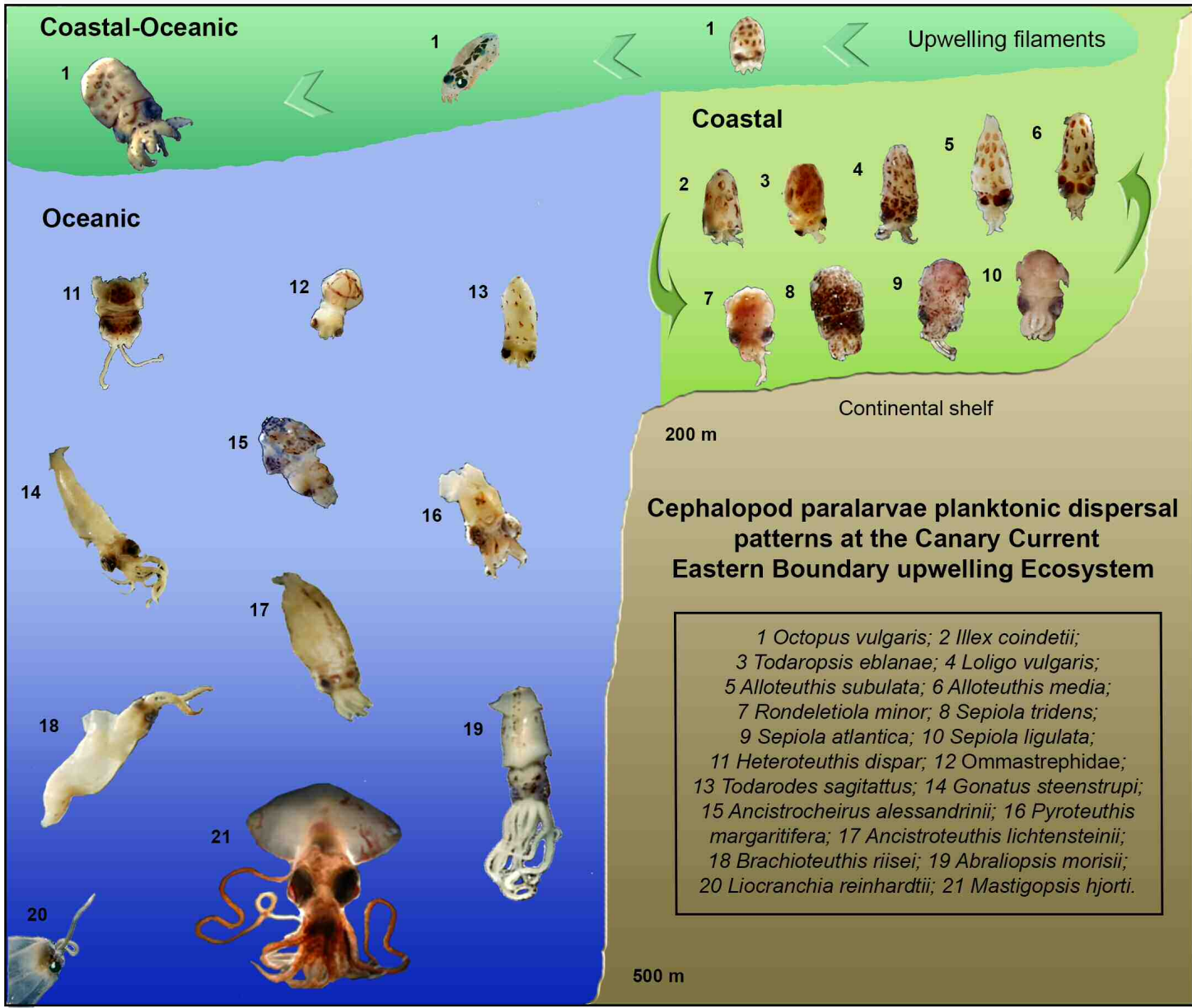


Highlights

1. Cephalopod paralarval richness was 2x higher in Moroccan than Iberian waters
2. Three planktonic dispersal patterns were identified in the Iberian–Canary current upwelling system
3. The interaction between vertical behaviour and oceanography led to these 3 dispersal patterns
4. Each planktonic pattern had different haplotype and nucleotide genetic signatures
5. *Octopus vulgaris* paralarvae shift within upwelling filaments from coast to ocean

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36



1
2
3
4 **1 Oceanographic processes shape genetic signatures of planktonic cephalopod paralarvae**
5 **2 in two upwelling regions**
6

7
8 3 Álvaro Roura^{a, b*}, Michael Amor^b, Ángel F. González^a, Ángel Guerra^a, Eric D. Barton^a, Jan
9 4 M. Strugnell^{b,c}
10

