecology of both species may be also due to the fact that until now, only the trophic ecology inferred from muscle isotope values (i.e., a medium-term integrator of the food assimilated) was considered because this was the tissue commonly sampled over the three periods. Indeed, as described above, variation in the plankton community *a priori* depend on short-term events such as phytoplankton blooms; so analysis of liver stable isotope values (a shorter-term integrative tissue) could be more relevant for comparison with resource variability. As such, in spring 2011, contrary to values measured in the muscle, $\delta^{15}N$ values in particular measured in the liver did not differ significantly between both species. Moreover, mixing models highlighted a common predominant group of prey (i.e., group 6), contributing to more that 20% in both species and corresponding to small to medium-sized organisms from the coast in the northern part. In the liver tissue of fish, carbon and nitrogen half-lives were shown to be considerably lower than in the muscle (e.g., Buchheister and Latour 2010 for flatfish) and in fact, from hepatic results, it was likely that both sardines and anchovies appeared to be short-term opportunistic feeders in spring 2011 (i.e., all prey groups contribution $\geq 5\%$). Although this pattern of quite similar average contributions for most prey groups may be an indication that the model cannot reliably find a fit for the data, this could be also related, in terms of ecological interpretation, to a temporary opportunistic behaviour of both species that are facing short-term variation in food availability. This would be also quite consistent with the fact that both species may feed during spawning season, with the spawning season potentially overlapping between the two species during this period (spring). However, the main contributing prey groups revealed by mixing models based on liver isotope values did not fully correspond to the most abundant prey items available in the Bay of Biscay at the period of sampling. So, results of mixing models performed on the liver tissue did not reveal any clear relationships between either the food available or that assimilated by the two fish species. Nonetheless in autumn 2011, results obtained in the livers corroborated those obtained in the muscles, with an apparent sharing of the mesozooplankton resource at this period. Finally, the lack of precise TEFs for planktivorous fish may be also responsible of potentially imprecise results, highlighting the recurrent crucial needs for more experimental studies in isotopic ecology (Martínez del Rio et al. 2009). This is particularly true for isotope values measured in fish liver, as many

dedicated studies focus on the muscle tissue as the reference tissue for the study of trophic interactions (Pinnegar and Polunin 1999).

Concluding remarks and further work for understanding small pelagic fish fluctuations

SIA represents an alternative and/or complementary method for determining the diets and feeding strategies of small sympatric pelagic fish species (e.g., Costalago et al. 2012). The results of the present study highlighted that it also provides useful information on potential trophic overlap between species in the very general context of understanding forage fish alternations and/or co-occurrence in a given area. In the Bay of Biscay, it effectively appeared that adults of sardines and anchovies do not compete strongly for the mesozooplankton resource in spring, where the spawning season of both species overlap and during which their energetic needs may be increased. In autumn, potential competition for the mesozooplankton resource may occur, although this may be compensated by the fact that both species feed throughout the year (ICES 2010b) and notably in spring when the food resource is abundant. Alternatively, in the present study, no clear relationships were revealed between the trophic ecology of adult sardines and anchovies depicted through SIA, and variations in the mesozooplankton resource in the Bay of Biscay area over the three different periods investigated. Other food resources than mesozooplankton (i.e., microplankton) may also contribute to their diet, and the lack of consideration of this compartment here may contribute to explain the lack of relationships (in addition to the other elements described above such as imprecise TEFs for plankton-feeding fish, for instance). However in the Bay of Biscay, the microplankton fraction (i.e., 50-200µm) appears in fact to be mainly constituted by phytoplankton (unpublished data). Moreover, several studies demonstrated that zooplankton, and notably copepods belonging to the mesozooplankton community, is by far the most important dietary component for both fish species compared to phytoplankton (e.g., Van der Lingen et al. 2006; Espinoza et al. 2009; Nikolioudakis et al. 2012). Furthermore, this is not the first time that a lack of relationship between food concentration and food ingestion in such small

710 plankton-feeding fish is found in the Bay of Biscay (e.g., Plounevez and Champalbert 1999; Bachiller
711 et al. 2012). Interestingly, in other systems, some authors have however already shown that feeding
712 mode and food consumption in adult sardines, for instance, can be highly dependent on food density
713 (Garrido et al. 2007), notably in the Mediterranean Sea (Nikolioudakis et al. 2011; Costalago et al.
714 2012).

715 Differences in the general function of the different systems may induce such differences in the
716 feeding strategies of small pelagic fish between systems. Indeed, in upwelling systems for instance,
717 alternative abundance fluctuations of sardines and anchovies have been demonstrated and partly
718 explained by both climatic (e.g., Lluch-Belda et al. 1989; Schwartzlose et al. 1999) and/or biological
719 factors (e.g., trophodynamic mediation suggested by Louw et al. 1998; Van der Lingen et al. 2006).
720 When two predator species show clear trophodynamic differences, as demonstrated for sardines and
721 anchovies in various ecosystems and notably in upwelling systems in terms of size of prey, there is
722 effectively a high potential for trophodynamically mediated fluctuations of both species abundances if
723 a peculiar food environment (dominated by either small or large particles) persist either spatially
724 and/or temporally under specific abiotic conditions. Indeed, it may favour the occurrence/maintenance
725 of one of the predator species relative to the other in the area, a phenomena that would be enhanced by
726 concurrent better reproductive success of this predator (Van der Lingen et al. 2006). In the Bay of
727 Biscay case study, sardines and anchovies are generally segregated in terms of trophic ecology,
728 highlighting a potential for trophodynamically fluctuations of both species' abundances in the area at
729 first sight. However, they both showed at the same time a certain trophic plasticity relative to the
730 composition of the mesozooplankton resource available, although this trophic plasticity appeared to be
731 higher in anchovy than in sardine. As such here, while anchovies were shown to efficiently remove
732 large particles in various systems (see Van der Lingen et al. 2006 for a review, and the present study
733 for the Bay of Biscay area), large organisms did not necessarily dominate the diet of anchovies when
734 the mesozooplankton resource contained a higher proportion of large organisms (such as in spring
735 2010). Conversely, while sardines were shown to efficiently remove or favour smaller particles in
736 various systems (see Van der Lingen et al. 2006 for a general review, and the present study for the Bay

737 of Biscay area), large organisms could also contribute to their diets when the mesozooplankton
738 resource was largely dominated by small to medium-sized organisms (such as in spring 2011).

739 In the Bay of Biscay ecosystem, no clear patterns of abundances of both fish species and no
740 potential explanation for fluctuations of their stocks have been reported yet. The present study
741 therefore emphasised that fluctuations in sardines and anchovies from the Bay of Biscay cannot also
742 be totally explained by the trophic ecology of adults of both species. Indeed, adult sardines and
743 anchovies do not compete strongly for food resource in the Bay of Biscay area. Furthermore, species
744 segregate diets, and although this can represent a potential for trophodynamically mediated
745 fluctuations under specific abiotic conditions, no clear link was made between food resource
746 availability and fish diets (i.e., no strict dependency of both species relative to the composition and
747 availability of the mesozooplankton resource). In this sense, our results seem to corroborate those of
748 Irigoien et al. (2009) who found a negative correlation between anchovy recruitment and zooplankton
749 biomass in the Bay of Biscay, suggesting then that the 2002-2006 failures in anchovy recruitment in
750 the area was not due to a decrease in mesozooplankton biomass.

751 Through the results of the present study, we provide further evidence that alternations of
752 species should be considered in conjunction with spawning success and year class formation (Van der
753 Lingen et al. 2006). Besides, a focus on the trophic ecology of larvae of both species may also
754 constitute a next step. This should be then coupled to an analysis of long-term fluctuations in
755 mesozooplankton and microplankton (may be preyed on by larvae as well) composition and
756 abundance, with consideration of abiotic factors too. Changes in the plankton community, in relation
757 to environmental parameters, have effectively been shown to directly affect survival of larvae and
758 consequently fish recruitment (e.g., Beaugrand et al. 2003). From a more theoretical ecological point
759 of view, investigating what maintains the trophic segregation between adults of both species, despite
760 variations in abundance and composition of the mesozooplankton community, should be also of
761 interest.

762

Acknowledgements

This work was supported by the European project REPRODUCE (EratNet-Marifish, FP7). Authors are very grateful to J. Massé, P. Petitgas, and M. Doray from IFREMER for facilitating the sampling during PELGAS surveys. Many thanks to M. Huret and P. Bourriau (IFREMER) too for their help during plankton sample collection, and P. Lespagnol (IFREMER) for fish sampling. Authors also thank G. Guillou and P. Richard (UMR LIENSs) for assistance in stable isotope analysis, and finally the anonymous reviewers of the earlier version of the manuscript for facilitating its improvement.

