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2 **Oceanographic structure and seasonal variation**
3 **contribute to high heterogeneity in mesozooplankton**
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5

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19

20 **Abstract**

21 The coastal oceans can be highly variable, especially near ocean fronts. The Ushant Front is the
22 dominant oceanographic feature in the Iroise Sea (NE Atlantic) during summer, separating warm

23 stratified offshore waters from cool vertically-mixed nearshore waters. Mesozooplankton
24 community structure was investigated over an annual cycle to examine relationships with
25 oceanographic conditions. DNA metabarcoding of COI and 18S genes was used in communities
26 from six sites along two cross-shelf transects. Taxonomic assignments of 380 and 296 OTUs (COI
27 and 18S respectively) identified 21 classes across 13 phyla. Meroplankton relative abundances
28 peaked in spring and summer, particularly for polychaete and decapod larvae respectively,
29 corresponding to the reproductive periods of these taxa. Meroplankton was most affected by
30 season, while holoplankton varied most by shelf position. Copepods with a mixed feeding strategy
31 were associated with the most offshore sites, especially in the presence of the front, while filter-
32 feeding or carnivorous copepods were associated with nearshore sites. In sum, mesozooplankton
33 communities in well-mixed coastal waters were distinct from those found in the Ushant Front (high
34 thermal stratification and chlorophyll-a). Furthermore, the benthic compartment, through its partial
35 life cycle in the water column, contributed to high heterogeneity in planktonic communities over
36 short temporal and spatial scales.

37

38 **Keywords:** Mesozooplankton, mitochondrial COI, 18S rRNA, DNA metabarcoding, High-
39 throughput sequencing, Marine Protected Area, Ocean front

40

41 **Introduction**

42 The coastal oceans can be highly variable, influenced not only by major oceanographic
43 regimes, but also by processes related to bordering landmasses, such as river discharge, runoff,
44 tidal forcing and currents influenced by the coastline and shallow water columns. Nearshore
45 conditions can therefore be highly contrasted to adjacent offshore water masses, with the most

46 striking examples being near ocean fronts. Ocean fronts occur where two distinct water masses
47 come into contact, and are among the main oceanographic structures affecting the distribution of
48 marine organisms as diverse as pelagic crustaceans, fish, marine mammals and birds (Acha *et al.*,
49 2015). They can arise from a diversity of processes and are defined by strong horizontal gradients
50 of salinity and/or temperature. These hydrological structures create vertical fluxes, leading to
51 aggregation processes, as well as increases in downward export or nutrient availability (Acha *et*
52 *al.*, 2015). Ocean fronts have been shown to play an important role on plankton distribution, by
53 aggregating, transporting, or separating specific assemblages (Marra *et al.*, 1990; Flint *et al.*, 2002;
54 Ohman *et al.*, 2012). In particular, the fronts formed in the transition between coastal bays and
55 their oceanic adjacent regions affect the distribution of meroplankton (i.e. mainly composed of
56 pelagic larvae of invertebrates) (Shanks *et al.*, 2003; Ayata *et al.*, 2011; Brandão *et al.*, 2020).
57 Consequently, fronts may play a role in the dispersal of species with a benthopelagic life cycle,
58 potentially altering their broad-scale connectivity and distribution along the coasts (Acha *et al.*,
59 2015). Furthermore, because they affect ecosystem functions such as primary productivity,
60 biogeochemical cycling and biomass production (Woodson and Litvin, 2015), ocean fronts also
61 affect ecosystem services (Martinetto *et al.*, 2020), particularly fisheries. As such, the influence of
62 ocean fronts on coastal communities merits adequate investigations.

63 The Iroise Sea, where the northeastern Atlantic transitions into the English Channel,
64 constitutes an interesting environment for examining the effects of ocean fronts on pelagic
65 communities. This part of the coast of western Brittany (France) is characterized by a megatidal
66 regime (5 - 10 m tidal amplitudes), which generates strong tidal currents. Tidal friction produces
67 enough turbulence to constantly mix the entire water column in nearshore areas, but as depth
68 increases, tidal mixing is not sufficient to erode the seasonal thermocline (Brumer *et al.*, 2020).

69 The result is a tidally influenced thermal front called the Ushant Front that is characterized by a
70 strong temperature gradient that separates warm stratified offshore waters from cool vertically
71 mixed nearshore waters (Le Boyer *et al.*, 2009; Chevallier *et al.*, 2014; Cadier *et al.*, 2017a). Where
72 nearshore and offshore water masses meet, a frontal zone is generated with specific characteristics
73 between both water masses, such as thermal stratification coupled with high chlorophyll-a near the
74 thermocline, which results from either subduction of productive coastal upper layers or nutrient
75 enrichment due to mixing of the two water masses (or both). The Ushant Front is the dominant
76 feature of the summer oceanographic structure of the Iroise Sea, which usually occurs from
77 April/May to October every year (Chevallier *et al.*, 2014; Cadier *et al.*, 2017a). This front is known
78 to increase primary production (Videau, 1987) but also to influence the size and abundance of
79 phytoplankton (Pingree *et al.*, 1975; Landeira *et al.*, 2014), with longer diatom chains during the
80 frontal establishment (Landeira *et al.*, 2014). This coastal front also acts as an ecological boundary
81 for free-living bacteria, with photosynthetic bacteria being most abundant offshore of the front,
82 oligotrophic bacteria being more abundant in water masses with low phytoplankton and high
83 inorganic nutrients, and opportunistic copiotrophic bacteria being most abundant in the most
84 productive period of the year, associated with the front (Lemonnier *et al.*, 2020). In addition, the
85 mesozooplankton community showed its highest diversity at the front as a result of the co-
86 occurrence of species from stratified and mixed waters (Schultes *et al.*, 2013).

87 The Ushant Front occurs within the boundaries of the Iroise Marine Natural Park (*Parc*
88 *Naturel Marin d'Iroise*, PNMI), the oldest marine protected area (MPA) in France. Due to its
89 influence on primary productivity and planktonic communities the front may affect the stocks of
90 planktivorous fish, in particular sardines and anchovies, which are important fisheries in the Iroise

91 Sea. To better understand these potential effects, the PNMI has established a monitoring program
92 that has been in place for 10 years.

93 DNA metabarcoding, which combines high-throughput sequencing technologies with
94 DNA barcoding, is revolutionizing how biodiversity is assessed in the marine environment (Baird
95 and Hajibabaei, 2012; Cristescu, 2014), by allowing DNA taken from environmental (e.g. water,
96 sediment) or bulk (e.g. plankton) samples to be sequenced concurrently, followed by taxonomic
97 assignment by comparison to a reference sequence database, such as the Barcode of Life Data
98 Systems for COI (Ratnasingham and Hebert, 2007). Thus, species diversity can be recovered
99 across many broad taxonomic groups (Valentini *et al.*, 2016). In order to capture a representative
100 sample of community biodiversity, multiple loci are often used in metabarcoding due to their
101 complementarity, as there is a trade-off between the taxonomic resolution and the taxonomic range
102 recovered (Carroll *et al.*, 2019; Couton *et al.*, 2019). Metabarcoding has been broadly applied for
103 benthic monitoring of marine ecosystems (Fonseca *et al.*, 2010; Lejzerowicz *et al.*, 2015; Cowart
104 *et al.*, 2020), and a growing number of studies are now using DNA metabarcoding to examine
105 zooplankton community dynamics (Chain *et al.*, 2016; Stefanni *et al.*, 2018; Carroll *et al.*, 2019;
106 Couton *et al.*, 2019; Questel *et al.*, 2021). Although metabarcoding is considered semi-quantitative
107 due to methodological biases (Clarke *et al.*, 2017; Carroll *et al.*, 2019), other studies have recently
108 revealed significant positive correlations between total abundance counts from morphological
109 taxonomic identification and metabarcoding sequence numbers (Bucklin *et al.*, 2019; Schroeder
110 *et al.*, 2020), thus indicating that relative abundances of reads can be considered as a proxy of
111 relative abundances of taxa. Furthermore, metabarcoding applied to a time-series of meroplankton
112 showed variations in read abundances that corresponded well with expectations based on the
113 reproductive season of the identified species (Couton *et al.*, 2019). In addition, despite the potential

114 biases associated with metabarcoding, the method does present various advantages, such as the
115 greater power for taxonomic identification for some groups, including undescribed or cryptic taxa,
116 reducing time-consuming counts under the microscope, and the ability to recover information for
117 a broad range of organisms (Lindeque *et al.*, 2013; Clarke *et al.*, 2017; Deagle *et al.*, 2018).

118 In this study, we investigated the composition of the mesozooplankton in the Iroise Sea
119 and the potential role of an ocean front in shaping this community over the course of the year. We
120 were particularly interested in the two contrasting groups of the mesozooplankton: meroplankton
121 (partial life-cycle in the water column) and holoplankton (full life-cycle in the water column). Our
122 specific goals were (i) to investigate the spatial and temporal distribution of mero- and
123 holoplankton in the Iroise Sea using DNA metabarcoding of the mitochondrial COI and the nuclear
124 ribosomal 18S genes, and (ii) to examine the environmental variables, including the ocean front
125 characteristics, that best explain differences observed in the mero- and holoplankton community
126 composition. Our central hypothesis is that the establishment of the Ushant Front in summer has
127 an impact on the structure of the mesozooplankton community in the Iroise Sea, increasing its
128 diversity.

129

130 **Materials and Methods**

131

132 **Sample collection**

133 Mesozooplankton sampling took place in the Iroise Marine Natural Park (*Parc Naturel*
134 *Marin d'Iroise*, PNMI), a marine protected area (MPA) that covers 3,550 km² of the Iroise Sea.
135 Sampling in the MPA was achieved on board the Valbelle (PM 509) and Augustine (PM 510)
136 PNMI vessels, and was part of a regular monitoring program, the PNMIR, which started in 2010.

137 The PNMIR consists in sampling along two parallel cross-shelf transects, in front of the bays of
138 Brest (seven stations) and Douarnenez (six stations). For the present study, zooplankton was
139 sampled in six stations, three in each transect, in spring (23-05-2019), summer (11-07-2019) and
140 fall (22-10-2019 to 24-10-2019) (Fig. 1a). Previous work has shown that the Ushant front usually
141 occurs in summer and fall (June – September). In order to examine the potential effect of frontal
142 conditions, sampling dates were chosen to capture conditions before, during and after the
143 establishment of the Ushant Front. Vertical tows were performed from 5 m above the bottom to
144 the surface with a WP2 plankton net with a 200- μ m mesh size (UNESCO, 1968), equipped with a
145 flowmeter (KC Denmark). Plankton was then sieved (200 μ m) in order to remove seawater, and
146 preserved onboard in a guanidinium thiocyanate buffer, not exceeding the volume of 100 ml.

147 To describe the abiotic conditions associated with each plankton sample, vertical profiles
148 of temperature, salinity, fluorescence and turbidity were performed using a CTD probe (Seabird
149 SBE19) coupled to a fluorimeter (NKE Mpx). Data from additional stations (four in the transect
150 of Bay of Brest and three in the transect of Bay of Douarnenez; Fig. 1b-d) followed in the PNMIR
151 monitoring program, taken on the same day of sampling, were included to better characterize the
152 water column. An index of the thermal stratification (ΔT) was calculated as the difference between
153 the surface and the bottom temperatures. A value higher than 1.5°C was considered as
154 characterizing a stratified water column (Schultes *et al.*, 2013).