11
12 6 ^a Instituto de Investigaciones Marinas (IIM-CSIC), 36208 Vigo, Spain. Email: AFG,
13 7 afg@iim.csic.es; AG, angelguerra@iim.csic.es; EDB: e.d.barton@iim.csic.es

14
15 8 ^b Department of Ecology, Environment and Evolution, La Trobe University, Melbourne 3086,
16 9 Australia. Email: michael.amor88@gmail.com

17
18 10 ^c Centre for Sustainable Tropical Fisheries and Aquaculture, James Cook University,
19 11 Townsville, Queensland, 4810, Australia. Email: JMS, jan.strugnell@jcu.edu.au

20
21 12 *Corresponding author: aroura@iim.csic.es
22
23

24
25 13 **Abstract**
26

27 14 The planktonic paralarval stage of cephalopods (octopus, squids and cuttlefishes) is an
28 15 important dispersal phase, particularly of benthic species, that lasts from days to months.
29 16 Cephalopod paralarvae modify their vertical position in the water in upwelling ecosystems
30 17 and such behaviour influences their spatial distribution and genetic structure, but to what
31 18 extent? In this work specific water masses were sampled with Lagrangian buoys in two
32 19 contrasting upwelling systems (Iberian Peninsula and Morocco) of the Iberian–Canary
33 20 current eastern boundary upwelling (ICC) in order to: i) identify the cephalopod assemblage
34 21 in the different upwelling systems ii) define their planktonic dispersal patterns and iii)
35 22 analyse the effect of different dispersal patterns on genetic structure and connectivity.
36 23 Cephalopod paralarvae were identified using the cytochrome *c* oxidase subunit I gene (COI),
37 24 revealing 21 different species and F_{st} values showed no population structure between both
38 25 upwelling systems. Cephalopod species richness was two times higher in the Moroccan
39 26 upwelling than in the Iberian Peninsula, with an undescribed Ancistrocheiridae species
40 27 identified in Moroccan waters. Three common planktonic dispersal patterns were identified in
41 28 the ICC: coastal, coastal-oceanic and oceanic. Coastal and oceanic dispersal patterns
42 29 favoured spatio-temporal paralarval retention or “schooling” of different cohorts over the
43 30 continental shelf and continental slope in 9 and 11 species, respectively. Such spatio-temporal
44 31 retention was reflected in the complex haplotype networks and high nucleotide / haplotype
45 32 diversity recorded for these two groups. The only cephalopod species displaying a coastal-
46
47
48
49
50
51
52
53
54
55
56
57
58
59

60
61
62
63
64
65
66
67
68
69
70
71
72
73
74
75
76
77
78
79
80
81
82
83
84
85
86
87
88
89
90
91
92
93
94
95
96
97
98
99
100
101
102
103
104
105
106
107
108
109
110
111
112
113
114
115
116
117
118

33 oceanic dispersal pattern was *Octopus vulgaris*, where low nucleotide and haplotype diversity
34 was observed. The observed decline in genetic structure resulted from the dispersal of similar
35 cohorts within upwelling currents and upwelling filaments to the oceanic realm. Seascape
36 analysis revealed that cephalopod paralarvae from two coastal upwelling ecosystems of the
37 ICC display three planktonic dispersal patterns with contrasting distributions and signatures
38 at the genetic level.

39

40 **Keywords:** upwelling filaments, cephalopod paralarvae, seascape genetics, eastern boundary
41 upwelling system, planktonic dispersal patterns, *Octopus vulgaris*, Northeastern Atlantic.

42

43 **Author Contributions**

44 Conceived and designed the experiment: AR, AFG, EDB; collected samples: AR, AFG;
45 processed the samples: AR; analysed the data: AR, MA, JMS; contributed reagents /
46 materials / analysis tools: AFG, JMS and AG; AR wrote the first draft of the manuscript, and
47 all authors contributed substantially to revisions.

48 All authors declare no conflict of interest and have approved the final version of the
49 manuscript.

119
120
121 **50 1. Introduction**
122
123

124 51 Understanding larval exchange between populations of marine organisms is a
125 52 fundamental aspect of population ecology and essential for the management of fishery stocks.
126 53 Among the different types of larvae present in the water, planktotrophic larvae spend more
127 54 time in the plankton than their lecithotrophic counterparts (Vance, 1973). It is expected that
130 55 longer pelagic duration would result in increased dispersal, increased gene flow and,
131 56 consequently, decreased levels of population genetic structure. Although there are many
132 57 studies that suggest this hypothesis (Bohonak, 1999; Siegel et al., 2003), Weersing and
133 58 Toonen (2009) concluded from a meta-analysis of 87 studies of pelagic larval duration (PLD)
134 59 and population genetic estimates - measured as global F_{st} - that mean PLD is a poor predictor
138 60 of connectivity. Most models assume that planktotrophic larvae behave as passive particles
139 61 (e.g. Teske et al. 2015), however when coupling vertical migration behaviour with mesoscale
140 62 or sub-mesoscale circulations, long distance dispersal can be minimized and larvae are
141 63 retained in close proximity to the area that they hatched (Shanks and Eckert, 2005; Queiroga
142 64 et al. 2007; Morgan and Fisher, 2010).