References

Albaina A, Irigoien X (2004) Relationships between frontal structures and zooplankton communities along a cross-shelf transect in the Bay of Biscay (1995 to 2003). Mar Ecol Prog Ser 284:65-75

Alheit J, Pohlmann T, Casini M, Greve W, Hinrichs R, Mathis M, O'Driscoll K, Vorberg R, Wagner C (2012) Climate variability drives anchovies and sardines into the North and Baltic Seas. Prog Oceanogr 96:128-139

Bachiller E, Cotano U, Boyra G, Irigoien X (2012) Spatial distribution of the stomach weights of juvenile anchovy (*Engraulis encrasicolus* L.) in the Bay of Biscay. ICES J Mar Sci. doi.10.1093/icesjms/fss176

Barange M, Coetzee J, Takasuka A, Hill K, Gutierrez M, Oozeki Y, Van der Lingen C, Agostini V (2009) Habitat expansion and contraction in anchovy and sardine populations. Prog Oceanogr 83:251-260

Beaugrand G, Brander KM, Lindley JA, Souissi S, Reid PC (2003) Plankton effect on cod recruitment in the North Sea. Nature 426:661-664

787 Bode A, Álvarez-Ossorio MT, Carrera P, Lorenzo J (2004) Reconstruction of trophic pathways
788 between plankton and the North Iberian sardine (*Sardina pilchardus*) using stable isotopes. Sci Mar
789 68:165-178

790 Bond AL, Diamond AW (2011) Recent Bayesian stable-isotope mixing models are highly sensitive to
791 variation in discrimination factors. Ecol Appl 21:1017-1023

792 Buchheister A, Latour RJ (2010) Turnover and fractionation of carbon and nitrogen stable isotopes in
793 tissues of a migratory coastal predator, summer flounder (*Paralichthys dentatus*). Can J Fish Aquat Sci
794 67:445-461

795 Cabal J, González-Nuevo G, Nogueira E (2008) Mesozooplankton species distribution in the NW and
796 N Iberian shelf during spring 2004: Relationship with frontal structures. J Mar Syst 72:282-297

797 Caut S, Angulo E, Courchamp F (2009) Variation in discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$): the effect
798 of diet isotopic values and applications for diet reconstruction. J Appl Ecol 46:443-453

799 Chavez FP, Ryan J, Lluch-Cota SE, Ñiquen CM (2003) From anchovies to sardines and back:
800 multidecadal change in the Pacific Ocean. Science 299:217-221

801 Chouvelon T, Chappuis A, Bustamante P, Lefebvre S, Mornet F, Guillou G, Violamer L, Dupuy C
802 (2014) Trophic ecology of European sardine *Sardina pilchardus* and European anchovy *Engraulis*
803 *encrasicolus* in the Bay of Biscay (north-east Atlantic) inferred from $\delta^{13}C$ and $\delta^{15}N$ values of fish and
804 identified mesozooplanktonic organisms. J Sea Res 85:277-291

805 Chouvelon T, Spitz J, Caurant F, Mèndez-Fernandez P, Chappuis A, Laugier F, Le Goff E,
806 Bustamante P (2012) Revisiting the use of $\delta^{15}N$ in meso-scale studies of marine food webs by
807 considering spatio-temporal variations in stable isotopic signatures – The case of an open ecosystem:
808 the Bay of Biscay (North-East Atlantic). Prog Oceanogr 101:92-105

809 Costalago D, Navarro J, Álvarez-Calleja I, Palomera I (2012) Ontogenetic and seasonal changes in the
810 feeding habits and trophic levels of two small pelagic fish species. Mar Ecol Prog Ser 460:169-181

811 Cury P, Bakun A, Crawford RJM, Jarre A, Quiñones RA, Shannon LJ, Verheye HM (2000) Small
812 pelagics in upwelling systems: patterns of interaction and structural changes in “wasp-waist”
813 ecosystems. ICES J Mar Sci 57:603-618

814 Cury P, Roy C (1989) Optimal environmental window and pelagic fish recruitment success in
815 upwelling areas. Can J Fish Aquat Sci 46:670-680

816 De Niro MJ, Epstein S (1977) Mechanism of carbon fractionation associated with lipid synthesis.
817 Science 197:261-263

818 De Niro MJ, Epstein S (1978) Influence of diet on the distribution of carbon isotopes in animals.
819 Geochim Cosmochim Acta 42:495-506

820 De Niro MJ, Epstein S (1981) Influence of diet on the distribution of nitrogen isotopes in animals.
821 Geochim Cosmochim Acta 45:341-351

822 Dubreuil J, Petitgas P (2009) Energy density of anchovy *Engraulis encrasicolus* in the Bay of Biscay.
823 Journal of Fish Biology 74:521-534

824 Dupuy C, Talarmin A, Hartmann HJ, Delmas D, Courties C, Marquis E (2011) Community structure
825 and grazing of the nano-microzooplankton on the continental shelf of the Bay of Biscay. Estuar Coast
826 Shelf S 95:1-13

827 Espinoza P, Bertrand A, Van der Lingen CD, Garrido S, Rojas de Mendiola B (2009) Diet of sardine
828 (Sardinops sagax) in the northern Humboldt Current system and comparison with the diets of
829 clupeoids in this and other eastern boundary upwelling systems. Prog Oceanogr 83:242-250

830 FAO (Food and Agriculture Organization of the United Nations) (2012) The state of world fisheries
831 and aquaculture 2010. Rome, FAO, 2012, 209 p

832 France RL (1995) Carbon-13 enrichment in benthic compared to planktonic algae: food web
833 implications. Mar Ecol Prog Ser 124:307-312

834 Fry B (1988) Food web structure on Georges Bank from stable C, N, and S isotopic compositions.
835 Limnol Oceanogr 33:1182-1190

836 Garrido S, Marçalo A, Zwolinski J, Van der Lingen CD (2007) Laboratory investigations on the effect
837 of prey size and concentration on the feeding behaviour of *Sardina pilchardus*. Mar Ecol Prog Ser
838 330:189-199

839 Garrido S, Rosa R, Ben-Hamadou R, Cunha ME, Chícharo MA, Van der Lingen CD (2008) Spatio-
840 temporal variability in fatty acid trophic biomarkers in stomach contents and muscle of Iberian sardine
841 (*Sardina pilchardus*) and its relationship with spawning. Mar Biol 154:1053-1065

842 Gohin F, Lampert L, Guillaud JF, Herbland A, Nézan E (2003) Satellite and in situ observations of a
843 late winter phytoplankton bloom, in the northern Bay of Biscay. Cont Shelf Res 23:1117-1141

844 Guelinckx J, Maes J, Van Den Driessche P, Geysen B, Dehairs F, Ollevier F (2007) Changes in $\delta^{13}C$
845 and $\delta^{15}N$ in different tissues of juvenile sand goby *Pomatoschistus minutus*: a laboratory diet-switch
846 experiment. Mar Ecol Prog Ser 341:205-215

847 Herbland A, Delmas D, Laborde P, Sautour B, Artigas F (1998) Phytoplankton spring bloom of the
848 Gironde plume waters in the Bay of Biscay: early phosphorus limitation and food web consequences.
849 Oceanol Acta 21:279-291

850 Hobson KA (1999) Tracing origins and migration of wildlife using stable isotopes: a review.
851 Oecologia 120:314-326

852 Hobson KA, Clark RG (1992) Assessing avian diets using stable isotopes. I: Turnover of C in tissues.
853 Condor 94:181-188

854 Holligan PM, Groom SB (1986) Phytoplankton distributions along the shelf break. P Roy Soc Edinb B
855 88:239-263

856 ICES (2010a) Report of the Working Group on Anchovy and Sardine (WGANSA). ICES WGANSA
857 Report 2010, ICES, 289 p.

858 ICES (2010b) Life-cycle spatial patterns of small pelagic fish in the Northeast Atlantic. ICES
859 Cooperative Research Report, ICES, 94 p.

860 Irigoien X, Fernandes JA, Grosjean P, Denis K, Albaina A, Santos M (2009) Spring zooplankton
861 distribution in the Bay of Biscay from 1998 to 2006 in relation with anchovy recruitment. J Plankton
862 Res 31:1-17

863 Irigoien X, Head RN, Harris RP, Cummings D, Harbour D, Meyer-Harms B (2000) Feeding
864 selectivity and egg production of *Calanus helgolandicus* in the English Channel. Limnol Oceanogr
865 45:44-54

866 Irigoien X, De Roos A (2011) The role of intraguild predation in the population dynamics of small
867 pelagic fish. Mar Biol 158:1683-1690

868 Isla JA, Ceballos S, Huskin I, Anadón R, Álvarez-Marqués F (2004) Mesozooplankton distribution,
869 metabolism and grazing in an anticyclonic slope water oceanic eddy (SWODDY) in the Bay of
870 Biscay. Mar Biol 145:1201-1212

871 James AG, Findlay KP (1989) Effect of particle size and concentration on feeding behavior, selectivity
872 and rates of food ingestion by the Cape anchovy *Engraulis capensis*. Mar Ecol Prog Ser 50:275-294

873 James AG, Probyn T (1989) The relationship between respiration rate, swimming speed and feeding
874 behaviour in the Cape anchovy *Engraulis capensis* Gilchrist. J Exp Mar Biol Ecol 131:81-100

875 Koutsikopoulos C, Le Cann B (1996) Physical processes and hydrological structures related to the Bay
876 of Biscay anchovy. Sci Mar 60:9-19

877 Labry C, Herbland A, Delmas D, Laborde P, Lazure P, Froidefond JM, Jégou AM, Sautour B (2001)
878 Initiation of winter phytoplankton blooms within the Gironde plume waters in the Bay of Biscay. Mar
879 Ecol Prog Ser 212:117-130

880 Lassalle G, Lobry J, Le Loc'h F, Bustamante P, Certain G, Delmas D, Dupuy C, Hily C, Labry C, Le
881 Pape O, Marquis E, Petitgas P, Pusineri C, Ridoux V, Spitz J, Niquil N (2011) Lower trophic levels

882 and detrital biomass control the Bay of Biscay continental shelf food web: implications for ecosystem
883 management. Prog Oceanogr 91:561-575