155

156 **DNA extraction, PCR amplification and sequencing**

157 DNA was preserved in a guanidine thiocyanate buffer which effectively lysed the plankton
158 samples at room temperature over the course of one week, thereby eliminating the need for filtering
159 samples post-collection. Two replicate DNA extractions were performed using 20 mL of each

160 plankton sample following homogenization by inversion, with a 2:1 volume of
161 phenol:chloroform:isoamyl alcohol and 1:1 of an extraction buffer, followed by precipitation with
162 2:1 volume of isopropanol, 2 washes with 70% ethanol, resuspension in Milli-Q water and
163 treatment with RNase A (10 mg/ml; Sigma-Aldrich, St. Louis, MO, USA) (Fukami *et al.*, 2004).

164 The primers mlCOIintF (5'-GGWACWGGWTGAACWGTWTAYCCYCC-3') (Leray *et*
165 *al.*, 2013) and jgHCO2198 (5'-TAIACYTCIGGRTGICCRAARAAYCA-3') (Geller *et al.*, 2013)
166 were used to amplify a 313-bp portion of the highly variable mitochondrial Cytochrome c Oxidase
167 subunit I (COI) gene. In addition, the primers SSU_F04 (5'-GCTTGTCTCAAAGATTAAGCC-
168 3') (Fonseca *et al.*, 2010) and SSU_R22mod (5'-CCTGCTGCCTTCCTTRGA-3') (Sinniger *et al.*,
169 2016) were used to amplify a ca. 450-bp portion of the V1–V2 regions of the nuclear small subunit
170 rDNA (18S rDNA). Primers including Illumina sequencing adaptors are detailed in Table S1.

171 For each marker, four PCR reactions (20 µl final volume) were run for each of the two
172 DNA extractions, for a total of eight PCRs per biological sample, containing 1µl of total DNA
173 template, 1µl of each primer, 0.5µl of BSA (New England Biolabs), 6.5µl of ultrapure water, and
174 10µl of Q5 High-Fidelity 2X Master Mix (New England Biolabs). For COI, due to inosine content
175 in the reverse primer, the Q5U Hot Start High-Fidelity DNA Polymerase (New England Biolabs)
176 was used instead. Cycling conditions for COI started with a 2-min denaturation step followed by
177 16 initial cycles of 98°C for 10s, 30s at 62°C (-1°C per cycle), 72°C for 60s, followed by 25 cycles
178 at 46°C annealing temperature, and a final extension of 72°C for 2 min (modified from Leray *et*
179 *al.*, 2013). For 18S, cycling included a 2-min denaturation step followed by 30 cycles of 98°C for
180 10s, 30s at 57°C, 72°C for 30s, and a final extension of 72°C for 2 min (modified from Fonseca *et*
181 *al.*, 2010). The positive amplifications were identified on a 1.5% agarose gel stained with GelRed
182 (Biotium). PCR products from the eight replicates per biological sample were pooled at equal

183 volumes. Then a second PCR was done to add the Illumina index. These PCR products were
184 quantified using an Epoch spectrophotometer (BioTek Instruments) and pooled at equimolar
185 amounts. Paired-end reads (2×250 bp) were generated on a MiSeq (Illumina) at the Get-PlaGe
186 Genomics platform (Toulouse, France).

187

188 **Read processing and taxonomic assignment**

189 Resulting raw sequence reads were processed with the FROGS v.2.0 pipeline (Escudié *et*
190 *al.*, 2018), implemented on a Galaxy interface (Goecks *et al.*, 2010). First, paired-end reads were
191 merged into contigs with the FLASH algorithm (Magoč and Salzberg, 2011) before filtering by
192 length (minimum 250 bp for COI, and 350 bp for 18S). Chimera and singleton were filtered using
193 the VSEARCH tool (Rognes *et al.*, 2016) in each sample, and the remaining sequences underwent
194 SWARM v3.0 clustering, using the default parameters (Mahé *et al.*, 2014). SWARM uses a
195 clustering algorithm with a threshold corresponding to the maximum number of differences
196 between two sequences in an OTU (Mahé *et al.*, 2014).

197 Each representative OTU sequence was aligned and assigned to a taxon using BLAST,
198 with the following reference databases: BOLD_COI (<https://www.boldsystems.org/>) and Silva
199 18S rRNA v132 (<https://www.arb-silva.de/>) for COI and 18S, respectively. Potential false-positive
200 OTUs were removed by filtering OTUs representing less than 0.005% of the OTU reads (Bokulich
201 *et al.*, 2013).

202 In addition, we assigned trophic regime traits for the copepods, due to their high relative
203 abundance and important role in the pelagic community. Traits respective to trophic regime
204 (omnivore-herbivore, omnivore-detritivore, omnivore, carnivore), and feeding strategy (filter,

205 cruise, active ambush and mixed) were determined based on two available reference datasets
206 (Benedetti *et al.*, 2015; Brun *et al.*, 2017).

207

208 **Data analyses**

209 Vertical profiles of environmental data (temperature, salinity and fluorescence) were
210 generated using CTD cast data in Ocean Data View (Schlitzer, 2016). In addition, we used weekly
211 averaged satellite images of sea surface temperatures (SST) for the sampling dates. Images were
212 obtained from NASA MODIS Aqua Global Level 3 (NASA OBPG, 2020).

213 In order to account for differences in sequencing depth among samples, rarefaction to an
214 equivalent number of reads (minimum of 59,000 and 56,000 reads for COI and 18S, respectively)
215 was applied to all samples. Filtered rarefied data were used for Nonmetric Multidimensional
216 Scaling (nMDS) ordination and hierarchical clustering, according to the community composition
217 of mero- and holoplankton, which were sorted at the family level (Table S2). A resemblance matrix
218 was generated using Euclidean distances based on Hellinger-transformed rarefied read
219 abundances. Permutational multivariate analysis of variance (PERMANOVA) was conducted to
220 test the statistical significance of groups formed *a priori* (factors shelf position and season), as
221 well as of their interaction (Anderson, 2001).

222 In order to assess the relationship between the distribution of the main mesozooplankton
223 groups and the environmental variables, a Redundancy Analysis (RDA) was conducted. In order
224 to avoid collinearity of explanatory variables, we applied a variance inflation factor (VIF) and
225 removed collinear variables. A cut-off VIF value of 10 (Zuur *et al.*, 2009) was applied to get the
226 final set of covariates (distance from the coast, temperature, salinity, fluorescence, and thermal
227 stratification), which excluded depth and turbidity. Mean values of temperature, salinity and

228 fluorescence of each vertical CTD profiles were used in the RDA analysis. Read abundances were
229 transformed using the Hellinger transformation to reduce the wide disparity in magnitude of the
230 number of reads between taxa (Legendre and Gallagher, 2001). Taxa that represented less than 5%
231 of the reads were not included in this analysis.

232 In addition, we calculated the Shannon diversity index, considering OTUs as proxies of
233 species, as used previously for estimating plankton diversity (e.g. Ibarbalz *et al.*, 2019).

234 The analyses were performed with the R v3.5.2 environment (R foundation Core Team,
235 2018). The normalization, RDA, nMDS, clustering, PERMANOVA and additional tests were
236 conducted with the packages *vegan* (Oksanen *et al.*, 2008) and *HH* (Heiberger, 2020).

237

238 **Results**

239

240 **Hydrological structure**

241 Variations in temperature profiles between sampling dates were observed in the Iroise Sea
242 (Fig.1b-d; Fig. S1). Surface temperature reached 18°C in both transects during summer (Fig.1c;
243 Fig. S1b), while it did not exceed 15°C during spring and fall (Fig. 1b,d; Fig. S1a,c). The water
244 column was well-mixed during spring and fall (Fig. 1b,d), while it was strongly stratified during
245 summer in the offshore stations, in both transects, as shown by the stratification index (ΔT) (Fig.
246 1c). This showed that the westernmost stations (B7 and D6) were located in the frontal zone, where
247 water masses come into contact and mix (Fig. 1b). To a lesser extent, the nearshore stations,
248 especially the one located at the entrance of the bay of Douarnenez (D2), were also stratified (Fig.
249 1c).

250 Regarding fluorescence, values ranged between 0.5 and 15 $\mu\text{g l}^{-1}$, with highest values
251 observed during spring and summer (Fig. S2). Peaks were observed below 5 m during spring, and
252 at mid-depth (15-20 m) during summer for the Brest transect, especially at station B7 (Fig. S2a,b).
253 Salinity increased offshore, especially during spring (Fig. S3). However, the range of values was
254 very narrow (between 34.5 and 35.5) regardless of the sampling time or location.

255

256 **Taxonomic composition**

257 After bioinformatic filtering, the high-throughput sequencing effort yielded a total of
258 1,143,663 reads for the COI, and 1,039,947 for the 18S datasets. Additional information on number
259 of reads before processing, as well as mean number of reads per sample can be found in Table S3.

260 From the 380 and 296 metazoan OTUs (for COI and 18S, respectively), 95% belonged to
261 six phyla: Arthropoda (241 and 169 OTUs), Mollusca (47 and 33 OTUs), Cnidaria (31 and 40
262 OTUs), Annelida (19 and 12 OTUs), Chordata (12 and 14 OTUs), and Echinodermata (15 and 9
263 OTUs) (Table 1), with Arthropoda, especially copepods, representing nearly 60% of those. The
264 remaining 5% belonged to seven phyla that contained no more than 14 OTUs each (COI and 18S
265 combined) (Table 1).

266 Regarding the relative abundance (proportion of reads) over all samples combined, the
267 mesozooplankton community was dominated by copepods (61.2%), hydrozoans (9.7%),
268 appendicularians (8.5%), branchiopods (mainly cladocerans, 5.9%), chaetognaths (5.4%),
269 ctenophores (3.4%), polychaetes (2.5%), and malacostracans (euphausiids, amphipods and
270 decapods, 1.8%) (Table 1). The results obtained with both molecular markers were generally
271 consistent. However, the spatial distribution of some groups, such as Chaetognatha in spring (Fig.
272 2a,d) or Malacostraca in summer (Fig. 2b,e), was quite different when estimated with COI or 18S.

273 In addition, Porifera and Ostracoda were only detected with COI, while Cnidaria and
274 Appendicularia had lower relative abundances with COI (Fig. 2a-f).

275 Community structure varied over the course of the year, for instance, with polychaetes and
276 chaetognaths being more representative in spring (Fig. 2a,d), and malacostracans, branchiopods
277 and ctenophores in summer (Fig. 2b,e). During fall, the community composition was mainly
278 homogeneous along the stations, largely dominated by copepods and appendicularians (Fig. 2i,l).
279 The Shannon diversity index of the mesozooplankton community was slightly higher with COI as
280 compared with 18S (Fig. 2a-f). Overall, a cross-shelf pattern was observed for diversity, with
281 higher values closer to the coast and lower values in the most offshore stations (Fig. 2a-f). This
282 trend did not vary over time.