146 65 In cephalopods, there are two major life-history dispersal patterns based on egg
147 66 morphology. The “holobenthic” dispersal pattern, utilized by cuttlefishes and most sepiolids
148 67 (Boletzky, 2003), is the production of relatively few, large eggs (>10-12% of adult mantle
149 68 length) resulting in young cephalopods that adopt the benthic mode of life after hatching.
150 69 Conversely, the “merobenthic” dispersal pattern of octopods, loliginids, ommastrephids or
151 70 oceanic squids (Boletzky 2003; Villanueva and Norman 2008) is the production of numerous
152 71 small eggs that hatch into free-swimming, planktonic hatchlings called paralarvae (Young
153 72 and Harman, 1988). These cephalopod paralarvae actively migrate up and down through the
154 73 water column (Passarella and Hopkins, 1991; Shea and Vecchione, 2010), affecting their
155 74 horizontal distribution along the continental shelf (Roura et al. 2016). The horizontal
156 75 distances travelled post-hatching range from less than one kilometre in small bottom-
157 76 dwelling species with no distinct planktonic phase (e.g. sepiolid squids of the subfamily
158 77 Sepiolinae, Boyle and Boletzky, 1996) to probably hundreds of kilometres in pelagic species,
159 78 especially teuthid squids (Roberts, 2005) and octopods (Villanueva and Norman, 2008; Roura
160 79 et al., 2017). The different dispersal patterns of merobenthic paralarvae seem to result from a
161 80 complex interaction between diel vertical behaviour and mesoscale processes, and horizontal
162 81 dispersal is inversely related with the size of the planktonic hatchlings for squids and
163 82 octopods (Villanueva et al. 2016). However, the effect of such dispersal at the genetic level
164
165
166
167
168
169
170
171
172
173
174
175
176
177

178
179
180
181
182
183
184
185
186
187
188
189
190
191
192
193
194
195
196
197
198
199
200
201
202
203
204
205
206
207
208
209
210
211
212
213
214
215
216
217
218
219
220
221
222
223
224
225
226
227
228
229
230
231
232
233
234
235
236

83 remains to be studied in cephalopods, especially in highly dynamic ecosystems like the
84 upwelling regions.

85 The Iberian–Canary current eastern boundary upwelling system (ICC) constitutes one
86 of the four main eastern boundary upwelling systems of the world’s oceans (Barton, 1998).
87 The ICC covers the latitudinal range between 12–43° N and it is divided into five sub-regions
88 (Aristegui et al., 2009). This work is centred in the Galician, Portuguese and the Moroccan
89 sub-regions. The western Iberian sub-region (Galician and Portuguese coasts) is the
90 northernmost part of the ICC. During spring and summer (from March–April to September–
91 October) north-easterly winds predominate in the Iberian basin and mesoscale upwelling
92 filaments develop intermittently in association with irregularities in the coastline like capes
93 (Barton et al., 2001; Joint et al., 2001; Peliz et al., 2005; Cordeiro et al. 2015). The Moroccan
94 sub-region, on the other hand, experiences year-round upwelling varying seasonally,
95 occurrence of extended upwelling filaments, absence of freshwater inputs and massive dust
96 inputs from the adjacent Sahara Desert (Navarro-Pérez and Barton, 1998; Aristegui et al.,
97 2009).

98 Throughout the ICC, coastal upwelling is enhanced in the vicinity of topographic
99 features such as capes, producing filaments of upwelled water that export coastal biomass
100 into the open ocean (Van Camp et al., 1991). In this sense, the cool filaments of Cape Silleiro
101 (NW Iberian Peninsula, 42-43° N) and Cape Ghir (30-31° N; W Morocco) export 4 and 31 x
102 10⁸ kg of organic carbon per year to the adjacent shelf, which corresponds to 20 and 60 % of
103 the phytoplankton primary production (Álvarez-Salgado et al., 2007), respectively. These
104 filaments break the strongly stratified oceanic waters - thermocline and nutricline located at
105 80-100 m depth - creating clear community gradients that contrast with the open ocean
106 communities (Hernández-León et al. 2002). Studies of larval distribution in the ICC suggest
107 that vertical behaviour is one of the contributing mechanisms for retention over the shelf
108 (Queiroga and Blanton, 2004; Peliz et al., 2007; Queiroga et al., 2007), as well as dispersion
109 (Rodríguez et al. 2001, Bécognée et al., 2006).