884 Lefebvre S, Marín Leal JC, Dubois S, Orvain F, Blin JL, Bataillé MP, Ourry A, Galois R (2009)
885 Seasonal dynamics of trophic relationships among co-occurring suspension feeders in two shellfish
886 culture dominated ecosystems. Estuar Coastal Shelf Sci 82:415-425

887 Lluch-Belda D, Crawford RJM, Kawasaki T, MacCall AD, Parrish RH, Schwartzlose RA, Smith PE
888 (1989) World-wide fluctuations of sardine and anchovy stocks: the regime problem. S Afr J Mar Sci
889 8:195-205

890 Lluch-Belda D, Schwartzlose RA, Serra R, Parrish R, Kawasaki T, Hedgecock D, Crawford RJM
891 (1992) Sardine and anchovy regime fluctuations of abundance in four regions of the world oceans: a
892 workshop report. Fish Oceanogr 1:339-347

893 Lorance P, Bertrand JA, Brind'Amour A, Rochet MJ, Trenkel VM (2009) Assessment of impacts from
894 human activities on ecosystem components in the Bay of Biscay in the early 1990s. Aquat Living
895 Resour 22:409-431

896 Louw GG, Van der Lingen CD, Gibbons MJ (1998) Differential feeding by sardine *Sardinops Sagax*
897 and anchovy *Engraulis capensis* recruits in mixed shoals. S Afr J Mar Sci 19:227-232

898 Loyer S, Lampert L, Menesguen A, Cann P, Labasque T (2006) Seasonal evolution of the nutrient
899 pattern on Biscay Bay continental shelf over the years 1999-2000. Sci Mar 70:31-46

900 Marquis E, Niquil N, Vézina AF, Petitgas P, Dupuy C (2011) Influence of planktonic food web
901 structure on a system's capacity to support pelagic production: an inverse analysis approach. ICES J
902 Mar Sci 68:803-812

903 Martínez del Rio C, Wolf N, Carleton SA, Gannes LZ (2009) Isotopic ecology ten years after a call for
904 more laboratory experiments. Biol Rev 84:91-111

McClelland JM, Valiela I, Michener RH (1997) Nitrogen-stable isotope signatures in estuarine food webs: a record of increasing urbanization in coastal watersheds. Limnol Oceanogr 42:930-937

Michener RH, Kaufman L (2007) Stable isotope ratios as tracers in marine food webs: an update. In: Michener R, Lajtha K (eds) Stable isotopes in ecology and environmental science. Blackwell Publishing Ltd, pp 238-282

Montoya JP (2007) Natural abundance of ^{15}N in marine planktonic ecosystems. In: Michener R, Lajtha K (eds) Stable isotopes in ecology and environmental science. Blackwell Publishing Ltd, pp 76-201

Motos L (1996) Reproductive biology and fecundity of the Bay of Biscay anchovy population (*Engraulis encrasicolus* L.). Scientia Marina 60:195-207

Nikolioudakis N, Palomera I, Machias A, Somarakis S (2011) Diel feeding intensity and daily ration of the sardine *Sardina pilchardus*. Mar Ecol Prog Ser 437:215-228

Nikolioudakis N, Isari S, Pitta P, Somarakis S (2012) Diet of sardine *Sardina pilchardus*: an "end-to-end" field study. Mar Ecol Prog Ser 453:173-188

Nøttestad L, Giske J, Holst JC, Huse G (1999) A length-based hypothesis for feeding migrations in pelagic fish. Canadian J Fish Aquat Sci 56:26-34.

OSPAR (2010) Quality Status Report 2010. OSPAR Commission, London, 176 p

Parnell AC, Inger R, Bearhop S, Jackson AL (2010) Source partitioning using stable isotopes: coping with too much variation. Plos One 5:e9672, doi:10.1371/journal.pone.0009672

Parnell AC, Phillips DL, Bearhop S, Semmens BX, Ward EJ, Moore JW, Jackson AL, Grey J, Kelly DJ, Inger R (2013) Bayesian stable isotope mixing models. Environmetrics 24: 387-399

Peterson BJ, Fry B (1987) Stable isotopes in ecosystem studies. Annu Rev Ecol Syst 18:293-320

Phillips DL, Gregg JW (2003) Source partitioning using stable isotopes: coping with too many sources. Oecologia 136:261-269

928 Phillips DL, Inger R, Bearhop S, Jackson AL, Moore JW, Parnell AC, Semmens BX, Ward EJ (2014)
929 Best practices for use of stable isotope mixing models in food web studies. Can J Zool 10.1139/cjz-
930 2014-0127

931 Phillips DL, Newsome SD, Gregg JW (2005) Combining sources in stable isotope mixing models:
932 alternative methods. Oecologia 144:520-527

933 Pinnegar JK, Polunin NVC (1999) Differential fractionation of $\delta^{13}C$ and $\delta^{15}N$ among fish tissues:
934 implications for the study of trophic interactions. Funct Ecol 13:225-231

935 Planque B, Lazure P, Jégou AM (2004) Detecting hydrological landscapes over the Bay of Biscay
936 continental shelf in spring. Climate Res 28:41-52

937 Plounevez S, Champalbert G (1999) Feeding behaviour and trophic environment of *Engraulis*
938 *encrasicolus* (L.) in the Bay of Biscay. Estuar Coast Shelf S 49:177-191

939 Post DM (2002) Using stable isotopes to estimate trophic position: models, methods and assumptions.
940 Ecology 83:703-718

941 Puillat I, Lazure P, Jégou AM, Lampert L, Miller PI (2004) Hydrographical variability on the French
942 continental shelf in the Bay of Biscay, during the 1990s. Cont Shelf Res 24:1143-1163

943 Puillat I, Lazure P, Jégou AM, Lampert L, Miller PI (2006) Mesoscale hydrological variability
944 induced by northwesterly wind on the French continental shelf of the Bay of Biscay. Sci Mar 70S1:15-
945 26

946 Raab K, Nagelkerke LAJ, Boerée C, Rijnsdorp AD, Temming A, Dickey-Collas M (2011) Anchovy
947 *Engraulis encrasicolus* diet in the North and Baltic Seas. J Sea Res 65:131-140

948 Rose M (1933) Copépodes pélagiques. Faune de France 26, Paris

949 Sautour B, Artigas LF, Delmas D, Herbland A, Laborde P (2000) Grazing impact of micro and
950 mesozooplankton during a spring situation in coastal waters off the Gironde estuary. J Plankton Res
951 22:531-552

952 Schimmelmann A, De Niro MJ (1986) Stable isotopic studies on chitin. II. The $^{13}C/^{12}C$ and $^{15}N/^{14}N$
953 ratios in arthropod chitin. Contrib. Mar Sci 29:113-130

954 Schwartzlose RA, Alheit J, Bakun A, Baumgartner TR, Cloete R, Crawford RJM, Fltechern WJ,
955 Green-Ruiz Y, Hagen E, Kawasaki T, Lluch-Belda D, Lluch-Cota SE, MacCall AD, Matsuura Y,
956 Nevarez-Martinez MO, Parrish RH, Roy C, Serra R, Shust KV, Ward MN, Zuzunaga JZ (1999)
957 Worlwide large-scale fluctuations of sardine and anchovy populations. S Afr J Mar Sci 21:289-347

958 Sherwood GD, Rose GA (2005) Stable isotope analysis of some representative fish and invertebrates
959 of the Newfoundland and Labrador continental shelf food web. Estuar Coast Shelf S 63:537-549

960 Sourisseau M, Carlotti F (2006) Spatial distribution of zooplankton size spectra on the French
961 continental shelf of the Bay of Biscay during spring 2000 and 2001. J Geophys Res Oceans
962 111:C05S09

963 Sponheimer M, Robinson TF, Cerling TE, Tegland L, Roeder BL, Ayliffe L, Dearing MD, Ehleringer
964 JR (2006) Turnover of stable carbon isotopes in the muscle, liver, and breath CO of alpacas (*Lama*
965 *pacos*). Rapid Commun Mass Spectrom 20:1395-1399

966 Stergiou KI, Karpouzi VS (2002) Feeding habits and trophic levels of Mediterranean fish. Reviews in
967 Fish Biology and Fisheries 11:217-254

968 Suzuki KW, Kasai A, Nakayama K, Tanaka M (2005) Differential isotopic enrichment and half-life
969 among tissues in Japanese temperate bass (*Lateolabrax japonicus*) juveniles: implications for
970 analysing migration. Can J Fish Aquat Sci 62:671-678

971 Sweeting CJ, Barry JT, Barnes C, Polunin NVC, Jennings S (2007a) Effects of body size and
972 environment on diet-tissue $\delta^{15}N$ fractionation in fishes. J Exp Mar Biol Ecol 340:1-10

973 Sweeting CJ, Barry JT, Barnes C, Polunin NVC, Jennings S (2007b) Effects of body size and
974 environment on diet-tissue $\delta^{13}C$ fractionation in fishes. J Exp Mar Biol Ecol 352:165-176

975 Takasuka A, Oozeki Y, Aoki I (2007) Optimal growth temperature hypothesis: why do anchovy
976 flourish and sardine collapse or vice versa under the same ocean regime? Can J Fish Aquat Sci 64:
977 768-776

978 Tieszen LL, Boutton TW, Tesdahl KG, Slade NA (1983) Fractionation and turnover of stable carbon
979 isotopes in animal tissues: implications for $\delta^{13}C$ analysis of diet. Oecologia 57:32-37