283 Holoplankton dominated the mesozooplankton community in the Iroise Sea, whatever the
284 transect and sampling time (Fig. 2g-l). Overall, meroplankton depicted an increase towards the
285 coast, and were more abundant during summer.

286 Taking a closer look at the meroplankton community, the effect of sampling time was the
287 most striking feature observed (Fig. 3a-c). This was exemplified by the near absence of reads
288 assigned to Decapoda in spring and their large relative abundance in summer in both transects.
289 This feature was statistically confirmed by the PERMANOVA that showed a significant effect of
290 sampling time on the composition of meroplankton (Table 2), and was also observed in the nMDS
291 and clustering analyses (Fig. 4a-b for COI; Fig. S4a-b for 18S). Conversely, no significant effect
292 of the shelf position was observed on the taxonomic composition of meroplankton, meaning that
293 the composition is stable across the shelf, at these high taxonomic levels (Table 2). Regarding the
294 taxonomic composition, polychaete larvae were remarkably more abundant in spring (Fig. 3a). In
295 turn, cnidarians (here exclusively hydroids), were mostly dominant in fall (Fig. 3c). Echinoderm

296 larvae were also found in greater numbers during fall (Fig. 3c), while decapod larvae were very
297 abundant during summer, and mainly in the most nearshore stations (Fig. 3b). Overall, the Shannon
298 index for meroplankton showed an increase towards the coast (Fig. 3a-c).

299 Meroplankton composition is shown at the family level for the most represented phylum
300 (Echinodermata), classes (Polychaeta and Bivalvia), and order (Decapoda) in Fig. 5. Overall, the
301 Shannon diversity index was higher towards the shore for most groups, with a few exceptions, in
302 accordance with the general trend observed for the whole meroplankton community (Fig. 3a-c). In
303 addition, Shannon indices were generally higher during spring and summer, and lower during fall.
304 Decapods were however one exception, presenting the highest diversity values during summer.
305 Generally, the meroplankton community showed a highly heterogeneous distribution over
306 relatively short spatial distances (stations were less than 30 km apart), as well as over time.
307 Substantial changes were also observed over seasons. For instance, amid echinoderms, larvae of
308 echinoids and holothurians were dominant during spring and summer, while larvae of ophiuroids
309 were mainly dominant during summer and fall (Fig. 5a-c). Polychaete larvae were dominated by
310 the families Oweniidae and Pectinariidae (Fig. 5d-f). Among bivalves, Mytilidae were dominant
311 during all three seasons, with contribution of Pectinidae during spring, and of Hiatellidae during
312 summer and fall (Fig. 5g-i). For decapods, caridean shrimps and brachyuran crabs were present
313 during all seasons, while anomuran crabs were only present during spring and summer, being
314 notably dominant during the warmest season (Fig. 5j-l).

315 In contrast to meroplankton, the Shannon diversity of holoplankton was relatively
316 homogeneous among months, but showed a decrease towards the nearshore stations (Fig. 3d-f).
317 The community was largely dominated by copepods, then by chaetognaths, appendicularians,
318 brachiopods and ctenophores. Pelagic polychaetes were also present in the community, mainly

319 during spring (Fig. 3d), while copepods were less represented in the offshore stations in spring and
320 summer (Fig. 3d,e). Shelf position was a significant factor for the holopelagic community with
321 both markers (Fig. 4c-d; Table 2; Fig. S4c-d). Season was also a significant factor for the
322 holoplankton community, but only with the 18S marker (Table 2; Fig. S4c-d), probably due to
323 more marked differences between months in comparison with the COI dataset. For instance, 18S
324 revealed higher relative abundance of appendicularians in fall and of polychaetes in spring (Fig.
325 2d-f).

326 Regarding the holoplankton, 183 OTUs were assigned to copepods with COI, which
327 belonged to 16 families (Fig. 6a-c). Shannon diversity values were relatively constant, showing
328 however a decreasing trend towards the coast. Calanoids were largely dominant in the area during
329 the sampling seasons, followed by cyclopoids. The calanoids Acartiidae, Temoridae and
330 Paracalanidae were the most abundant overall (Fig. 6a-c). During fall, a higher number of families
331 were present in the area, with the notable contribution of Centropagidae, Corycaeidae and
332 Oncaeidae, in addition to the regularly dominant calanoids (Fig. 6c).

333 Regarding copepod feeding traits, most were filter feeders with an omnivore-herbivore
334 trophic regime (Fig. 6d-i). Some spatial patterns were observed, with an increased proportion of
335 copepods with a mixed feeding strategy towards the offshore stations (Fig. 6g,i). In addition,
336 omnivore-detritivore copepods increased offshore during fall (Fig. 6f).

337

338 **Mesozooplankton distribution in relation to the oceanographic structure**

339 Regarding the influence of environmental variables on the distribution of the
340 meroplankton, holoplankton and copepod feeding traits, the first and second axes of the
341 Redundancy Analysis (RDA) ordination accounted together for 58.0%, 63.8, and 53.6% of the

342 constrained variance, respectively (Fig. 7; Table S4). All ordinations showed the segregation of
343 sampling times, with samples collected in summer being more distinct than those collected in
344 spring and fall (Fig. 7). This seems to be mainly driven by the unique thermohaline condition of
345 summer in the Iroise Sea, with a warmer and stratified water column, especially in the outer shelf
346 stations (yellow squares) (Fig. 7). Distance from the coast was an important factor, thus describing
347 the cross-shelf gradient, as clearly depicted along the ordinations (Fig. 7). Among meroplankton,
348 polychaete and mollusc larvae were mainly related to nearshore and intermediate shelf
349 environmental conditions, as opposed to cnidarians, which were found offshore (Fig. 7a). Amid
350 holoplankton, copepods showed an association with the nearshore stations (Fig. 7b). Regarding
351 the ordination for holoplanktonic groups, axis 2 mainly described the thermohaline gradient, with
352 highest temperature and thermal stratification (ΔT) opposed to salinity (Fig. 7b). Among the taxa,
353 brachiopods and ctenophores seemed to be the most associated with the summer conditions (Fig.
354 7b). For meroplankton, malacostracans (decapod larvae) were mostly associated with nearshore
355 summer stations (yellow circles) (Fig. 7a). Filter feeders and omnivore-herbivore copepods were
356 associated with summer warmer and stratified conditions (Fig. 7c).

357

358 **Discussion**

359 Here we investigated spatio-temporal variation in the composition of the mesozooplankton
360 community in a marine protected area (MPA) in the northeast Atlantic Ocean, based on a multi-
361 locus DNA metabarcoding approach.

362

363 **Meroplankton contributes to mesozooplankton community structure across**
364 **seasons**

365 The mesozooplankton community composition based on DNA metabarcoding data showed
366 significant differences throughout the year, consistently with both markers. This is coherent with
367 temporal variation described for temperate oceans, in response to oceanographic conditions
368 (Thomas and Nielsen, 1994; Beaugrand *et al.*, 2010). The CTD profiles revealed a clear spring-
369 to-summer transition in the Iroise Sea, represented by the stratification of the water column, and
370 the establishment of the Ushant Front in July, with the most offshore stations (B7 and D6) being
371 located in the frontal zone. Although our dataset did not include monthly sampling, inspection of
372 satellite SST images showed that the front persisted in August and September 2019 (data not
373 shown), while it was no longer present in October.

374 Mesozooplankton community structure was particularly strong in the meroplankton
375 component over the course of the year, with abundant polychaete larvae during spring and decapod
376 larvae during summer, for instance. In agreement with the present findings, and, importantly, using
377 the same sampling gear (i.e. 200- μ m WP2 net), a 20-year time-series from the Western English
378 Channel (station L4), also reported a peak of polychaete larvae in late spring, of decapod larvae in
379 June/July, and of echinoderm larvae in August and September (Highfield *et al.*, 2010). This time
380 series also found that seasonal variation accounted for the main changes in the composition of the
381 meroplankton, in addition to inter-annual variability. We hypothesize that the marked temporal
382 variation found for meroplankton in the Iroise Sea was due to two main factors, the
383 spawning/reproductive periods of species and the availability of food for the larvae.

384 Known spawning times for the most abundant benthopelagic species in the Iroise Sea
385 coincided with peaks in meroplankton proportions in spring and summer. For instance, the
386 polychaete *Lagis koreni* (Pectinariidae) is known to have its main spawning period from April to
387 June (Nicolaidou, 1983) in agreement with our detection of a peak in polychaete larvae in spring.

388 Another study in the NE Atlantic, including the area off the bay of Douarnenez, also found larvae
389 of *L. koreni* in May and June (Ayata *et al.*, 2011). In addition, *Ophiothrix fragilis* and *Ophiocolina*
390 *nigra*, two co-occurring ophiuroids very abundant in the area (Blanchet-Aurigny *et al.*, 2012;
391 Guillam *et al.*, 2020), were detected in all sampling periods, particularly in fall. Both have extended
392 breeding seasons, with spawning taking place at approximately monthly intervals from April to
393 October (Smith, 1940), in agreement with our observations.

394 Changes in phytoplankton community structure may reflect differential bloom timings and
395 intensities, with high contributions of diatoms over dinoflagellates in spring and summer, observed
396 in previous reports in this region (Birrien *et al.*, 1991; Ragueneau *et al.*, 1994; Cadier *et al.*, 2017a).
397 In the Iroise Sea, spring blooms are associated with higher proportions of diatoms, followed by
398 moderate drops in diatom abundances in summer, with dinoflagellates being more preponderant
399 in fall (Cadier *et al.*, 2017a; Benedetti *et al.*, 2019). Diatom blooms are triggered by the seasonal
400 stratification in April-May, which creates favorable conditions in nutrients and irradiance allowing
401 large opportunistic diatoms to grow (Cadier *et al.*, 2017a). Phytoplankton continue to show high
402 concentrations throughout summer due to the replenishment of nutrients in the coastal waters
403 through the establishment of the Ushant Front (Cadier *et al.*, 2017a). The front may therefore favor
404 larval survival by increasing resources available in the zone of the front as compared to nearshore
405 waters (Cadier *et al.*, 2017a, 2017b).

406
407 Like meroplankton, copepods also showed strong temporal variations. Regarding family
408 compositions, Acartiidae were dominant in spring, while Temoridae and Paracalanidae were
409 present during all seasons. During fall, a higher number of copepod families were present. When
410 evaluating the temporal variability of mesozooplankton in the area, Benedetti *et al.* (2019) also

411 found higher contributions of Acartiidae in spring and a higher number of unidentified Calanoida
412 in fall. With respect to the feeding traits, we observed the dominance of omnivore-herbivore filter-
413 feeders overall, with an increase of omnivore-detritivores and carnivores in fall. In the North
414 Atlantic, peaks of copepods that graze on phytoplankton have been seen in spring, while copepods
415 that feed on microzooplankton and detritus were dominant in other seasons (Friedland *et al.*, 2016).
416 Overall, omnivorous filter-feeding taxa (i.e. Temoridae and Paracalanidae copepods,
417 appendicularians and cladocerans) represented the dominant trophic trait over the
418 mesozooplankton community throughout the water column. To a lesser degree, carnivory was also
419 an important feeding behavior, with Corycaeidae copepods peaking during fall, and hydrozoans
420 and chaetognaths constituting the second and third most abundant groups in the area.