110 The study of cephalopod paralarvae in the ICC has been mostly carried out in
111 continental shelf waters off the NW Iberian Peninsula (Rocha et al., 1999; González et al.
112 2005; Otero et al. 2008; Olmos-Pérez et al. 2017a, b; Roura et al., 2012, 2016), the
113 Portuguese coast (Moreno et al., 2009) and Mauritanian waters (Faure et al. 2000).
114 Collectively, these studies showed that cephalopod paralarvae are scarce in the zooplankton
115 and positively linked with the upwelling index. Marked changes in their horizontal and
116 vertical distributions suggest that different dispersal patterns may be co-occurring within the

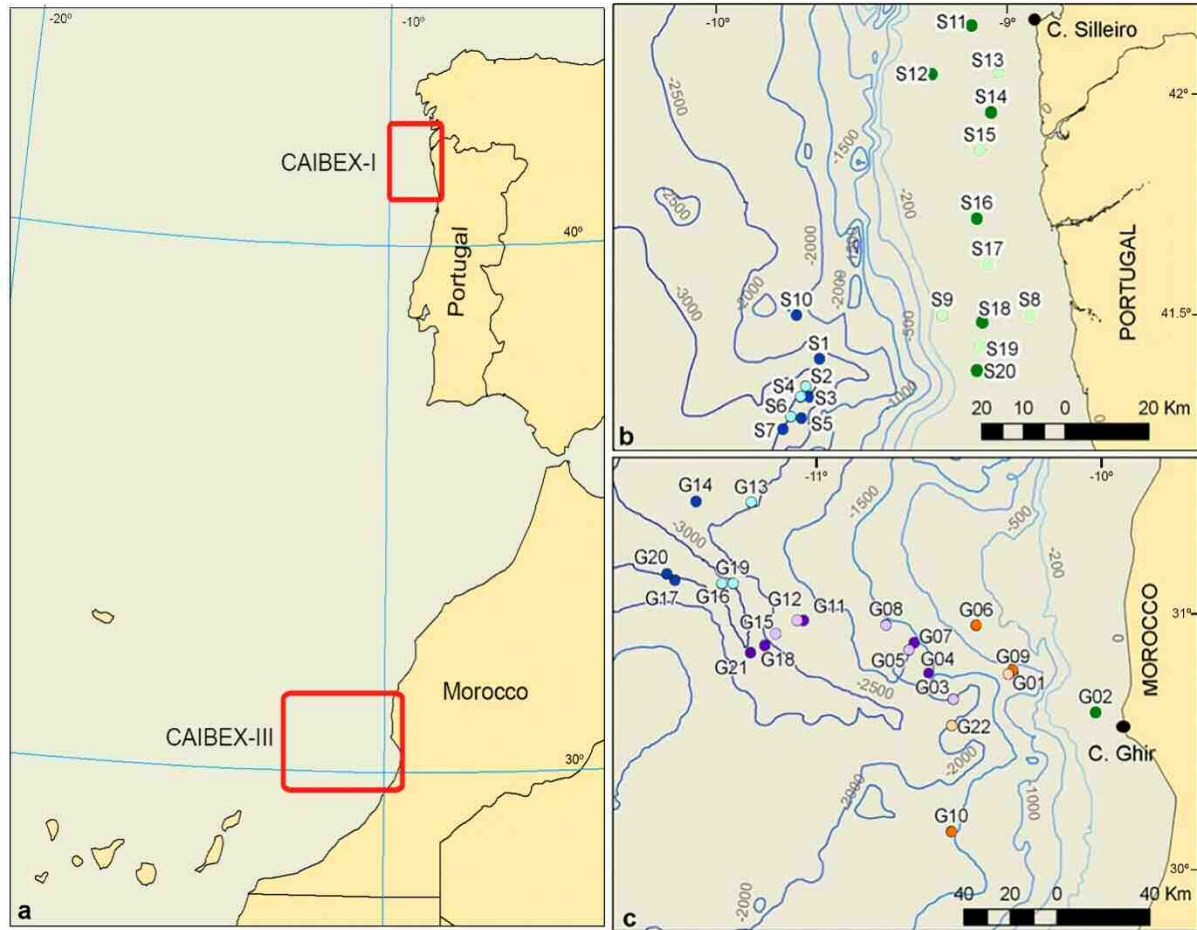
237
238
239 117 cephalopod assemblage of the ICC (Roura et al., 2016). However, difficulties in the
240
241 118 taxonomic identification of most cephalopod paralarvae, especially within the loliginid,
242
243 119 sepiolid and ommastrephid groups (Fernández-Álvarez et al. 2016; Olmos-Pérez et al. 2017a,
244
245 120 b), hamper the accurate interpretation of the different adaptations of the paralarvae to the
246
247 121 oceanography of the ICC. Accordingly, an integrative approach combining oceanographic,
248
249 122 behavioural, morphological and genetic data, known as seascape genetics (Selkoe et al.
250
251 123 2008), is required to help elucidate the spatial ecology of cephalopod paralarvae in the ICC
252
253 124 upwelling ecosystems.