980 Trueman CN, McGill RAR, Guyard PH (2005) The effect of growth rate on tissue-diet isotopic
981 spacing in rapidly growing animals. An experimental study with Atlantic salmon (*Salmo salar*). Rapid
982 Commun Mass Spectrom 19:3239-3247

983 Valdés L, López-Urrutia A, Cabal J, Alvarez-Ossorio M, Bode A, Miranda A, Cabanas M, Huskin I,
984 Anadón R, Alvarez-Marqués F, Llope M, Rodríguez N (2007) A decade of sampling in the Bay of
985 Biscay: What are the zooplankton time series telling us? Prog Oceanogr 74:98-114

986 Valdés L, Moral M (1998) Time-series analysis of copepod diversity and species richness in the
987 southern Bay of Biscay off Santander, Spain, in relation to environmental conditions. ICES J Mar Sci
988 55:783-792

989 Vanderklift MA, Ponsard S (2003) Source of variation in consumer-diet $\delta^{15}N$ enrichment: a meta-
990 analysis. Oecologia 136:169-182

991 Van der Lingen CD (1994) Effect of particle size and concentration on the feeding behaviour of adult
992 pilchard *Sardinops sagax*. Mar Ecol Prog Ser 109:1-13

993 Van der Lingen CD, Hutchings L, Field JG (2006) Comparative trophodynamics of anchovy
994 *Engraulis encrasicolus* and sardine *Sardinops sagax* in the southern Benguela: are species alternations
995 between small pelagic fish trophodynamically mediated? Afr J Mar Sci 28:465-478

996 Vander Zanden MJ, Cabana G, Rasmussen JB (1997) Comparing trophic position of freshwater fish
997 calculated using stable nitrogen isotope ratios ($\delta^{15}N$) and literature dietary data. Can J Fish Aquat Sci
998 54:1142-1158

Villate F, Moral M, Valencia V (1997) Mesozooplankton community indicates climate changes in a shelf area of the inner Bay of Biscay throughout 1988 to 1990. J Plankton Res 19:1617-1636

Vizzini S, Mazzola A (2006) The effects of anthropogenic organic matter inputs on stable carbon and nitrogen isotopes in organisms from different trophic levels in a southern Mediterranean coastal area. Sci Total Environ 368:723-731

Williams R, Conway DVP, Hunt HG (1994) The role of copepods in the planktonic ecosystems of mixed and stratified waters of the European shelf seas. Hydrobiologia 292/293:521-530

Zarauz L, Irigoien X, Urtizberea A, Gonzalez M (2007) Mapping plankton distribution in the Bay of Biscay during three consecutive spring surveys. Mar Ecol Prog Ser 345:27-39

Table 1: Average size and size-class classification used for mesozooplanktonic organisms identified and analysed in this study.

Size-class classification used in this study	Taxa	Average size (mm)[a]
Small organisms	Copepod nauplii	0.2
	Euterpina sp.	0.6
	Oithona sp.	0.7
	Oncaea sp.	0.7
	Coryceus sp.	0.9
	Appendicularia	0.9
	Evadne / *Podon* sp.	0.9
	Acartia sp.	1.0
	Small undetermined Calanoids including copepodites	1.1
Medium-sized organisms	*Temora* sp.	1.4
	Centropages sp.	1.5
	Medium undetermined Calanoids	1.9
Large organisms	*Calanus helgolandicus*	2.9
	Decapod larvae	3.5

[a] Average size corresponds to an average value of sizes (total length) reported for species included in the taxa (i.e., mostly genus) that may be found in the Bay of Biscay area and/or in the North-East Atlantic. Main references for the reported species: Plounevez and Champalbert 1999; Isla et al. 2004; Valdés et al. 2007; Cabal et al. 2008. Main reference for the size of species: Rose 1933.

Table 2: $\delta^{13}C$ and $\delta^{15}N$ values (Mean ± Standard Deviation in ‰) of the mesozooplanktonic prey groups defined by hierarchical cluster analysis and used in mixing models for the 3 periods studied (Spring 2010, Spring 2011 and Autumn 2011). The values presented here are corrected for the effects of preservation and/or delipidation, for consistency of treatment between prey and predators (see text). As groups are not strictly identical between the three periods investigated, they are numbered and presented in the default order for each hierarchical classification performed.

	Group	Average range of sizes (mm)	Species forming the group and associated stations	$\delta^{13}C$ Mean ± SD	$\delta^{15}N$ Mean ± SD
Spring 2010	1. Small to medium-sized organisms from the slope, north	0.7 – 1.9	*Oithona* sp. (Sl1) Medium und. Calanoid (Sl1, Sl2)	-20.3 ± 0.4	4.2 ± 0.6
	2. Medium-sized organisms from the coast to the shelf, central to north	1.4 – 1.9	*Temora* sp. (C2) Medium und. Calanoid (Sh3)	-19.4 ± 0.3	4.9 ± 0.3
	3. Small to medium-sized organisms from the slope, central to south	0.7 – 1.9	*Oithona* sp. (Sl3) Medium und. Calanoid (Sl3, Sl4)	-21.7 ± 0.3	4.3 ± 1.0
	4. Small to medium-sized organisms from the coast to the shelf, north	0.2 – 1.9	Copepod nauplii (C1) *Euterpina* sp. (C1) *Acartia* sp. (C2) *Temora* sp. (C1, Sh1) Medium und. Calanoid (C1, Sh1)	-19.8 ± 0.2	7.0 ± 0.6
	5. Large organisms from the shelf to the slope, north	2.9	*C. helgolandicus* (Sh1, Sh2, Sl1, Sl2)	-20.6 ± 0.7	7.1 ± 0.9
	6. Large organisms from the slope, central to south	2.9	*C. helgolandicus* (Sl3, Sl4, Sl5)	-22.2 ± 0.1	6.8 ± 0.4
	7. Small to medium-sized organisms from the coast to the slope, south	1.0 – 1.9	*Acartia* sp. (Sh5, Sl4, Sl5) *Temora* sp. (C4, Sh5) Medium und. Calanoid (Sh5)	-20.4 ± 0.7	6.2 ± 0.6
	8. Small to medium-sized organisms from the coast to the shelf, central to south	0.7 – 1.4	*Oithona* sp. (C3) *Oncaea* sp. (C3) *Evadne / Podon* sp. (Sh3) *Acartia* sp. (C3, C4) *Temora* sp. (C3)	-19.8 ± 0.5	7.2 ± 0.9

Table 2. Continued.

Spring 2011	1. Small to medium-sized organisms from the slope, north	1.1 – 1.9	Small und. Calanoid (Sl1) Medium und. Calanoid (Sl1)	-21.8 ± 0.4	4.6 ± 0.3
	2. Small to medium-sized organisms from the shelf to the slope, north	1.0 – 1.9	*Oithona* sp. (Sl1, Sl2) *Oncaea* sp. (Sh2) *Corycaeus* sp. (Sh1) *Centropages* sp. (Sl1, Sl2) Small und. Calanoid (Sl2) Medium und. Calanoid (Sh1, Sh2, Sl2)	-20.6 ± 0.5	5.8 ± 0.6
	3. Large organisms from the shelf to the slope, north	2.9	*C. helgolandicus* (Sh1, Sh2, Sl2)	-20.8 ± 0.7	6.9 ± 1.1
	4. Small to medium-sized organisms from the shelf to the slope, central to south	0.7 – 1.9	*Oithona* sp. (Sh3, Sh4, Sl3, Sl4, Sl5) *Oncaea* sp. (Sh3, Sh4) Small und. Calanoid (Sh3, Sh4, Sl4, Sl5) Medium und. Calanoid (Sl3, Sl4, Sl5)	-20.3 ± 0.4	5.6 ± 0.5
	5. Small to medium-sized organisms from the coast to the shelf, central	2.9	*Temora* sp. (C2, C3) Small und. Calanoid (C3) Medium und. Calanoid (C2, C3, C4, Sh3, Sh4)	-20.4 ± 0.4	6.2 ± 0.5
	6. Small to medium-sized organisms from the coast, north	1.0 – 1.9	*Acartia* sp. (C1) *Temora* sp. (C1) Medium und. Calanoid (C1)	-19.2 ± 0.4	8.9 ± 0.6
	7. Large organisms from the shelf to the slope, south	2.9	*C. helgolandicus* (Sh5, Sl5)	-19.0 ± 0.6	7.6 ± 0.7
	8. Small to medium-sized organisms from the coast to the shelf, south	0.7 – 1.9	*Oithona* sp. (C5) Appendicularia (C4, C5) *Acartia* sp. (C4, C5, Sh4, Sh5) *Temora* sp. (C5) Medium und. Calanoid (C5, Sh5)	-19.1 ± 0.3	7.2 ± 0.3
	9. Small to medium-sized organisms from the oceanic area	0.7 – 1.9	*Oithona* sp. (O4) Small und. Calanoid (O4) Medium und. Calanoid (O4)	-20.5 ± 0.5	4.9 ± 0.8

Table 2. Continued.