421

422 **High spatial heterogeneity in the mesozooplankton community from the** 423 **nearshore to offshore**

424 Stratification of the water column and temperature were key factors in determining
425 mesozooplankton community structure together with distance from the coast, reflecting the
426 nearshore-offshore gradient, as these explain most of the variation of the first RDA components.
427 In a previous study, using a nearshore-offshore transect in September 2009, Schultes *et al.* (2013)
428 found that the East-West gradient was the main pattern of zooplankton abundance variations in the
429 Iroise Sea. In our study, a noticeable differentiation along the cross-shelf gradient was observed,
430 mainly in terms of community composition for holoplankton and Shannon diversity index for
431 meroplankton. The inner stations had the highest level of diversity for meroplankton. This likely
432 reflects the influence of larval export from the bays, which hosts abundant and highly diverse
433 benthic communities (Chauvaud *et al.*, 2000; Gallon *et al.*, 2017). In addition, groups that are

434 known for their coastal affiliations (e.g. malacostracans and echinoderms) were associated with
435 nearshore and intermediate shelf environmental conditions.

436 Furthermore, we found that the mesozooplankton community was highly heterogeneous at
437 a small spatial scale, especially along the cross-shelf gradient. Changes in zooplankton community
438 composition between relatively nearby sites have been observed elsewhere, mainly attributed to
439 plankton patchiness and the presence of ocean fronts (Seda and Devetter, 2000; Trudnowska *et al.*,
440 2016). Plankton distribution and its patchiness are regulated by both biological and physical
441 processes that govern its abundance and community composition (Mackas *et al.*, 1985; Acha *et*
442 *al.*, 2015). In addition, the outflows from each of the bays may also play a role in the community
443 observed in the adjacent sampled area. The mixing intensity from the bays to the front leads to
444 localized and variable small oceanographic processes, contributing to the spatial variability even
445 within the coastal zone for the meroplankton community.

446 Mesozooplankton in the Iroise Sea provides a wide range of food resources for the higher
447 trophic levels. For example, crustaceans (small copepods, and decapod and cirriped larvae) are a
448 significant part of the diet of the sardine species *Sardina pilchardus*, a locally important
449 commercial species (Garrido *et al.*, 2008). Crustaceans are also an important food resource for the
450 exclusively planktivorous basking shark (*Cetorhinus maximus*), which annual sightings in the
451 Iroise Sea are associated with the Ushant Front (Sims, 2008), and for seabirds (Springer *et al.*,
452 2007). In addition, recent evidence showed that gelatinous zooplankton (ctenophores,
453 hydromedusae, chaetognaths and salps), which were also very abundant in our samples, form an
454 important part of fish diet with higher levels of protein, carbon content and other nutrients than
455 previously thought (Henschke *et al.*, 2016; Diaz Briz *et al.*, 2017).

456 Safeguarding the biodiversity of coastal waters and managing the sustainable exploitation
457 of marine resources are the main objectives within the establishment of the Iroise Marine Natural
458 Park. The plankton monitoring program (PNMIR) aims particularly to evaluate the interannual
459 variations in the phyto- and zooplankton communities and their link with fisheries of commercially
460 important species in the area, such as the European sardine *S. pilchardus* (Benedetti *et al.*, 2019).
461 Regarding the influence of the Ushant Front in the pelagic communities, a longer monitoring time-
462 series, with more frequent sampling would aid to follow the full cycle of the front. One further
463 limitation of our observations is the absence of sampling in fully oligotrophic waters, offshore of
464 the frontal zone. Sampling offshore would have allowed us to determine whether communities in
465 the frontal zone represented a combination of nearshore and offshore taxa, or whether taxa specific
466 to the frontal zone were present, as observed for phytoplankton (Landeira *et al.*, 2014). Offshore
467 sampling was logistically difficult to implement with the resources available to the MPA, but
468 future sampling and monitoring efforts that include sites beyond the frontal zone may improve our
469 understanding of the dynamics of planktonic communities and their effects on trophic webs and
470 biogeochemical cycles in the area. In addition, we recommend incorporating multi-marker DNA
471 metabarcoding as a standard tool for biodiversity assessment and environmental monitoring time-
472 series, as multiple compartments of the planktonic community sampled concurrently can be
473 examined in parallel.

474

475 **Conclusions**

476 Our study is the first to implement DNA metabarcoding to document the spatial and
477 temporal variations in the community composition and diversity of mesozooplankton in the Iroise
478 Sea. Metabarcoding of mixed tissue samples allowed us to detect a diverse array of marine

479 zooplankton and examine changes in community structure across several different phyla. Although
480 the mesozooplankton community was overwhelmingly dominated by holoplankton in terms of
481 relative abundance of reads, meroplankton showed a relatively high contribution of taxa present
482 in the area, expressed by the number of OTUs. It is worth pointing out that some meroplanktonic
483 groups may be underestimated due to the size of the mesh (200 μm), notably bivalve larvae.

484 Our findings indicate that spring and summer are the periods with higher relative
485 abundances and diversity for most meroplankton groups in the Iroise Sea, associated with the
486 spawning period of benthic species and with favorable environmental conditions for the
487 development of larvae, probably due to the presence of the Ushant Front. During fall, despite the
488 presence of some meroplanktonic groups, holoplankton taxa dominate the mesozooplankton
489 community in terms of relative abundance. Most meroplankters have likely settled by this time
490 period, or less larvae are released, and that the holoplankton taxa present feed mainly on sinking
491 and suspended particles (detritivores) and/or microzooplankton (carnivorous).

492 Our results for copepod feeding traits are in general agreement with observations for
493 bacterial communities associated with the Ushant Front. Copiotrophic bacteria that recycle detrital
494 organic matter were also dominant in communities following the period of the phytoplankton
495 blooms (Lemonnier *et al.*, 2020). Both copepod and bacterial communities therefore appear to
496 respond to increases in available organic matter post-bloom.

497 The significant small-scale spatial variation observed also reflect the strong nearshore-offshore
498 gradient. In addition, the importance of the stratification of the water column reflects the influence
499 of the Ushant Front in structuring the mesozooplankton community. In the future, the present
500 findings should be integrated with the characterization of other target communities, such as

501 phytoplankton, bacterioplankton, prokaryotes and higher trophic levels, in order to present a full
502 picture of the influence of the Ushant Front in the pelagic component of the Iroise Sea.

503

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515

516 **Data Availability Statement**

517 The data will be made available within a DOI once the acceptance of the manuscript.

518

519 **References**

- 520 Acha, E.M., Piola, A., Iribarne, O. and Mianzan, E. 2015. Ecological Processes at Marine Fronts: Oases in the Ocean.
521 Springer, Cham (68 pp).
- 522 Anderson, M. J. 2001. A new method for non-parametric multivariate analysis of variance. *Austral Ecology*, 26: 32–
523 46.
- 524 Ayata, S. D., Stolba, R., Comtet, T., and Thiébaud, É. 2011. Meroplankton distribution and its relationship to coastal
525 mesoscale hydrological structure in the northern Bay of Biscay (NE Atlantic). *Journal of Plankton Research*,
526 33: 1193–1211.
- 527 Baird, D. J., and Hajibabaei, M. 2012. Biomonitoring 2.0: A new paradigm in ecosystem assessment made possible
528 by next-generation DNA sequencing. *Molecular Ecology*, 21: 2039–2044.

529 Beaugrand, G., Edwards, M., and Legendre, L. 2010. Marine biodiversity, ecosystem functioning, and carbon
530 cycles. *Proceedings of the National Academy of Sciences of the United States of America*, 107: 10120–10124.

531 Benedetti, F., Gasparini, S., and Ayata, S. D. 2015. Identifying copepod functional groups from species functional
532 traits. *Journal of Plankton Research*, 38: 159–166.

533 Benedetti, F., Jalabert, L., Sourisseau, M., Becker, B., Cailliau, C., Desnos, C., Elineau, A., *et al.* 2019. The seasonal
534 and inter-annual fluctuations of plankton abundance and community structure in a North Atlantic Marine
535 Protected Area. *Frontiers in Marine Science*, 6: 1–16.

536 Birrien, J. L., Wafar, M. V. M., Corre, P. Le, and Riso, R. 1991. Nutrients and primary production in a shallow
537 stratified ecosystem in the Iroise Sea. *Journal of Plankton Research*, 13: 721–742.

538 Blanchet-Aurigny, A., Dubois, S. F., Hily, C., Rochette, S., Le Goaster, E., and Guillou, M. 2012. Multi-decadal
539 changes in two co-occurring ophiuroid populations. *Marine Ecology Progress Series*, 460: 79–90.

540 Bokulich, N. A., Subramanian, S., Faith, J. J., Gevers, D., Gordon, J. I., Knight, R., Mills, D. A., *et al.* 2013.
541 Quality-filtering vastly improves diversity estimates from Illumina amplicon sequencing. *Nature Methods*, 10:
542 57–59.

543 Brandão, M. C., Garcia, C. A. E., and Freire, A. S. 2020. Meroplankton community structure across oceanographic
544 fronts along the South Brazil Shelf. *Journal of Marine Systems*, 208.

545 Brumer, S. E., Garnier, V., Redelsperger, J. L., Bouin, M. N., Arduin, F., and Accensi, M. 2020. Impacts of surface
546 gravity waves on a tidal front: A coupled model perspective. *Ocean Modelling*, 154: 101677. Elsevier Ltd.
547 <https://doi.org/10.1016/j.ocemod.2020.101677>.

548 Brun, P., Payne, M. R., and Kiørboe, T. 2017. A trait database for marine copepods. *Earth System Science Data*, 9:
549 99–113.

550 Bucklin, A., Yeh, H. D., Questel, J. M., Richardson, D. E., Reese, B., Copley, N. J., and Wiebe, P. H. 2019. Time-
551 series metabarcoding analysis of zooplankton diversity of the NW Atlantic continental shelf. *ICES Journal of*
552 *Marine Science*, 76: 1162–1176.

553 Cadier, M., Sourisseau, M., Gorgues, T., Edwards, C. A., and Memery, L. 2017a. Assessing spatial and temporal
554 variability of phytoplankton communities' composition in the Iroise Sea ecosystem (Brittany, France): A 3D
555 modeling approach: Part 2: Linking summer mesoscale distribution of phenotypic diversity to
556 hydrodynamism. *Journal of Marine Systems*, 169: 111–126.

557 Cadier, M., Gorgues, T., Sourisseau, M., Edwards, C. A., Aumont, O., Marié, L., and Memery, L. 2017b.
558 Assessing spatial and temporal variability of phytoplankton communities' composition in the Iroise Sea
559 ecosystem (Brittany, France): A 3D modeling approach. Part 1: Biophysical control over plankton functional
560 types succession and distribution. *Journal of Marine Systems*, 165: 47–68. The Authors.
561 <http://dx.doi.org/10.1016/j.jmarsys.2016.09.009>.