252 125 Here, we explore the interaction between cephalopod paralarvae dispersal patterns and
253
254 126 their genetic diversity / population structure in two contrasting upwelling systems: the
255
256 127 seasonal upwelling of Cape Silleiro (NW Iberian Peninsula) and the permanent upwelling of
257
258 128 Cape Ghir (W Morocco). We identified planktonic cephalopod species via DNA barcoding
259
260 129 and morphological taxonomy, then categorized them into different planktonic dispersal
261
262 130 patterns. Genetic diversity and population structure were then evaluated for each of these
263
264 131 patterns under the oceanographic scenario of the ICC. This integrative approach combining
265
266 132 fine-scale oceanography, morphology, behaviour and genetic data shed light on the effects of
267
268 133 different planktonic dispersal patterns as drivers of genetic diversity and population structure
269
270 134 of cephalopod paralarvae from one of the most important upwelling ecosystems of the world.

269 135 **2. Material and methods**

271
272 136 Data acquisition for this study was carried out under the framework of the
273
274 137 multidisciplinary project “**Canaries–Iberian Marine Ecosystem Exchanges (CAIBEX)**” in
275
276 138 2009 on board the RV *Sarmiento de Gamboa* in the seasonal upwelling system off Cape
277
278 139 Silleiro (41-43°N, CAIBEX-I) from July 7 to 24, and the permanent upwelling system off
279
280 140 Cape Ghir (30-32°N, CAIBEX-III) from August 16 to September 5 (Fig. 1). High-resolution
281
282 141 mapping of the study area determined the oceanographic conditions (temperature, salinity and
283
284 142 Chl-fluorescence) *in situ* using a towed vehicle (SeaSoar) that undulates between the surface
285
286 143 and 400 m depth. The information collected with the SeaSoar, together with real time satellite
287
288 144 images of sea surface temperature (SST) and chlorophyll provided by the Plymouth Marine
289
290 145 Laboratory (NEODAAS), helped to determine the location of upwelled water masses. Once
291
292 146 detected, we aimed to follow the course of the upwelled water mass carrying out three
293
294 147 Lagrangian experiments with an instrumented drifting buoy (IDB), with a length of 100m,
295
148 which was deployed in the core of the upwelled water. The IDB was equipped with Global

296
 297
 298 149 Positioning System and Iridium communication, an Acoustic Doppler Current Profiler
 299 (ADCP) at 2 m to determine current direction and velocities down to 100 m depth, and
 300 150 (ADCP) at 2 m to determine current direction and velocities down to 100 m depth, and
 301 151 temperature sensors at 10, 20, 40, 60, 65, 80 and 100 m depth.



152
 153 **Fig. 1.** a) Schematic map of the Iberian-Canary Upwelling system showing the areas sampled
 154 (red boxes). b) Zooplankton samples collected during CAIBEX-I off the NW coast of the
 155 Iberian Peninsula, around Cape Silleiro (42°N). Samples S1 to S7 correspond to the first
 156 Lagrangian experiment (L1) carried out in the open ocean (blue dots) and S13 to S20 to the
 157 second Lagrangian experiment (L2) carried out over the continental shelf (green dots). c)
 158 Zooplankton samples collected during CAIBEX-III off the NW Africa coast, around Cape
 159 Ghir (30°N, G1-G22). Samples were collected over the continental shelf at night (green dots,
 160 <200 m depth), in an area affected by the upwelling in the open ocean (orange dots, >200 m
 161 depth), following the upwelling filament during the third Lagrangian experiment (violet dots,
 162 L3) and in the open ocean (blue dots). Light / dark colours represent day / night samplings.
 163 See Table A.1 for details.