Autumn 2011	1. Large Decapod larvae from the shelf, north	3.5	Decapod larvae (Sh2)	-20.3 ± 0.2	2.4 ± 0.2
	2. Small to medium-sized organisms from the slope, north	0.7 – 1.9	*Oithona* sp. (Sl1) Small und. Calanoids (Sl1) Medium und. Calanoids (Sl1)	-20.5 ± 0.1	5.4 ± 0.9
	3. Small to medium-sized organisms from the shelf to the slope, central to north	0.7 – 1.9	*Oithona* sp. (Sh2, Sl3) Small und. Calanoids (Sl3) Medium und. Calanoids (Sh2, Sl3)	-20.5 ± 0.1	3.9 ± 0.4
	4. Small to medium-sized organisms from the coast to the shelf, north	0.7 – 1.9	*Oncaea* sp. (Sh1) *Temora* sp. (Sh1) Medium und. Calanoids (Sh1)	-20.9 ± 0.0	7.0 ± 0.4
	5. Small to medium-sized organisms from the coast to the shelf, central	0.7 – 1.9	*Oithona* sp. (Sh3) *Oncaea* sp. (C3, Sh3) *Temora* sp. (C3, Sh3) Medium und. Calanoids (Sh3)	-20.2 ± 0.2	6.2 ± 0.6
	6. Small to medium-sized organisms from the coast to the slope, south	0.7 – 1.9	*Oithona* sp. (Sl5) *Oncaea* sp. (C4, C5, Sh4) *Corycaeus* sp. (C4) Small und. Calanoids (Sl5) Medium und. Calanoids (C4, C5, Sh4, Sl5)	-20.3 ± 0.4	5.8 ± 0.7

Table 3: Results of statistical tests for significant differences between periods or between species for $\delta^{13}C$ and $\delta^{15}N$ values measured in muscle and/or liver of fishes. For the muscle tissue, within a species and for one element at time (i.e., carbon or nitrogen), vertical bars (|) indicate means that do not differ significantly following post hoc Tukey's tests in the case of ANOVA, or multiple comparison test with Holm's adjustment method in the case of Kruskal-Wallis (test chosen according to satisfying conditions for parametric statistics or not). For the liver tissue, analysed at two periods only, vertical bars (|) indicate means that do not differ significantly following the Student t tests or Mann-Whitney-Wilcoxon tests applied (according to satisfying conditions for parametric statistics or not). To assess significant between-species differences, Student t tests or Mann-Whitney-Wilcoxon tests were also performed and p-values are indicated for each period and for each tissue considered. *N* = number of individuals. $\delta^{13}C$ and $\delta^{15}N$ values (Mean ± Standard Deviation in ‰) for each group of fish are specifically represented in Fig. 5.

		$\delta^{13}C$							**$\delta^{15}N$**						
		Sardina pilchardus			***Engraulis encrasicolus***				***Sardina pilchardus***			***Engraulis encrasicolus***			
			Groups for non-significant difference between periods			**Groups for non-significant difference between periods**		**Significant difference between species**		**Groups for non-significant difference between periods**			**Groups for non-significant difference between periods**		**Significant difference between species**
Tissue	**Period**	***N***	**a**	**b**	***N***	**a**	**b**		***N***	**a**	**b**	***N***	**a**	**b**	
Muscle	Spring 2010	40	|		34	|		**p < 0.001**	40	|		34	|		**p < 0.001**
	Spring 2011	38	|		37	|		**p < 0.001**	38	|		37	|		**p < 0.001**
	Autumn 2011	31		|	33	|		**p < 0.001**	31	|		33		|	**p = 0.002**
Liver	Spring 2010	—			—				—			—			
	Spring 2011	38	|		37	|		**p < 0.001**	38	|		37	|		p = 0.696
	Autumn 2011	31		|	33	|		**p = 0.003**	31		|	33		|	p = 0.327

Table 4: Summary of estimated contributions (Mean values ± Standard Deviation in %) of mesozooplanktonic prey groups in the diet of European sardine *Sardina pilchardus* and European anchovy *Engraulis encrasicolus* resulting from the different mixing models applied with different Trophic Enrichment Factors (TEFs: $\Delta\delta^{13}C$ and $\Delta\delta^{15}N$) taken from the literature (i.e., sensitivity analysis). Values for groups of prey contributing on average to more than 10% in the diet of each species, when the four models performed by species and by tissue are considered, are in bold. For signification of each group of prey within each period, see Table 2.

	Model applied References for TEFs	**Model 1 Post 2002**	**Model 2 Sweeting et al. 2007ab**	**Model 3 Pinnegar and Polunin 1999**	**Model 4 Trueman et al. 2005**	**Mean ± SD**	**Model 1 Post 2002**	**Model 2 Sweeting et al. 2007ab**	**Model 3 Pinnegar and Polunin 1999**	**Model 4 Trueman et al. 2005**	**Mean ± SD**
	$\Delta\delta^{13}C$	0.4 ± 1.3	1.7 ± 1.1	2.5 ± 0.1	2.1 ± 0.1		0.4 ± 1.3	0.9 ± 1.3	1.2 ± 0.8	1.6 ± 0.3	
Period	**$\Delta\delta^{15}N$**	3.4 ± 1.0	3.2 ± 1.3	3.3 ± 0.2	2.3 ± 0.3		3.4 ± 1.0	2.3 ± 0.9	2.2 ± 0.2	0.0 ± 0.3	
						Sardina pilchardus (n = 40)					
	Prey group			**MUSCLE**					**LIVER**		
Spring 2010	1	1.2 ± 1.0	1.3 ± 1.2	1.1 ± 1.0	1.7 ± 1.6	1.3 ± 0.3	—	—	—	—	—
	2	2.1 ± 1.8	3.0 ± 2.7	1.3 ± 1.2	3.1 ± 3.0	2.4 ± 0.8	—	—	—	—	—
	3	0.9 ± 0.8	0.9 ± 0.8	1.1 ± 1.0	1.3 ± 1.2	1.1 ± 0.2	—	—	—	—	—
	4	34.5 ± 12.5	38.0 ± 9.3	16.4 ± 10.6	26.5 ± 12.7	**28.9 ± 9.6**	—	—	—	—	—
	5	10.3 ± 8.3	9.6 ± 5.9	28.9 ± 11.7	9.9 ± 6.5	**14.7 ± 9.5**	—	—	—	—	—
	6	2.1 ± 2.0	1.7 ± 1.4	10.7 ± 4.5	2.3 ± 1.8	4.2 ± 4.3	—	—	—	—	—
	7	3.3 ± 3.0	3.8 ± 3.2	3.7 ± 3.4	4.5 ± 4.1	3.8 ± 0.5	—	—	—	—	—
	8	45.6 ± 11.8	41.7 ± 8.3	36.8 ± 13.1	50.7 ± 12.8	**43.7 ± 5.9**	—	—	—	—	—
						Engraulis encrasicolus (n = 34)					
				MUSCLE					**LIVER**		
	1	4.0 ± 3.5	5.5 ± 3.8	5.2 ± 4.0	1.4 ± 1.3	4.0 ± 1.9	—	—	—	—	—
	2	26.0 ± 7.2	12.5 ± 5.3	4.9 ± 4.0	1.7 ± 1.6	**11.3 ± 10.8**	—	—	—	—	—
	3	1.7 ± 1.6	3.4 ± 2.6	8.2 ± 5.2	1.5 ± 1.4	3.7 ± 3.1	—	—	—	—	—
	4	27.9 ± 11.4	23.1 ± 6.7	10.7 ± 7.7	15.6 ± 10.3	**19.3 ± 7.7**	—	—	—	—	—
	5	6.0 ± 5.4	15.1 ± 6.2	19.3 ± 11.1	30.1 ± 13.3	**17.6 ± 10.0**	—	—	—	—	—
	6	1.7 ± 1.7	6.4 ± 3.7	28.5 ± 6.2	15.9 ± 5.3	**13.1 ± 11.8**	—	—	—	—	—
	7	6.0 ± 5.4	12.0 ± 6.2	11.6 ± 8.5	4.8 ± 4.6	8.6 ± 3.7	—	—	—	—	—
	8	26.7 ± 10.2	22.0 ± 6.2	11.6 ± 7.8	29.0 ± 12.5	**22.3 ± 7.7**	—	—	—	—	—

Table 4. Continued.