562 Carroll, E. L., Gallego, R., Sewell, M. A., Zeldis, J., Ranjard, L., Ross, H. A., Tooman, L. K., *et al.* 2019. Multi-
563 locus DNA metabarcoding of zooplankton communities and scat reveal trophic interactions of a generalist
564 predator. *Scientific Reports*, 9: 1–14.

565 Chain, F. J. J., Brown, E. A., Macisaac, H. J., and Cristescu, M. E. 2016. Metabarcoding reveals strong spatial
566 structure and temporal turnover of zooplankton communities among marine and freshwater ports. *Diversity*
567 *and Distributions*, 22: 493–504.

568 Chauvaud, L., Jean, F., Ragueneau, O., and Thouzeau, G. 2000. Long-term variation of the Bay of Brest ecosystem:
569 Benthic-pelagic coupling revisited. *Marine Ecology Progress Series*, 200: 35–48.

570 Chevallier, C., Herbette, S., Marié, L., Le Borgne, P., Marsouin, A., Péré, S., Levier, B., *et al.* 2014. Observations of
571 the Ushant front displacements with MSG/SEVIRI derived sea surface temperature data. *Remote Sensing of*
572 *Environment*, 146: 3–10. Elsevier Inc. <http://dx.doi.org/10.1016/j.rse.2013.07.038>.

573 Clarke, L. J., Beard, J. M., Swadling, K. M., and Deagle, B. E. 2017. Effect of marker choice and thermal cycling
574 protocol on zooplankton DNA metabarcoding studies. *Ecology and Evolution*, 7: 873–883.

575 Couton, M., Comtet, T., Le Cam, S., Corre, E., and Viard, F. 2019. Metabarcoding on planktonic larval stages: An
576 efficient approach for detecting and investigating life cycle dynamics of benthic aliens. *Management of*
577 *Biological Invasions*, 10: 657–689.

578 Cowart, D. A., Matabos, M., Brandt, M. I., Marticorena, J., and Sarrazin, J. 2020. Exploring Environmental DNA
579 (eDNA) to Assess Biodiversity of Hard Substratum Faunal Communities on the Lucky Strike Vent Field
580 (Mid-Atlantic Ridge) and Investigate Recolonization Dynamics After an Induced Disturbance. *Frontiers in*
581 *Marine Science*, 6: 1–21.

582 Cristescu, M. E. 2014. From barcoding single individuals to metabarcoding biological communities: Towards an
583 integrative approach to the study of global biodiversity. *Trends in Ecology and Evolution*, 29: 566–571.
584 Elsevier Ltd. <http://dx.doi.org/10.1016/j.tree.2014.08.001>.

585 Deagle, B. E., Clarke, L. J., Kitchener, J. A., Polanowski, A. M., and Davidson, A. T. 2018. Genetic monitoring of
586 open ocean biodiversity: An evaluation of DNA metabarcoding for processing continuous plankton recorder
587 samples. *Molecular Ecology Resources*, 18: 391–406.

588 Diaz Briz, L., Sánchez, F., Marí, N., Mianzan, H., and Genzano, G. 2017. Gelatinous zooplankton (ctenophores,
589 salps and medusae): an important food resource of fishes in the temperate SW Atlantic Ocean. *Marine Biology*
590 *Research*, 13: 630–644.

591 Escudié, F., Auer, L., Bernard, M., Mariadassou, M., Cauquil, L., Vidal, K., Maman, S., *et al.* 2018. FROGS: Find,
592 Rapidly, OTUs with Galaxy Solution. *Bioinformatics*, 34: 1287–1294.

593 Flint, M. V., Sukhanova, I. N., Kopylov, A. I., Poyarkov, S. G., and Whitley, T. E. 2002. Plankton distribution
594 associated with frontal zones in the vicinity of the Pribilof Islands. *Deep-Sea Research Part II: Topical Studies*
595 *in Oceanography*, 49: 6069–6093.

596 Fonseca, V. G., Carvalho, G. R., Sung, W., Johnson, H. F., Power, D. M., Neill, S. P., Packer, M., *et al.* 2010.
597 Second-generation environmental sequencing unmasks marine metazoan biodiversity. *Nature*
598 *Communications*, 1: 1–8. Nature Publishing Group. <http://dx.doi.org/10.1038/ncomms1095>.

599 Friedland, K. D., Record, N. R., Asch, R. G., Kristiansen, T., Saba, V. S., Drinkwater, K. F., Henson, S., *et al.* 2016.
600 Seasonal phytoplankton blooms in the North Atlantic linked to the overwintering strategies of copepods.
601 *Elementa*, 2016: 1–19.

602 Fukami, H., Budd, A. F., Levitan, D. R., Jara, J., Kersanach, R., and Knowlton, N. 2004. Geographic differences in
603 species boundaries among members of the *Montastraea annularis* complex based on molecular and
604 morphological markers. *Evolution*, 58: 324–337.

605 Gallon, R. K., Lavesque, N., Grall, J., Labrune, C., Gremare, A., Bachelet, G., Blanchet, H., *et al.* 2017. Regional
606 and latitudinal patterns of soft-bottom macrobenthic invertebrates along French coasts: Results from the
607 RESOMAR database. *Journal of Sea Research*, 130: 96–106.

608 Garrido, S., Ben-Hamadou, R., Oliveira, P. B., Cunha, M. E., Chícharo, M. A., and Van Der Lingen, C. D. 2008.
609 Diet and feeding intensity of sardine *Sardina pilchardus*: Correlation with satellite-derived chlorophyll data.
610 *Marine Ecology Progress Series*, 354: 245–256.

611 Geller, J., Meyer, C., Parker, M., and Hawk, H. 2013. Redesign of PCR primers for mitochondrial cytochrome c
612 oxidase subunit I for marine invertebrates and application in all-taxa biotic surveys. *Molecular Ecology*
613 *Resources*, 13: 851–861.

614 Goecks, J., Nekrutenko, A., Taylor, J., Afgan, E., Ananda, G., Baker, D., Blankenberg, D., *et al.* 2010. Galaxy: a
615 comprehensive approach for supporting accessible, reproducible, and transparent computational research in
616 the life sciences. *Genome Biology*, 11.

617 Guillam, M., Bessin, C., Blanchet-Aurigny, A., Cugier, P., Nicolle, A., Thiébaud, É., and Comtet, T. 2020. Vertical
618 distribution of brittle star larvae in two contrasting coastal embayments: implications for larval transport.
619 *Scientific Reports*, 10. Nature Publishing Group UK. <https://doi.org/10.1038/s41598-020-68750-4>.

620 Heiberger, M. R. M. 2020. Package ‘HH’.

621 Henschke, N., Everett, J. D., Richardson, A. J., and Suthers, I. M. 2016. Rethinking the Role of Salps in the Ocean.
622 *Trends in Ecology and Evolution*, 31: 720–733. Elsevier Ltd. <http://dx.doi.org/10.1016/j.tree.2016.06.007>.

623 Highfield, J. M., Eloire, D., Conway, D. V. P., Lindeque, P. K., Attrill, M. J., and Somerfield, P. J. 2010. Seasonal
624 dynamics of meroplankton assemblages at station L4. *Journal of Plankton Research*, 32: 681–691.

625 Ibarbalz, F. M., Henry, N., Brandão, M. C., Martini, S., Bussení, G., Byrne, H., Coelho, L. P., *et al.* 2019. Global
626 Trends in Marine Plankton Diversity across Kingdoms of Life. *Cell*, 179.

627 Landeira, J. M., Ferron, B., Lunven, M., Morin, P., Marie, L., and Sourisseau, M. 2014. Biophysical interactions
628 control the size and abundance of large phytoplankton chains at the ushant tidal front. *PLoS ONE*, 9.

629 Le Boyer, A., Cambon, G., Daniault, N., Herbette, S., Le Cann, B., Marié, L., and Morin, P. 2009. Observations of
630 the Ushant tidal front in September 2007. *Continental Shelf Research*, 29: 1026–1037.

631 Legendre, P., and Gallagher, E. D. 2001. Ecologically meaningful transformations for ordination of species data.
632 *Oecologia*, 129: 271–280.

633 Lejzerowicz, F., Esling, P., Pillet, L. L., Wilding, T. a., Black, K. D., and Pawlowski, J. 2015. High-throughput
634 sequencing and morphology perform equally well for benthic monitoring of marine ecosystems. *Scientific*
635 *Reports*, 5: 13932. Nature Publishing Group. <http://www.nature.com/doi/10.1038/srep13932>.

636 Lemonnier, C., Perennou, M., Eveillard, D., Fernandez-Guerra, A., Leynaert, A., Marié, L., Morrison, H. G., *et al.*
637 2020. Linking Spatial and Temporal Dynamic of Bacterioplankton Communities With Ecological Strategies
638 Across a Coastal Frontal Area. *Frontiers in Marine Science*, 7: 1–13.

639 Leray, M., Yang, J. Y., Meyer, C. P., Mills, S. C., Agudelo, N., Ranwez, V., Boehm, J. T., *et al.* 2013. A new
640 versatile primer set targeting a short fragment of the mitochondrial COI region for metabarcoding metazoan

641 diversity: Application for characterizing coral reef fish gut contents. *Frontiers in Zoology*, 10: 1–14.

642 Lindeque, P. K., Parry, H. E., Harmer, R. A., Somerfield, P. J., and Atkinson, A. 2013. Next generation sequencing
643 reveals the hidden diversity of zooplankton assemblages. *PLoS ONE*, 8: 1–14.

644 Mackas, D. L., Denman, K. L., and Abbott, M. R. 1985. Plankton patchiness: biology in the physical vernacular.
645 *Bulletin of Marine Science*, 37: 653–674.

646 Magoč, T., and Salzberg, S. L. 2011. FLASH: Fast length adjustment of short reads to improve genome assemblies.
647 *Bioinformatics*, 27: 2957–2963.

648 Mahé, F., Rognes, T., Quince, C., de Vargas, C., and Dunthorn, M. 2014. Swarm: Robust and fast clustering method
649 for amplicon-based studies. *PeerJ*, 2014: 1–13.

650 Marra, J., Houghton, R. W., and Garside, C. 1990. Phytoplankton growth at the shelf-break front in the Middle
651 Atlantic Bight. *Journal of Marine Research*, 48: 851–868.

652 Martinetto, P., Alemany, D., Botto, F., Mastrángelo, M., Falabella, V., Acha, E. M., Antón, G., *et al.* 2020. Linking
653 the scientific knowledge on marine frontal systems with ecosystem services. *Ambio*, 49: 541–556.

654 NASA Ocean Biology Processing Group. 2020. Moderate-resolution Imaging Spectroradiometer (MODIS) Aqua
655 SST Data; NASA OB.DAAC, Greenbelt, MD, USA. Accessed on 01/14/2021.

656 Nicolaidou, A. 1983. Life history and productivity of *Pectinaria koreni* Malmgren (polychaeta). *Estuarine, Coastal
657 and Shelf Science*, 17: 31–43.

658 Ohman, M. D., Powell, J. R., Picheral, M., and Jensen, D. W. 2012. Mesozooplankton and particulate matter
659 responses to a deep-water frontal system in the southern California Current System. *Journal of Plankton
660 Research*, 34: 815–827.