164 *2.1 Physical and biological sampling*

165 Meteorological conditions resulted in only weak development of upwelling during
 166 CAIBEX-I (Cordeiro et al., 2018). We employed a Lagrangian sampling approach, whereby

355
356
357 167 contrasting water masses were tracked and sampled: 1) oceanic waters over the continental
358 168 slope around 41° 25'N in the relaxation period after a brief upwelling event (L1: July 10-13
360 169 inclusive, samples S1-S7 in blue Fig. 1b); and 2) an incipient coastal upwelling with
362 170 alongshore transport over the shelf was sampled from 42°N to 41°23'N (L2: July 17-20
363 171 inclusive, samples S13-S20, Fig. 1b). Between these two Lagrangian experiments samples
365 172 were collected following a coastal-ocean gradient off the Portuguese coast (S8-S10), as well
366 173 as two samples in the continental shelf of Galicia (S11 and S12). Details of the sampling can
368 174 be found in Table A.1.

370 175 In contrast, constant NE winds during CAIBEX-III allowed the development of a
371 176 strong upwelling filament (Troupin et al., 2012, Sangrá et al., 2015). Once the core of the
372 177 filament was identified the IDB was deployed in the core of the filament, allowing sampling
373 178 of the upwelled water as it was advected from the coast into the ocean during the third
374 179 Lagrangian experiment (L3: August 23-31 inclusive, samples in violet, Fig. 1c). Samples
375 180 were also collected over the shelf (in green, Fig. 1c), in an area affected by the upwelling
376 181 over the continental slope (in orange, Fig. 1c) and in the open ocean (in blue, Fig. 1c), to
377 182 investigate the zooplankton communities surrounding the upwelling filament. Details of the
378 183 sampling can be found in Table A.2.

384 184 A CTD was deployed to 500 m depth in the open ocean and to 10 m above the sea-
385 185 bottom over the continental shelf (<200 m depth) before each zooplankton sampling.
386 186 Mesozooplankton samples were collected close to the IDB both at midnight and midday, to
387 187 identify vertical migrations, with two 750 mm diameter bongo nets equipped with 375 µm
388 188 mesh and a mechanical flow-meter. At a ship speed of 2.5 knots three double-oblique tows
389 189 were carried out at each station over the continental slope (>200m depth): at the deep
390 190 scattering layer (DSL: 500 m), at 100 m and at the surface (0-5 meters). Over the continental
391 191 shelf, samples were collected at 100 m (or 10 m above bottom when shallower) and at the
392 192 surface (0-5 m). The bongo net was first lowered to the desired depth, towed for 30 minutes
393 193 and subsequently hauled up at 0.5 m s⁻¹. Plankton samples were fixed with 96% ethanol and
394 194 stored at -20°C to allow DNA preservation.

403 195 *2.2 Cephalopod identification and barcoding*

406 196 Cephalopod paralarvae were sorted from the samples and were classified to the lowest
407 197 taxonomic level according to Sweeney et al. (1992) and Vecchione et al. (2001). The dorsal
408 198 mantle length (DML), width (W) and total length (TL) of each individual was recorded to the
409

414
415
416 199 nearest 0.1 mm using the software NIS-Elements 3.0 connected to a digital camera (Nikon
417
418 200 DXM1200F) under a binocular microscope (Nikon SMZ800). Furthermore, length of the
419
420 201 right tentacle (TeL) was measured in all decapod paralarvae and the number of suckers per
421
422 202 arm was counted in every *Octopus vulgaris* paralarva (Sweeney et al. 1992).

423 203 The soft body of most cephalopod paralarvae is typically damaged during capture,
424
425 204 especially in oceanic and neritic squid families, hampering the identification process. Three
426
427 205 animals for which the mantle was not present were not measured. Cephalopod paralarvae of
428
429 206 certain groups like oegopsids, loliginids or sepiolids lack morphological characters present in
430
431 207 juveniles and adults - such as photophores, hooks or developed tentacles - thus preventing
432
433 208 their identification to species level (Vecchione et al., 2001). Therefore, genetic identifications
434
435 209 were done with the barcoding gene cytochrome *c* oxidase I (COI) (Hebert et al., 2003), to
436
437 210 allow comparison with the vast database of cephalopod COI sequences available on
438
439 211 GenBank. A project called “Cephalopod paralarvae of the Eastern Atlantic” (CEPAR) was
440
441 212 created in collaboration with the Barcode of Life Data System ([BOLD](#)). A 654-bp region of
442
443 213 the COI gene (Ratnasingham and Hebert, 2007) was sequenced from a small piece of mantle
444
445 214 of each paralarvae. DNA extraction and sequencing were carried out at the University
446
447 215 Guelph, Canada. Prior to obtaining the COI sequences, a visual database from each specimen
448
449 216 was created using dorsal, ventral and lateral photographs. Sequence data are available on the
450
451 217 Barcode of Life Data System (project folder Cephalopod paralarvae of the Eastern Atlantic
452
453 218 “CEPAR”).