					Sardina pilchardus (n = 38)					
	Prey group			**MUSCLE**				**LIVER**		
Spring 2011	1	1.5 ± 1.4	2.1 ± 1.7	11.0 ± 4.6	2.3 ± 2.2	4.2 ± 4.5	6.0 ± 5.1	6.2 ± 4.2	9.1 ± 5.8	3.0 ± 5.2	6.1 ± 2.5
	2	3.4 ± 2.9	5.0 ± 3.3	3.6 ± 3.2	1.9 ± 1.8	3.5 ± 1.3	7.7 ± 6.1	9.9 ± 5.9	9.9 ± 7.1	2.3 ± 3.4	7.5 ± 3.6
	3	3.3 ± 2.7	8.6 ± 4.0	32.7 ± 5.7	25.0 ± 5.0	**17.4 ± 13.8**	2.2 ± 2.0	8.4 ± 5.1	19.9 ± 8.3	11.0 ± 16.7	**10.4 ± 7.3**
	4	4.2 ± 3.2	4.8 ± 3.2	1.9 ± 1.7	1.3 ± 1.3	3.1 ± 1.7	19.8 ± 9.5	11.0 ± 6.0	7.8 ± 6.0	1.7 ± 2.3	**10.1 ± 7.5**
	5	4.3 ± 3.4	7.3 ± 4.2	4.4 ± 3.7	2.2 ± 2.2	4.6 ± 2.1	6.2 ± 5.1	10.5 ± 6.1	10.3 ± 7.3	2.7 ± 3.7	7.4 ± 3.7
	6	27.2 ± 5.2	31.6 ± 4.0	41.3 ± 4.6	63.1 ± 5.5	**40.8 ± 16.0**	2.0 ± 1.8	14.6 ± 4.8	18.0 ± 6.3	73.0 ± 27.1	**26.9 ± 31.5**
	7	25.4 ± 7.0	19.2 ± 4.8	2.0 ± 1.8	1.7 ± 1.6	**12.1 ± 12.1**	8.1 ± 5.6	14.5 ± 5.8	10.2 ± 6.7	2.7 ± 2.6	8.9 ± 4.9
	8	27.7 ± 7.7	18.5 ± 5.2	1.7 ± 1.5	1.5 ± 1.5	**12.4 ± 13.0**	14.4 ± 7.5	15.1 ± 6.0	8.5 ± 6.0	2.2 ± 2.3	**10.1 ± 6.0**
	9	3.0 ± 2.4	2.9 ± 2.2	1.4 ± 1.3	1.0 ± 0.9	2.1 ± 1.0	33.6 ± 7.7	9.8 ± 5.4	6.3 ± 5.0	1.4 ± 1.9	**12.8 ± 14.3**
					Engraulis encrasicolus (n = 37)					
				MUSCLE				**LIVER**		
	1	2.5 ± 2.3	12.8 ± 5.8	24.9 ± 7.0	6.2 ± 5.1	**11.6 ± 9.8**	9.6 ± 6.5	15.8 ± 7.6	15.1 ± 7.9	4.0 ± 7.2	**11.1 ± 5.5**
	2	4.9 ± 4.5	10.9 ± 6.6	5.7 ± 5.1	4.0 ± 3.7	6.4 ± 3.1	9.8 ± 7.4	10.0 ± 7.4	7.8 ± 6.4	3.0 ± 3.5	7.7 ± 3.3
	3	2.2 ± 2.1	15.4 ± 6.4	46.6 ± 10.9	54.3 ± 12.0	**29.6 ± 24.8**	3.8 ± 3.3	25.3 ± 10.6	40.4 ± 14.5	11.4 ± 13.1	**20.2 ± 16.1**
	4	10.2 ± 7.5	9.1 ± 6.1	3.5 ± 3.1	2.6 ± 2.4	6.4 ± 3.9	17.3 ± 10.8	6.8 ± 5.6	5.0 ± 4.4	2.3 ± 2.5	7.9 ± 6.6
	5	4.9 ± 4.4	11.2 ± 6.7	5.7 ± 5.2	4.5 ± 4.0	6.6 ± 3.1	8.1 ± 6.6	10.6 ± 7.7	8.1 ± 6.6	3.4 ± 3.6	7.6 ± 3.0
	6	3.5 ± 3.2	14.7 ± 5.6	5.8 ± 4.6	20.9 ± 7.6	**11.2 ± 8.1**	2.8 ± 2.4	14.2 ± 6.7	11.3 ± 7.0	67.1 ± 22.5	**23.9 ± 29.2**
	7	23.4 ± 11.6	9.2 ± 5.8	2.5 ± 2.2	2.9 ± 2.7	9.5 ± 9.8	6.5 ± 4.8	6.1 ± 5.0	4.2 ± 3.8	3.9 ± 3.7	5.2 ± 1.3
	8	33.2 ± 13.1	8.6 ± 5.7	2.3 ± 2.2	2.5 ± 2.3	**11.7 ± 14.7**	8.7 ± 6.1	5.5 ± 4.6	3.9 ± 3.5	3.0 ± 2.9	5.3 ± 2.5
	9	15.2 ± 7.6	8.1 ± 5.6	3.0 ± 2.8	2.1 ± 1.9	7.1 ± 6.0	33.4 ± 10.6	5.7 ± 4.8	4.2 ± 3.8	1.9 ± 2.0	**11.3 ± 14.8**

Table 4. Continued.

					Sardina pilchardus (n = 31)					
	Prey group			**MUSCLE**				**LIVER**		
Autumn 2011	1	1.1 ± 1.1	2.1 ± 2.0	2.6 ± 2.4	2.7 ± 2.7	2.1 ± 0.7	1.7 ± 1.5	1.6 ± 1.6	1.3 ± 1.2	2.4 ± 2.3	1.8 ± 0.5
	2	3.7 ± 3.6	7.2 ± 6.4	6.9 ± 6.1	5.7 ± 5.4	5.9 ± 1.6	6.6 ± 5.6	7.2 ± 6.4	4.5 ± 4.4	6.8 ± 6.4	6.3 ± 1.2
	3	1.7 ± 1.6	2.9 ± 2.8	4.1 ± 3.8	3.1 ± 3.0	3.0 ± 1.0	2.6 ± 2.3	2.6 ± 2.5	2.0 ± 1.9	3.7 ± 3.7	2.7 ± 0.7
	4	73.2 ± 15.7	17.3 ± 11.8	73.5 ± 10.4	4.7 ± 4.2	**42.2 ± 36.4**	28.4 ± 11.1	45.8 ± 14.5	67.3 ± 12.1	73.4 ± 11.1	**53.7 ± 20.6**
	5	14.1 ± 13.0	50.9 ± 12.5	6.5 ± 6.0	65.8 ± 15.9	**34.3 ± 28.6**	43.2 ± 13.1	28.5 ± 11.4	17.4 ± 10.9	7.0 ± 6.2	**24.0 ± 15.5**
	6	6.2 ± 6.2	19.6 ± 12.9	6.4 ± 5.7	18.0 ± 14.3	**12.6 ± 7.2**	17.5 ± 11.1	14.3 ± 9.7	7.5 ± 6.7	6.7 ± 6.1	**11.5 ± 5.3**
					Engraulis encrasicolus (n = 33)					
				MUSCLE				**LIVER**		
	1	3.2 ± 3.0	3.1 ± 2.9	2.6 ± 2.4	2.5 ± 2.4	2.9 ± 0.4	5.2 ± 4.3	3.2 ± 2.9	2.3 ± 2.1	2.1 ± 2.0	3.2 ± 1.4
	2	10.4 ± 8.8	11.3 ± 9.2	6.9 ± 6.1	8.0 ± 7.2	9.2 ± 2.0	15.8 ± 9.1	14.6 ± 8.7	7.1 ± 5.9	5.1 ± 4.9	**10.7 ± 5.3**
	3	4.6 ± 4.3	4.8 ± 4.5	4.1 ± 3.8	4.0 ± 3.8	4.4 ± 0.4	7.8 ± 6.1	5.9 ± 5.0	3.6 ± 3.3	3.3 ± 3.3	5.2 ± 2.1
	4	18.6 ± 12.7	33.1 ± 12.3	73.5 ± 10.4	63.1 ± 13.5	**47.1 ± 25.6**	20.1 ± 8.6	38.8 ± 9.3	72.8 ± 8.0	80.1 ± 9.3	**53.0 ± 28.3**
	5	42.5 ± 18.0	29.0 ± 13.1	6.5 ± 6.0	12.4 ± 9.5	**22.6 ± 16.3**	28.6 ± 9.3	21.2 ± 9.3	7.6 ± 6.0	4.7 ± 4.6	**15.5 ± 11.3**
	6	20.7 ± 13.3	18.7 ± 12.0	6.4 ± 5.7	10.0 ± 8.4	**14.0 ± 6.8**	22.5 ± 9.4	16.3 ± 8.8	6.6 ± 5.4	4.7 ± 4.7	**12.5 ± 8.4**