661 Oksanen, J., Kindt, R., Legendre, P., O’Hara, B., Simpson, G. L., Solymos, P. M., Stevens, M. H. H., *et al.* 2008.
662 The vegan package. *Community ecology package*: 190.
663 <https://bcrb.bio.umass.edu/biometry/images/8/85/Vegan.pdf>.

664 Pingree, R. D., Pugh, P. R., Holligan, P. M., and Forster. 1975. Summer phytoplankton blooms and red tides along
665 tidal fronts in the approaches to the English Channel. *Nature*, 258: 672–677.

666 Questel, J. M., Hopcroft, R. R., DeHart, H. M., Smoot, C. A., Kosobokova, K. N., Bucklin, A. 2021. Metabarcoding
667 of zooplankton diversity within the Chukchi Borderland, Arctic Ocean: improved resolution from multi-gene
668 markers and region-specific DNA databases. *Marine Biodiversity*, 51: 4.

669 R Core Team. 2018. R: A Language and Environment for Statistical Computing. R Foundation for Statistical
670 Computing, Vienna, Austria, <https://www.R-project.org/>.

671 Ragueneau, O., Varela, E. D., Treguer, P., Queguiner, B., and Delamo, Y. 1994. Phytoplankton dynamics in relation
672 to the biogeochemical cycle of silicon in a coastal ecosystem of Western Europe. *Marine Ecology Progress
673 Series*, 106: 157–172.

674 Ratnasingham, S., and Hebert, P. D. N. 2007. The Barcode of Life Data System. *Molecular Ecology Notes*, 7: 355–
675 364.

676 Rognes, T., Flouri, T., Nichols, B., Quince, C., and Mahé, F. 2016. VSEARCH: A versatile open source tool for
677 metagenomics. *PeerJ*, 2016: 1–22.
678 <https://doi.org/10.1016/j.pocean.2019.102202>.

679 Schlitzer. 2016. Ocean Data View User’s Guide Acknowledgements: 1–173.
680 https://odv.awi.de/fileadmin/user_upload/odv/misc/odv4Guide.pdf.

681 Schroeder, A., Stanković, D., Pallavicini, A., Gionechetti, F., Pansera, M., and Camatti, E. 2020. DNA
682 metabarcoding and morphological analysis - Assessment of zooplankton biodiversity in transitional waters.
683 *Marine Environmental Research*, 160.

684 Schultes, S., Sourisseau, M., Le Masson, E., Lunven, M., and Marié, L. 2013. Influence of physical forcing on
685 mesozooplankton communities at the Ushant tidal front. *Journal of Marine Systems*, 109–110: S191–S202.
686 Elsevier B.V. <http://dx.doi.org/10.1016/j.jmarsys.2011.11.025>.

687 Seda, J., and Devetter, M. 2000. Zooplankton community structure along a trophic gradient in a canyon-shaped dam
688 reservoir. *Journal of Plankton Research*, 22: 1829–1840.

689 Shanks, A. L., McCulloch, A., and Miller, J. 2003. Topographically generated fronts, very nearshore oceanography
690 and the distribution of larval invertebrates and holoplankters. *Journal of Plankton Research*, 25: 1251–1277.

691 Sims, D. W. 2008. Chapter 3 Sieving a Living. A Review of the Biology, Ecology and Conservation Status of the
692 Plankton-Feeding Basking Shark *Cetorhinus Maximus*. *Advances in Marine Biology*, 54: 171–220.

693 Sinniger, F., Pawlowski, J., Harii, S., Gooday, A. J., Yamamoto, H., Chevalloné, P., Cedhagen, T., *et al.* 2016.
694 Worldwide analysis of sedimentary DNA reveals major gaps in taxonomic knowledge of deep-sea benthos.
695 *Frontiers in Marine Science*, 3: 1–14.

696 Smith, J. B. 1940. *Memoirs: The Reproductive System and Associated Organs of the Brittle-Star Ophiothrix*

697 Fragilis. *Journal of Cell Science*, s2-82: 267–309.

698 Springer, A. M., Byrd, G. V., and Iverson, S. J. 2007. Hot oceanography: Planktivorous seabirds reveal ecosystem
699 responses to warming of the Bering Sea. *Marine Ecology Progress Series*, 352: 289–297.

700 Stefanni, S., Stanković, D., Borme, D., de Olazabal, A., Juretić, T., Pallavicini, A., and Tirelli, V. 2018. Multi-
701 marker metabarcoding approach to study mesozooplankton at basin scale. *Scientific Reports*, 8: 1–13.

702 Thomas, K., and Nielsen, T. G. 1994. Regulation of zooplankton biomass and production in a temperate, coastal
703 ecosystem. 1. Copepods. *Limnology and Oceanography*, 39: 493–507.

704 Trudnowska, E., Gluchowska, M., Beszczynska-Möller, A., Blachowiak-Samolyk, K., and Kwasniewski, S. 2016.
705 Plankton patchiness in the Polar Front region of the west Spitsbergen Shelf. *Marine Ecology Progress Series*,
706 560: 1–18.

707 UNESCO. 1968. *Monographs on Oceanographic Methodology: Zooplankton Sampling*. UNESCO, Paris (174 pp).

708 Valentini, Taberlet P, Miaud C, Civade R, Herder J, Thomsen PF, Bellemain E, *et al.* 2016. Next-generation
709 monitoring of aquatic biodiversity using environmental {DNA} metabarcoding. *Molecular Ecology*, 25: 929–
710 942.

711 Videau, C. 1987. Primary production and physiological state of phytoplankton at the Ushant tidal front (west coast
712 of Brittany, France). *Marine Ecology Progress Series*, 35: 141–151.

713 Woodson, C. B., and Litvin, S. Y. 2015. Ocean fronts drive marine fishery production and biogeochemical cycling.
714 *Proceedings of the National Academy of Sciences of the United States of America*, 112: 1710–1715.

715 Zuur, A. F., Ieno, E. N., Walker, N. J., Saveliev, A. A., Smith, G. 2009. *Mixed Effects Models and Extensions in*
716 *Ecology with R*. Springer, New York (574 pp).

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718 **Tables and Figures**

719

720 **Table 1. Number of reads and OTUs by Phylum and Class.** Number of reads and OTUs
721 assigned to Phylum and Class for COI and 18S markers, ordered by higher to lower number of
722 reads.

Phylum	Class	Common name	OTUs		Reads	
			COI	18S	COI	18S
Arthropoda	Hexanauplia	copepods and barnacles	183	129	732,753	604,461
Cnidaria	Hydrozoa	jellyfish-like	31	39	86,787	124,656
Chordata	Appendicularia	tunicates	8	11	81,126	103,611
Arthropoda	Branchiopoda	planktonic crustaceans	13	8	50,340	79,308
Chaetognatha	Sagittoidea	arrow worms	6	7	61,803	56,037
Ctenophora	Tentaculata	comb jellies	8	6	64,110	10,926
Annelida	Polychaeta	polychaetes	19	12	25,458	28,851
Arthropoda	Malacostraca	crustaceans	41	32	22,356	16,944
Mollusca	Bivalvia	bivalves	33	21	8,757	7,788
Mollusca	Gastropoda	sea snails	14	12	2,655	1,926
Chordata	Actinopterygii	ray-finned fish	3	2	1,233	2,766
Chordata	Thaliacea	salps	1	1	1,269	1,875
Porifera	Demospongiae	sponges	1	-	3,075	-
Echinodermata	Ophiuroidea	brittle stars	6	4	771	426
Echinodermata	Echinoidea	sea urchins	6	4	411	117
Arthropoda	Ostracoda	ostracods	4	-	501	-
Echinodermata	Holothuroidea	sea cucumbers	3	1	258	171
Platyhelminthes	Rhabditophora	flatworm	-	1	-	27
Cnidaria	Anthozoa	jellyfish-like	-	1	-	18
Bryozoa	Gymnolaemata	moss animals	-	1	-	15

Phoronida		horseshoe worm	-	2	-	12
Nemertea	Palaeonemertea	marine worm	-	2	-	12
Metazoans			380	296	1,143,663	1,039,947

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724 **Table 2. Results of PERMANOVA.** Results of the PERMANOVA comparing the mero- and
725 holoplankton community composition between seasons and/or among shelf position, for COI and
726 18S genes.

COI				18S			
	df	F	<i>p</i>		df	F	<i>p</i>
Meroplankton				Meroplankton			
Season	2	1.89	0.011 *	Season	2	4.93	0.000 ***
Shelf position	2	0.67	0.907	Shelf position	2	1.98	0.298
S x SP	4	0.82	0.742	S x SP	4	1.16	0.460
Residual	9			Residual	9		
Holoplankton				Holoplankton			
Season	2	1.99	0.054	Season	2	3.45	0.000 ***
Shelf position	2	4.03	0.005 **	Shelf position	2	4.32	0.000 ***
S x SP	4	1.12	0.709	S x SP	4	1.04	0.512
Residual	9			Residual	9		