449 219 *2.3 Molecular analyses*

451 220 Sequence data were compared against those held in publicly available databases
452
453 221 (BOLD and GenBank) using the BLASTn algorithm. Species level identifications were based
454
455 222 on homologies above 97% and the taxonomic position of those below 97% was assessed
456
457 223 according to their location in a phylogenetic tree (Hebert et al., 2003). Cephalopod sequences
458
459 224 were collapsed into unique haplotypes using DnaSP (Librado and Rozas 2009).
460
461 225 jMODELTESTv.3.8 (Posada, 2008) was used to determine the most appropriate model of
462
463 226 sequence evolution for phylogenetic analyses. The AIC (Akaike information criterion,
464
465 227 Akaike 1974) favored the GTR + G + I model. A maximum likelihood (ML) method of
466
467 228 phylogenetic inference was used to construct a phylogenetic tree including all the different
468
469 229 haplotypes present using *PhyML* v3.1 (Guindon et al. 2010). The strength of support for
470
471 230 internal nodes of the ML phylogeny was measured using 1000 bootstrap replicates. Bayesian
472

473
474
475 231 marginal posterior probabilities were calculated using MrBayes v3.2 (Ronquist and
476
477 232 Huelsenbeck, 2003). Model parameter values were treated as unknown and were estimated.
478
479 233 Random starting trees were used and the analysis was run for 15 million generations,
480
481 234 sampling the Markov chain every 1000 generations. The program Tracer v1.3 (Rambaut and
482
483 235 Drummond, 2003) was used to ensure Markov chains had reached stationarity and to
484
485 236 determine the correct 'burn-in' for the analysis.

486 237 *2.4 Planktonic dispersal patterns: vertical and horizontal distribution*

487

488
489 238 Once the paralarvae were identified, cephalopod assemblages were examined with
490
491 239 multivariate techniques using the software packages PRIMER6 & PERMANOVA+
492
493 240 (Anderson MJ et al., 2008) to identify patterns of distribution. Abundance numbers were
494
495 241 transformed using the function $\log(x + 1)$ to reduce the contribution of highly abundant
496
497 242 species (Clarke and Green, 1988). The Bray-Curtis similarity matrix, which reflects
498
499 243 differences in relative abundance as well as in species composition, was used to calculate the
500
501 244 resemblance matrix among samples. A non-parametric permutational ANOVA
502
503 245 (PERMANOVA) analysis running 999 permutations was used to test for vertical migrations
504
505 246 using a two-factor nested design (factor day/night with two levels and strata, three levels:
506
507 247 surface, 0-100, 0-500 m), and also to test the different horizontal distribution patterns
508
509 248 observed (factor dispersal pattern, three levels: coast, coast-ocean, ocean). Non-parametric
510
511 249 analyses using Mann-Whitney U tests were also conducted to test whether the cephalopod
512
513 250 paralarvae abundances and DML varied significantly between the different habitats sampled.
514
515 251 These analyses were used in order to explore the relationship between the different
516
517 252 planktonic dispersal patterns displayed by the paralarvae in the genetic diversity metrics /
518
519 253 neutrality statistics / haplotype networks.

515 254 *2.5 Genetic diversity and population structure*

516

517
518 255 Ambiguous nucleotides were visually edited using the chromatograms available in
519
520 256 BOLD and using the aligned sequences as a reference. Only sequences >600bp, with no stop
521
522 257 codons, were used in analyses (n = 318). Of these, only the species for which there were more
523
524 258 than five sequences (Goodall-Copestake et al., 2012) were retained for further genetic
525
526 259 analysis. Individual sequences smaller than 620 bp were deleted and the last 20 bp of the 3'
527
528 260 end of all sequences were removed. As a result, a region of 624 bp was retained and used to
529
530 261 calculate genetic diversity and neutrality tests (11 species, n = 285 specimens). DnaSP
531