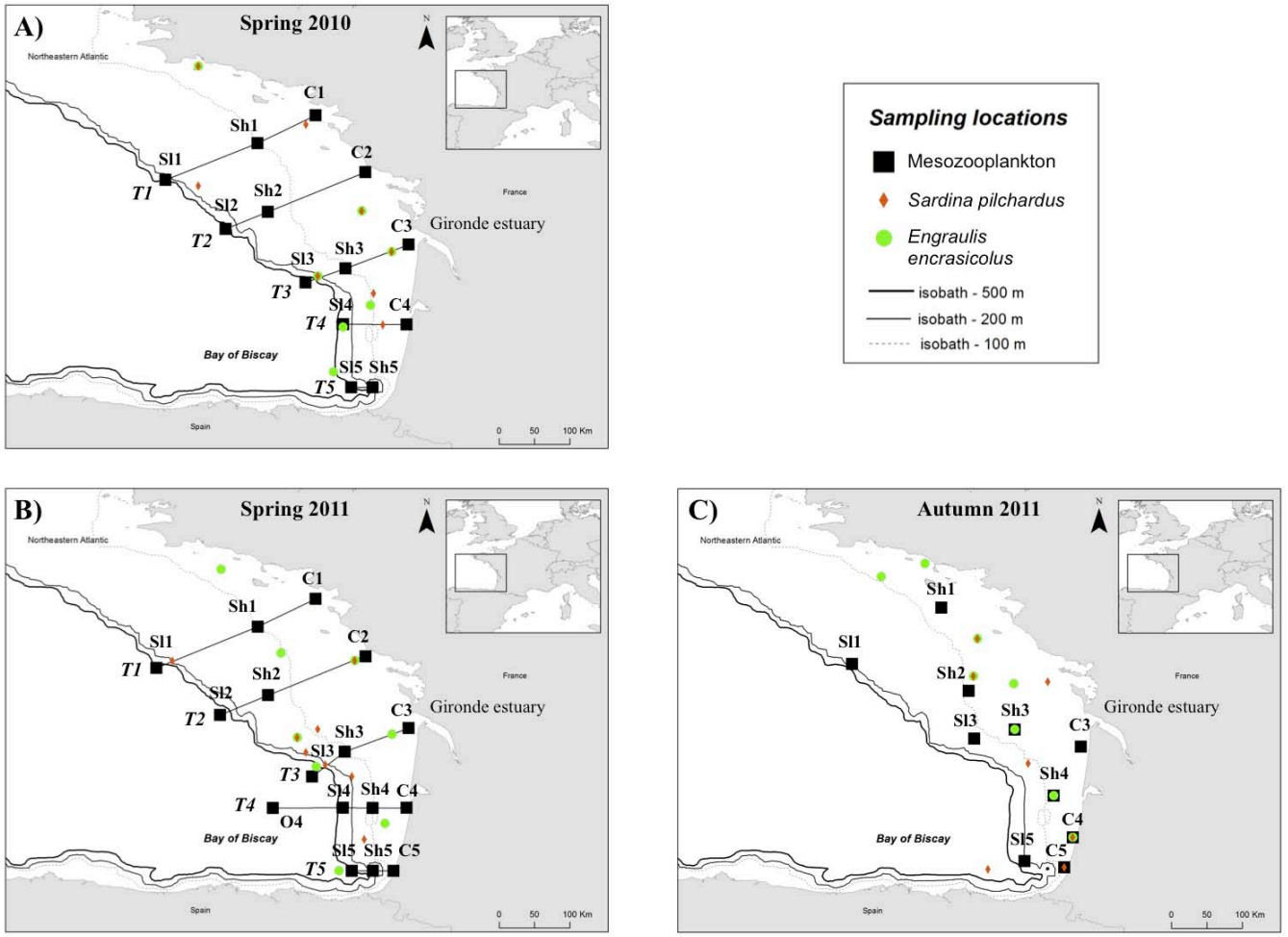

Fig. 1: Maps of the study area (Bay of Biscay), indicating the transects realised from the coastline to the slope and the stations selected for plankton sampling for the three periods considered in this study: A) Spring 2010, B) Spring 2011 and C) Autumn 2011. Trawls for fish sampling are also indicated. T = Transect; C = Coast; Sh = Shelf; Sl = Slope; O = Oceanic. The study area represented (outlines of the box) extends from 0° to 8°W and from 43°N to 48°N (Source: Etopo1 NOAA – LIENSs – 2013. Design and realization: Cellule Géomatique LIENSs – UMR 7266).

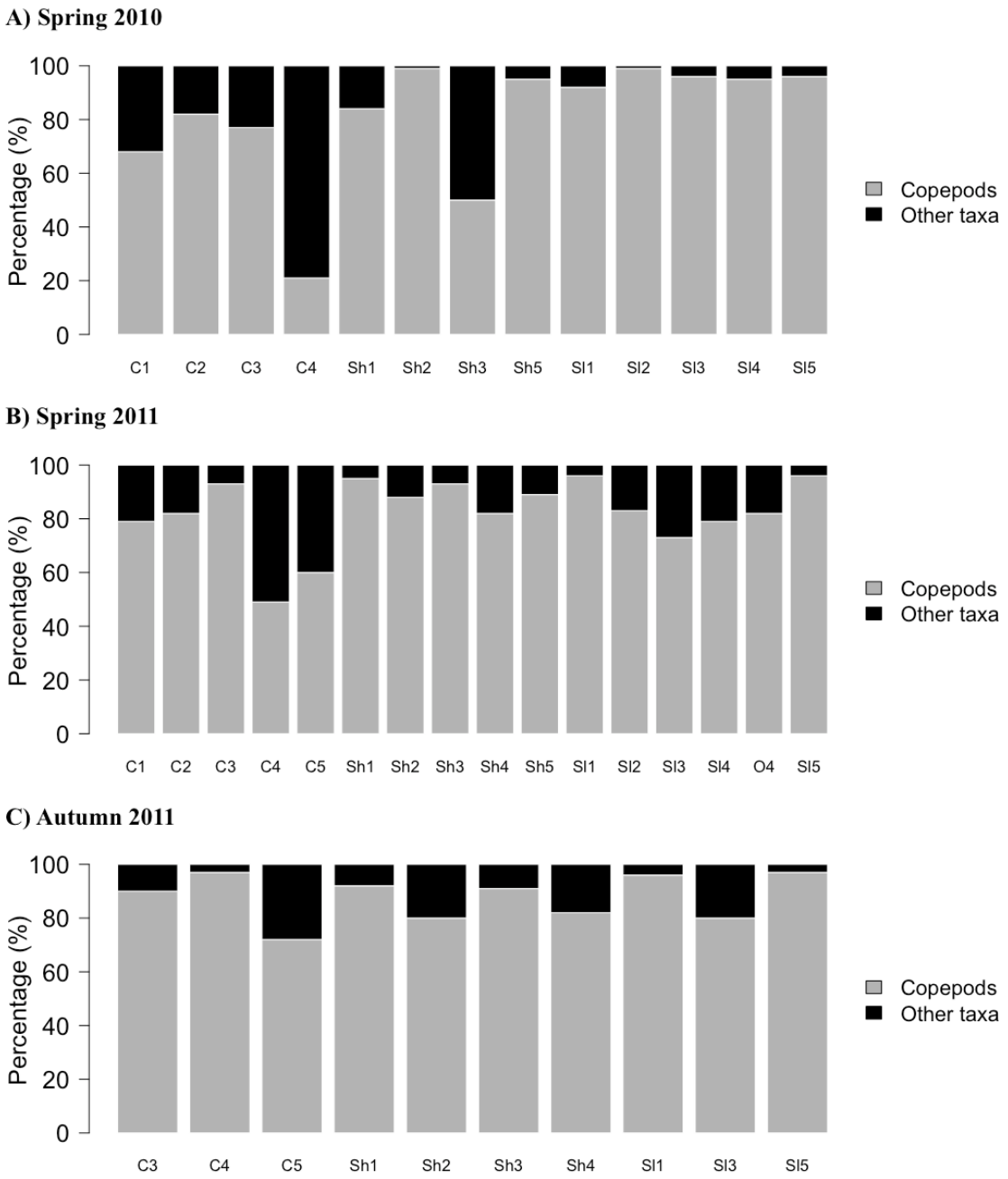

Fig. 2: Histograms presenting the percentage of copepods and of other taxa (relative to the total abundance of mesozooplanktonic organisms, in number of individuals. m^{-3}) for each station selected and considered for taxonomic identification in A) Spring 2010, B) Spring 2011 and C) Autumn 2011. C = Coast; Sh = Shelf; Sl = Slope; O = Oceanic; see Fig. 1 for the corresponding location of each station.

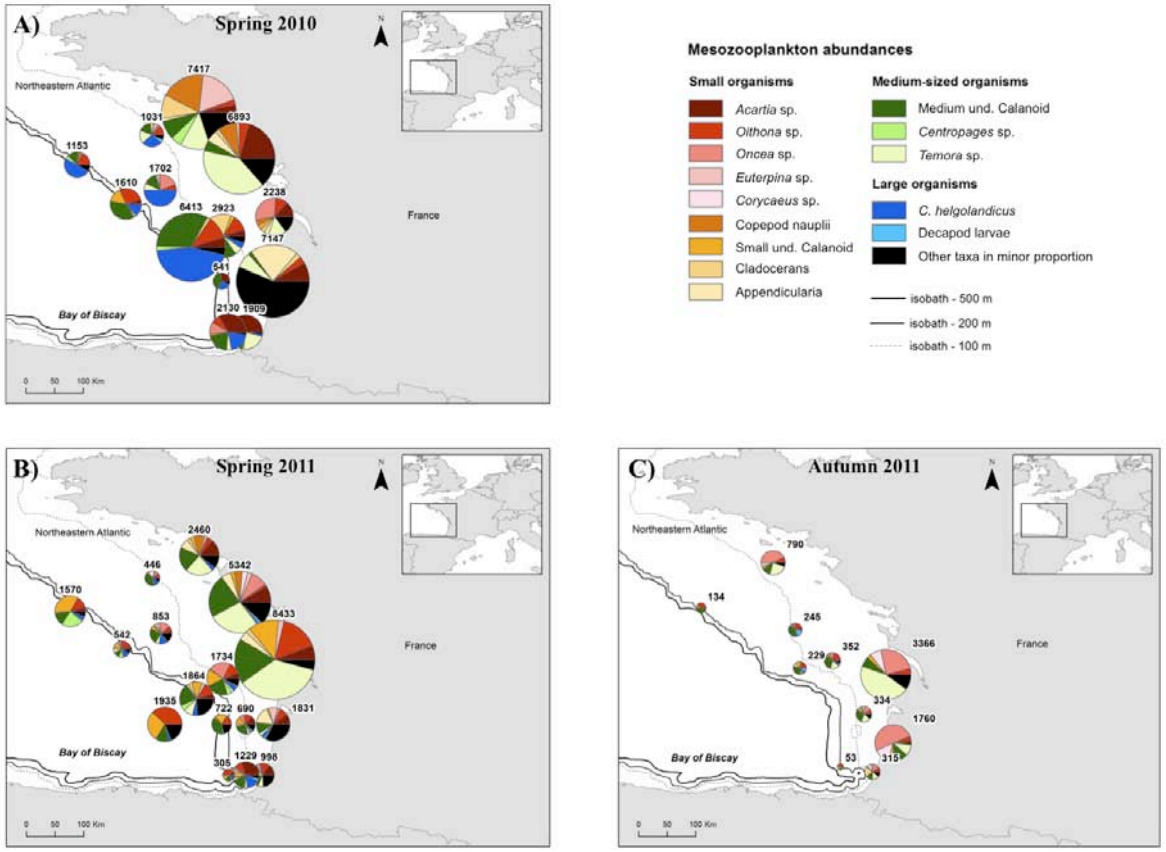

Fig. 3: Results of the taxonomic identification performed on mesozooplankton samples collected in A) Spring 2010, B) Spring 2011 and C) Autumn 2011. The number above a pie corresponds to the total abundance of organisms within a station (in number of individuals. m^{-3}), and the size of pies is proportional to the total abundance. Among the 'dominant taxa' (see text), small organisms are represented in shades of red to yellow, medium-sized organisms in shades of green, and large organisms in shades of blue (see Table 1). Within each station, the proportion of taxa in minor proportion (i.e., with an abundance < 5% when taken individually, and therefore not analysed for stable isotope ratios – see text) is indicated in black. The study area represented (outlines of the box) extends from 0° to 8°W and from 43°N to 48°N (Source: Etopo1 NOAA – LIENSs – 2013. Design and realization: Cellule Géomatique LIENSs – UMR 7266).