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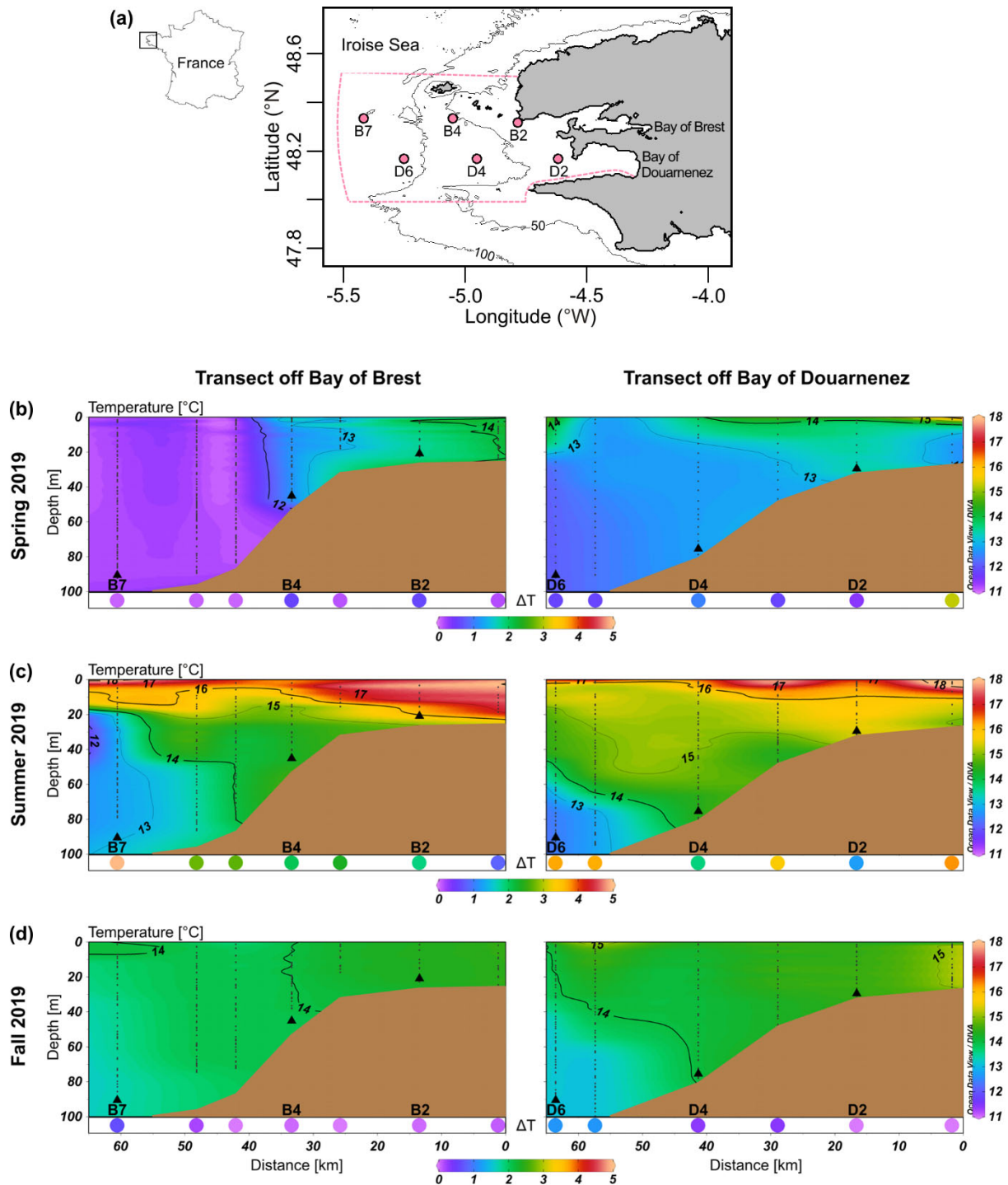
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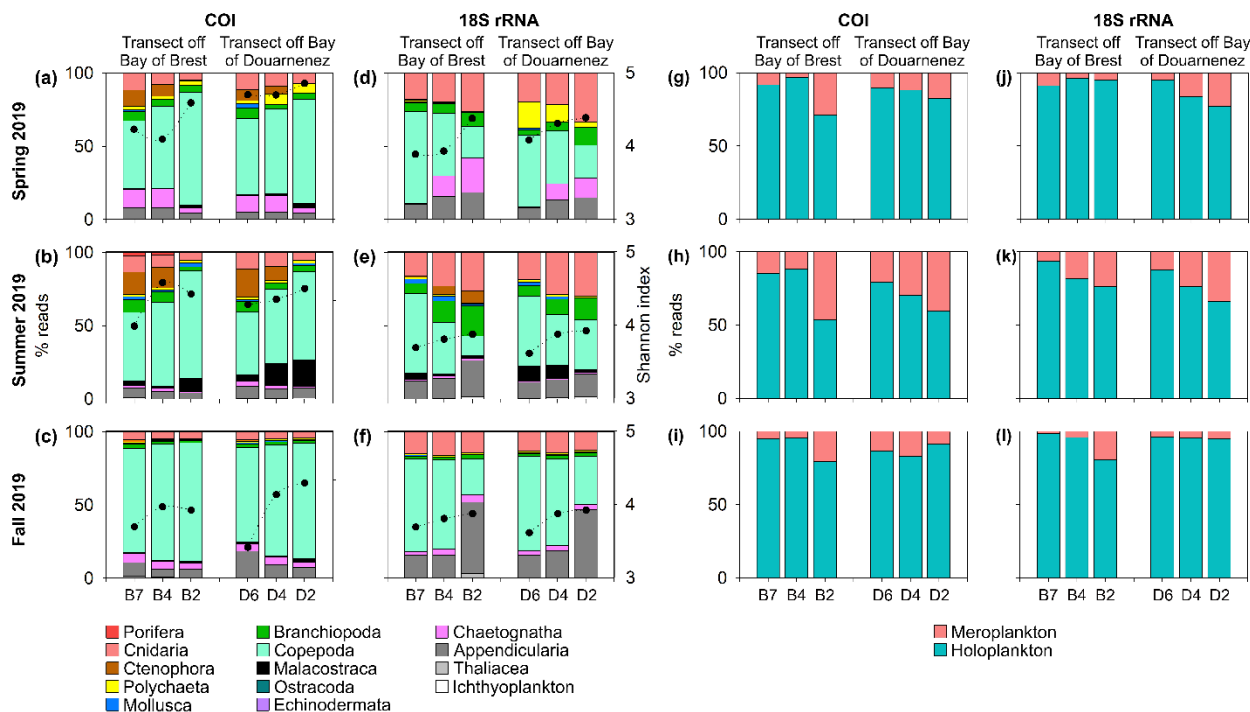
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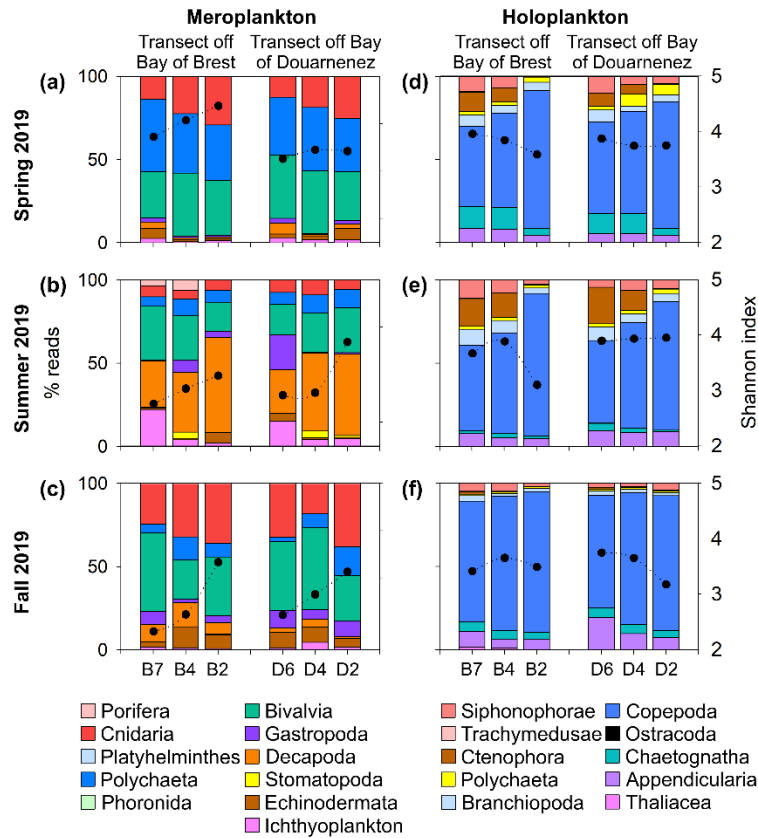
753 **Figure 1. Location of the study area and vertical profiles of temperature.** (a) Position of the
 754 sampling stations in the Iroise Sea, along the transects off the bays of Brest (B2-B7) and
 755 Douarnenez (D2-D6). The red dashed line represents the boundaries of the Iroise Marine Natural
 756 Park. Vertical profiles of temperature, and stratification index (ΔT ; indicated with color dots at the

757 bottom of the vertical temperature profiles) along the two transects during sampling in (b) spring,
 758 (c) summer, and (d) fall 2019. Triangles represent the starting depth of plankton tows. Vertical
 759 dashed lines indicate the location of CTD casts.
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 762 **Figure 2. Proportion of reads of the mesozooplankton taxa, and reads associated to mero-**
 763 **and holoplankton.** Proportion (percentage of reads) of mesozooplankton taxa with COI (a-c) and
 764 18S (d-f) genes, and of mero- and holoplankton with COI (g-i) and 18S (j-l), in the Iroise Sea
 765 during spring, summer and fall 2019, along the two study transects. Black dots represent the
 766 Shannon diversity index. Taxa that represented less than 0.02% of the reads are not shown.

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777 **Figure 3. Proportion of main taxa for mero- and holoplankton.** Proportion (percentage of
 778 reads) of the main taxa of mero- and ichthyoplankton (a-c) and holoplankton (d-f) in the Iroise Sea
 779 during spring, summer and fall 2019 with the COI gene. Black dots represent the Shannon diversity
 780 index.

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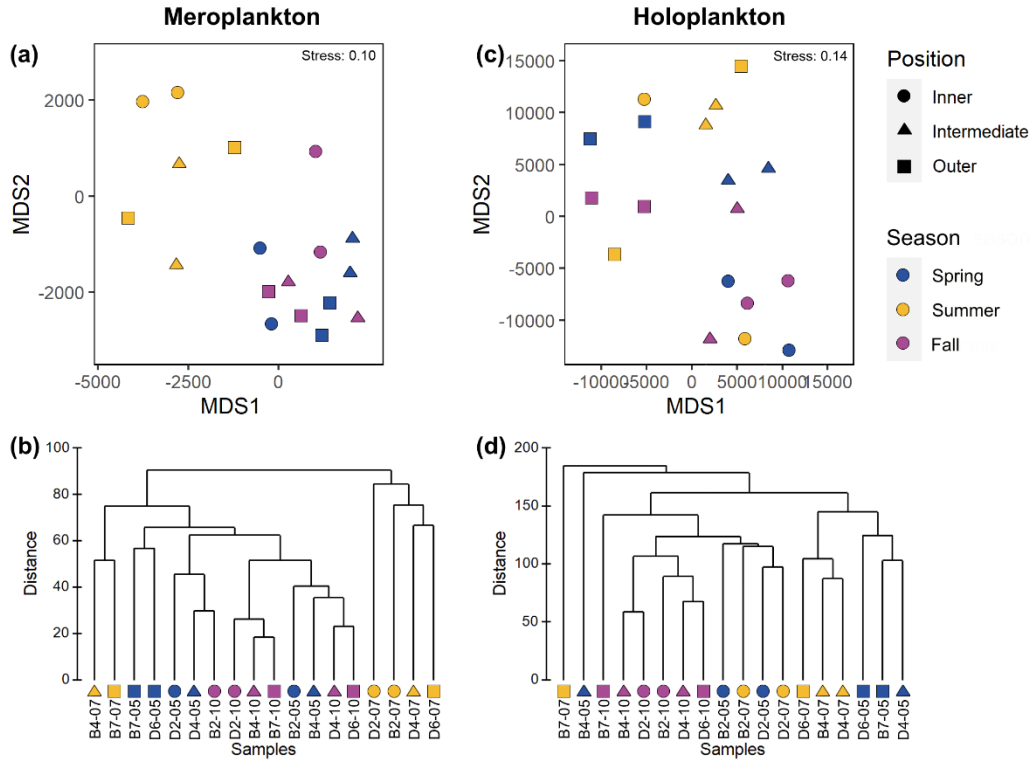
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794 **Figure 4. NMDS and clustering of the samples for mero- and holoplankton taxa.** Nonmetric
 795 multidimensional scaling (nMDS) ordination and clustering of samples collected in the Iroise Sea
 796 based on the rarefied abundance (number of reads) of OTUs assigned to mero- (a,b) and
 797 holoplankton (c,d) taxa with the COI gene. Samples are classified according to shelf position and
 798 season of sampling.