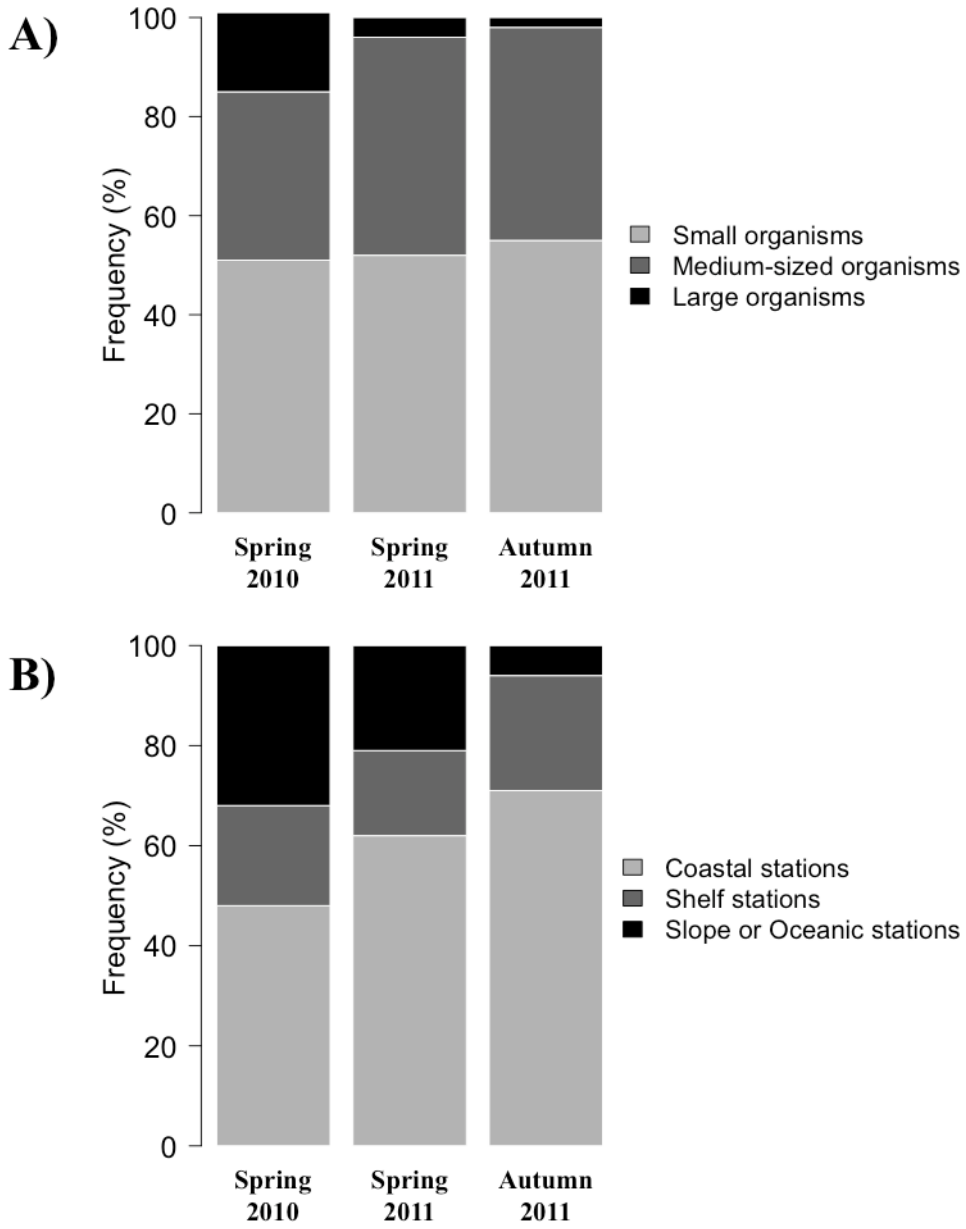

Fig. 4: Stacked bar charts presenting A) the frequency of identified organisms belonging to small, medium-sized or large organisms within each of the three periods considered in this study, all stations considered together over the Bay of Biscay; B) the frequency of organisms identified in coastal stations, stations from the shelf or stations from the slope and oceanic stations within each of the three periods considered in this study, all sizes considered together. Only organisms belonging to 'dominant taxa' were taken into account for both histograms (see text).

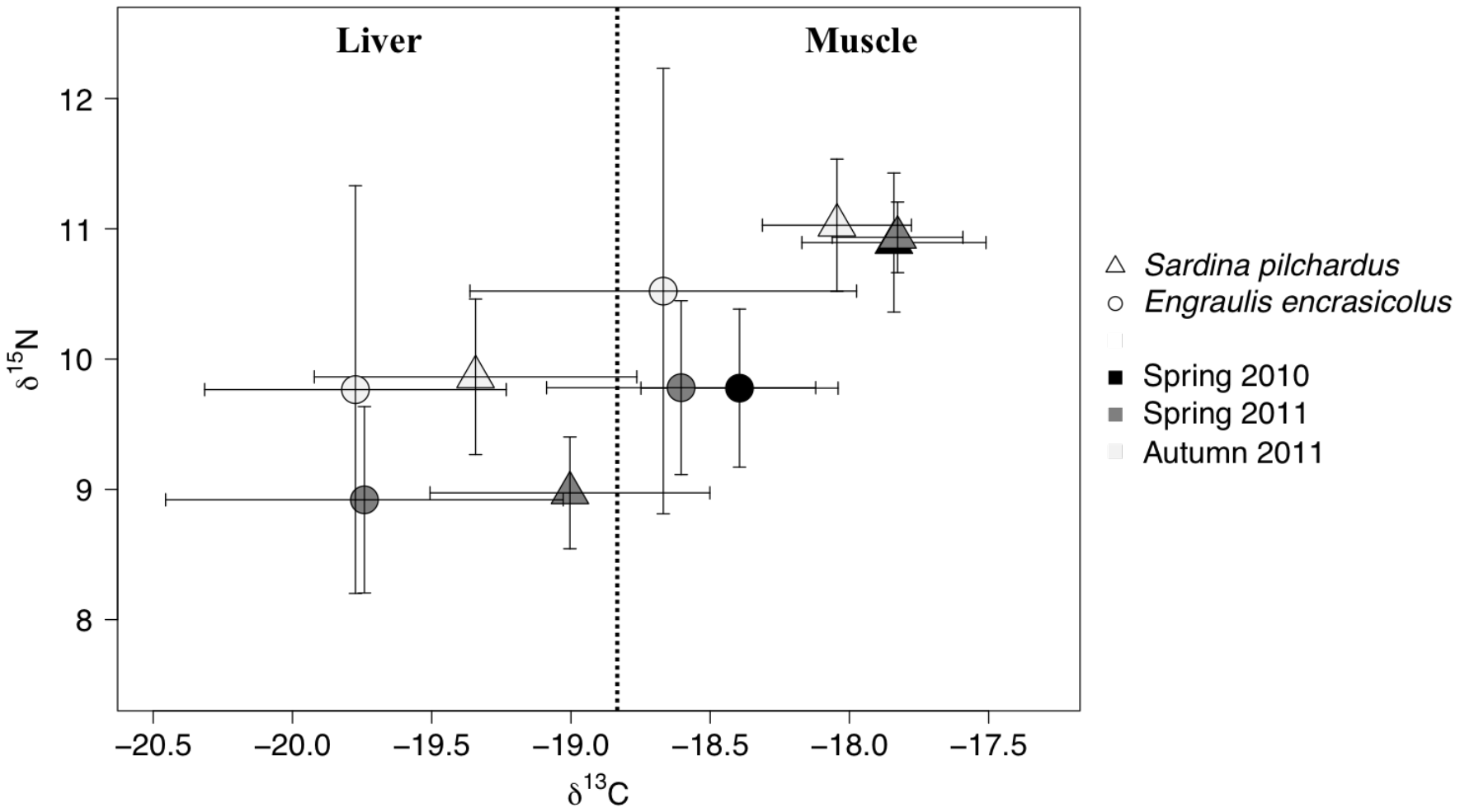

Fig. 5: $\delta^{13}C$ and $\delta^{15}N$ values (mean ± SD, in ‰) for European sardine *Sardina pilchardus* and European anchovy *Engraulis encrasicolus*, depending on the period considered and on the tissue analysed. Detailed results of statistical tests for significant difference between species or between periods for a given tissue or for a given element are presented in Table 3. The dotted line is only drawn as a guide to visually distinguish muscle and liver tissue values that are presented here on the same figure.