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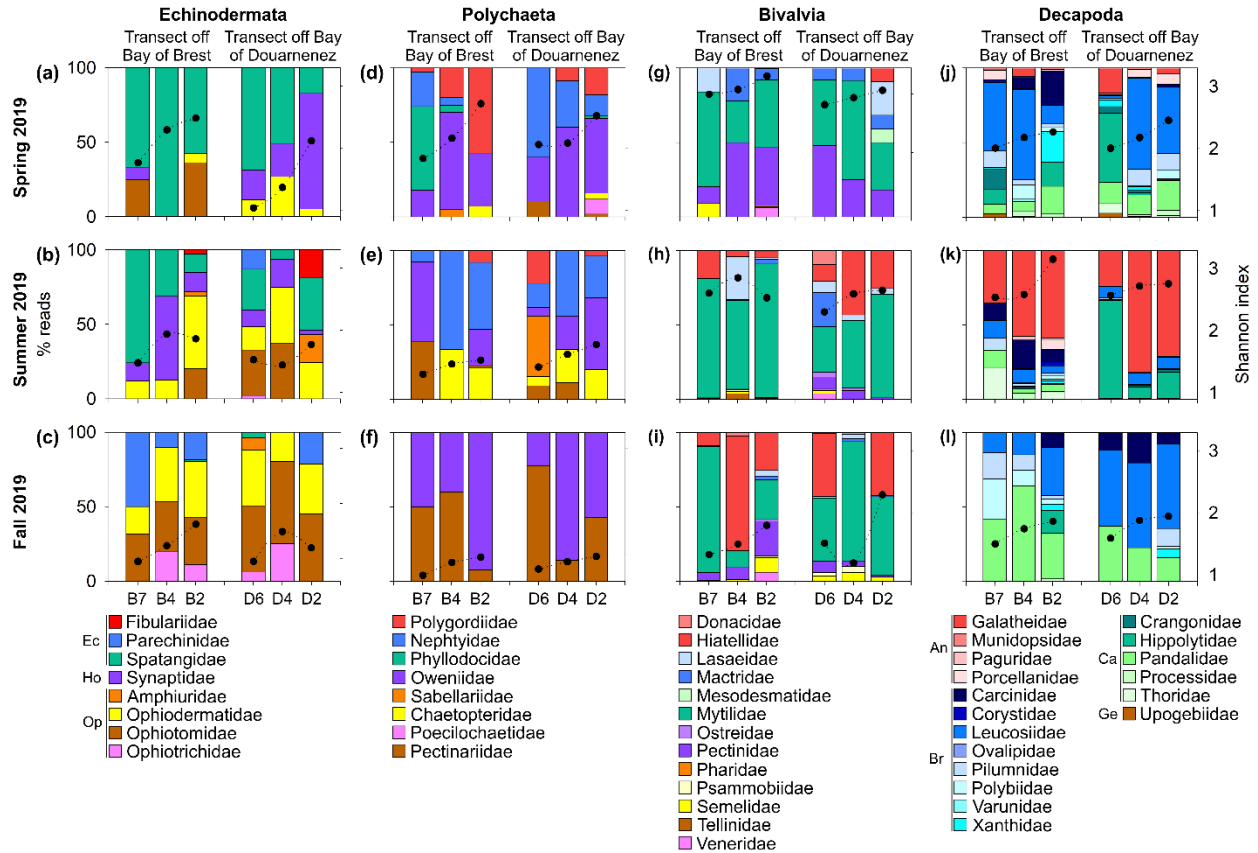
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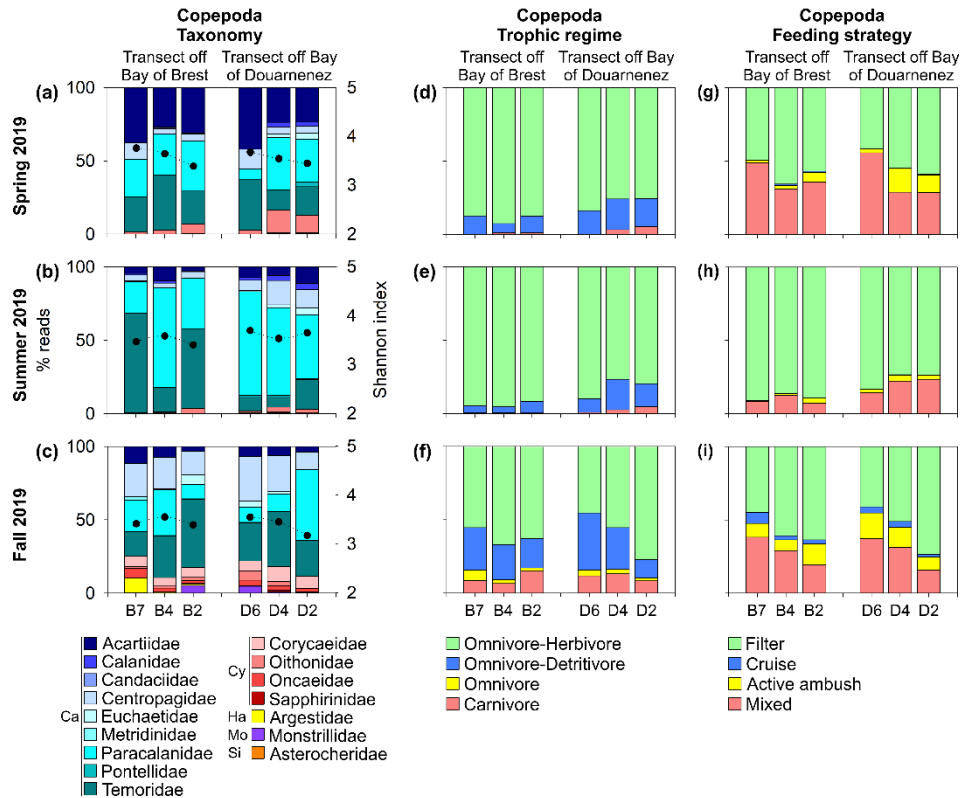
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 812 **Figure 5. Community composition, at the family level, for Echinodermata, Polychaeta,**
 813 **Bivalvia and Decapoda, focusing on meroplankton.** Proportion (percentage of reads) per family
 814 for Echinodermata (a-c), Polychaeta (d-f), Bivalvia (g-i) and Decapoda (j-l) in the Iroise Sea during
 815 spring, summer and fall 2019 with the COI gene. Black dots represent the Shannon diversity index.
 816 For echinoderms, Ec = Echinoidea, Ho = Holothuroidea, and Op = Ophiuroidea. For decapods, An
 817 = Anomura, Br = Brachyura, Ca = Caridea, and Ge = Gebiidea.

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828 **Figure 6. Taxonomic composition and feeding traits of copepods.** Proportion (percentage of
 829 reads) of copepod families (a-c), and copepods feeding traits (d-i) in the Iroise Sea during spring,
 830 summer and fall 2019 as obtained from taxonomic assignment with the COI gene. Black dots
 831 represent the Shannon diversity index. Copepods are colored according to their Orders: Ca =
 832 Calanoida (shades of blue), Cy = Cyclopoida (shades of pink), Ha = Harpacticoida (yellow), Mo
 833 = Monstrilloida (purple), and Si = Siphonostomatoida (orange). Copepods traits based on available
 834 datasets (Benedetti *et al.*, 2015; Brun *et al.*, 2017).

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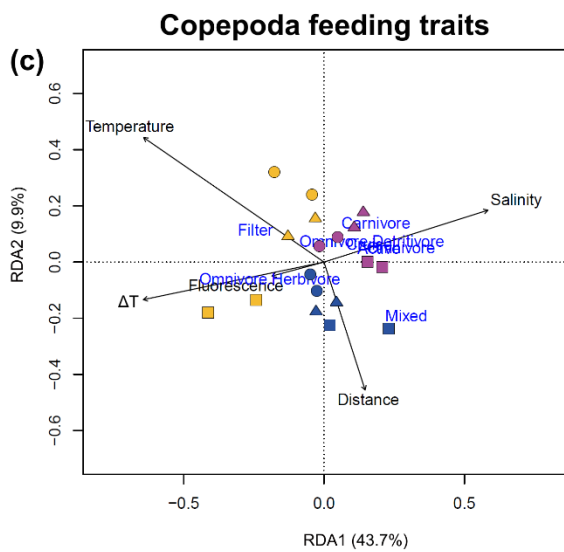
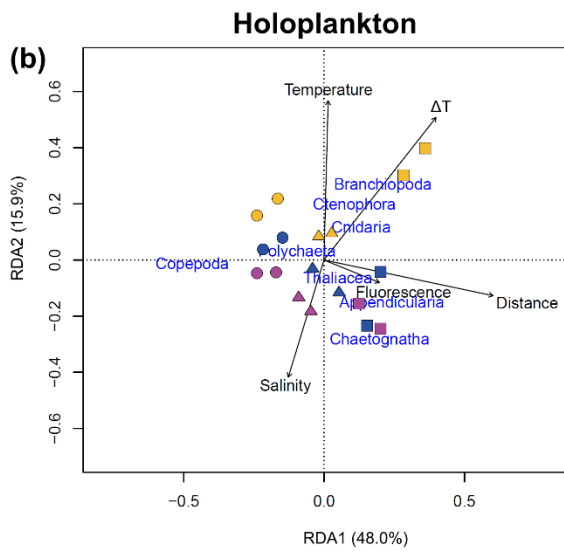
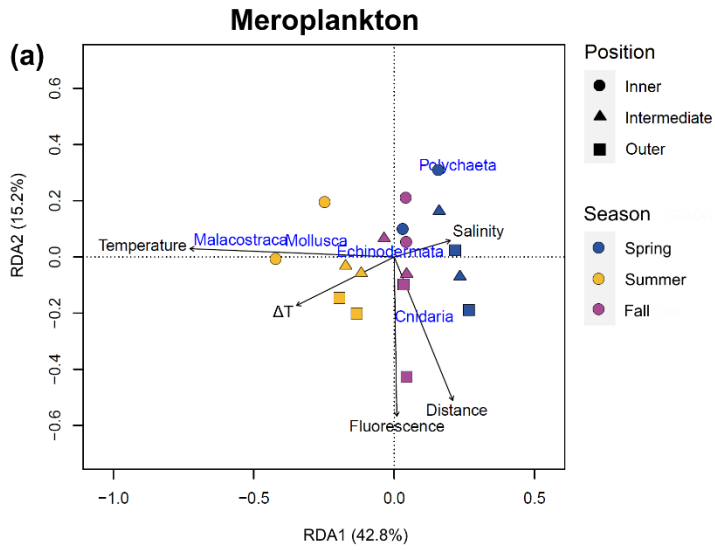
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845 **Figure 7. Redundancy Analysis (RDA) ordination for meroplankton, holoplankton and**
846 **copepods feeding traits in relation to environmental variables.** Triplot of the RDA ordination
847 with explanatory variables (arrows), taxa (abbreviations in blue) and samples (symbols), based on
848 OTUs retrieved from the COI dataset and assigned to meroplankton (a), holoplankton (b), and
849 copepods feeding traits (c). Samples are classified according to shelf position and season of
850 sampling.