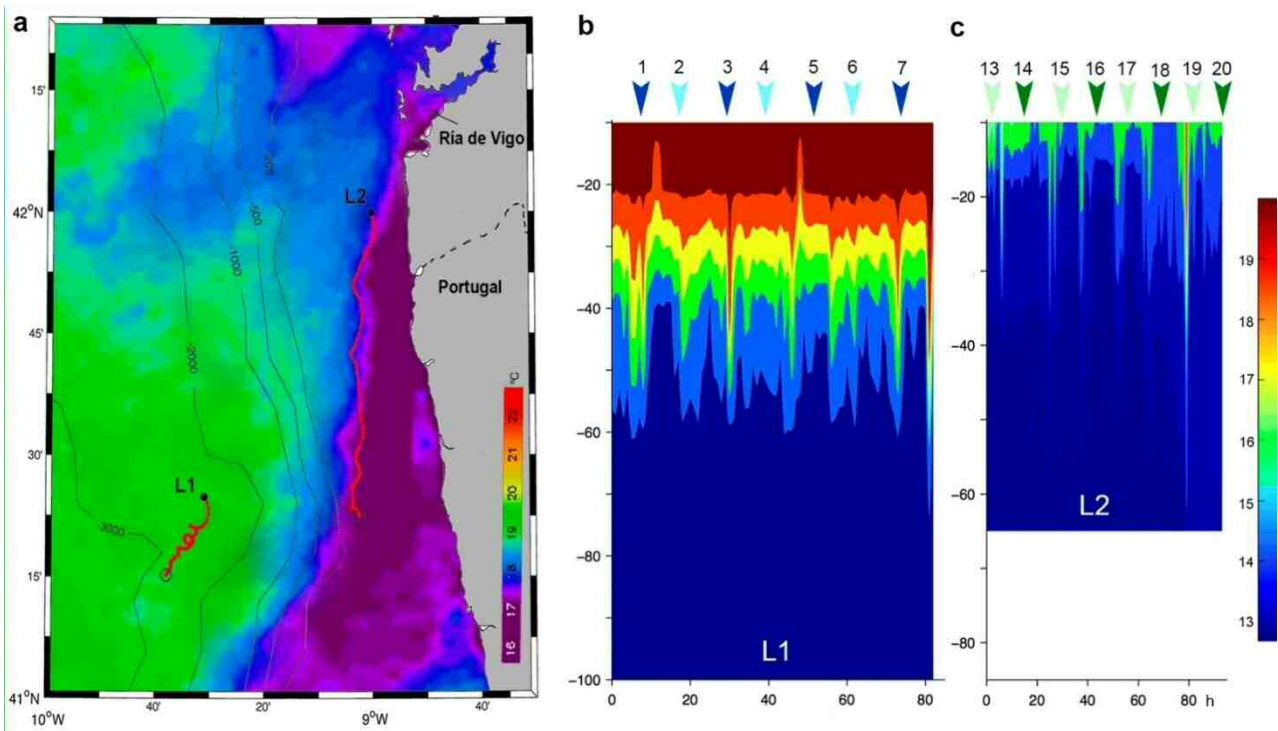
532
533
534
535
536
537
538
539
540
541
542
543
544
545
546
547
548
549
550
551
552
553
554
555
556
557
558
559
560
561
562
563
564
565
566
567
568
569
570
571
572
573
574
575
576
577
578
579
580
581
582
583
584
585
586
587
588
589
590

262 (Librado and Rozas 2009) was used to calculate genetic diversity metrics for each species:
263 haplotype number (H), number of polymorphic sites (S), haplotype diversity (h) and
264 nucleotide diversity per site (π). The validity of the diversity metrics obtained was assessed
265 using the 95th percentile boundary of the function $\pi = 0.0081h^2$ (Goodall-Copestake et al.,
266 2012).

267 Historical fluctuations in population size were investigated using Fu's F_s and
268 Tajima's D to test if the sequences were evolving neutrally or under selection using
269 ARLEQUIN v.3.5.1.2 (Excoffier et al. 2005). A randomization procedure using 1,000
270 samples was used to test the significance of Fu's F_s and Tajima's D. Haplotype networks
271 were generated from COI sequence data using Network v4.6 (Bandelt et al. 1999) to visualize
272 the relationships between the existing haplotypes of a given species under the following
273 criteria: with at least three haplotypes and a minimum of five individuals.

274 Three species (*Octopus vulgaris*, *Alloteuthis subulata* and *A. media*) were present in
275 both surveys (NW Iberian Peninsula and W Morocco, Tables 1 and 2). Pairwise F_{ST}
276 (Excoffier et al. 2005) was calculated between these populations using ARLEQUIN in order
277 to know the extent of genetic differentiation between populations of both upwelling
278 ecosystems.

279



280
281 **Fig. 2.** a) Trajectory of the buoys during the first (L1, July 10-13) and second Lagrangian
282 experiment (L2, July 17-20) in Iberian waters overlaid on sea surface temperature (SST) at

591
592
593
594
595
596
597
598
599
600
601
602
603
604
605
606
607
608
609
610
611
612
613
614
615
616
617
618
619
620
621
622
623
624
625
626
627
628
629
630
631
632
633
634
635
636
637
638
639
640
641
642
643
644
645
646
647
648
649

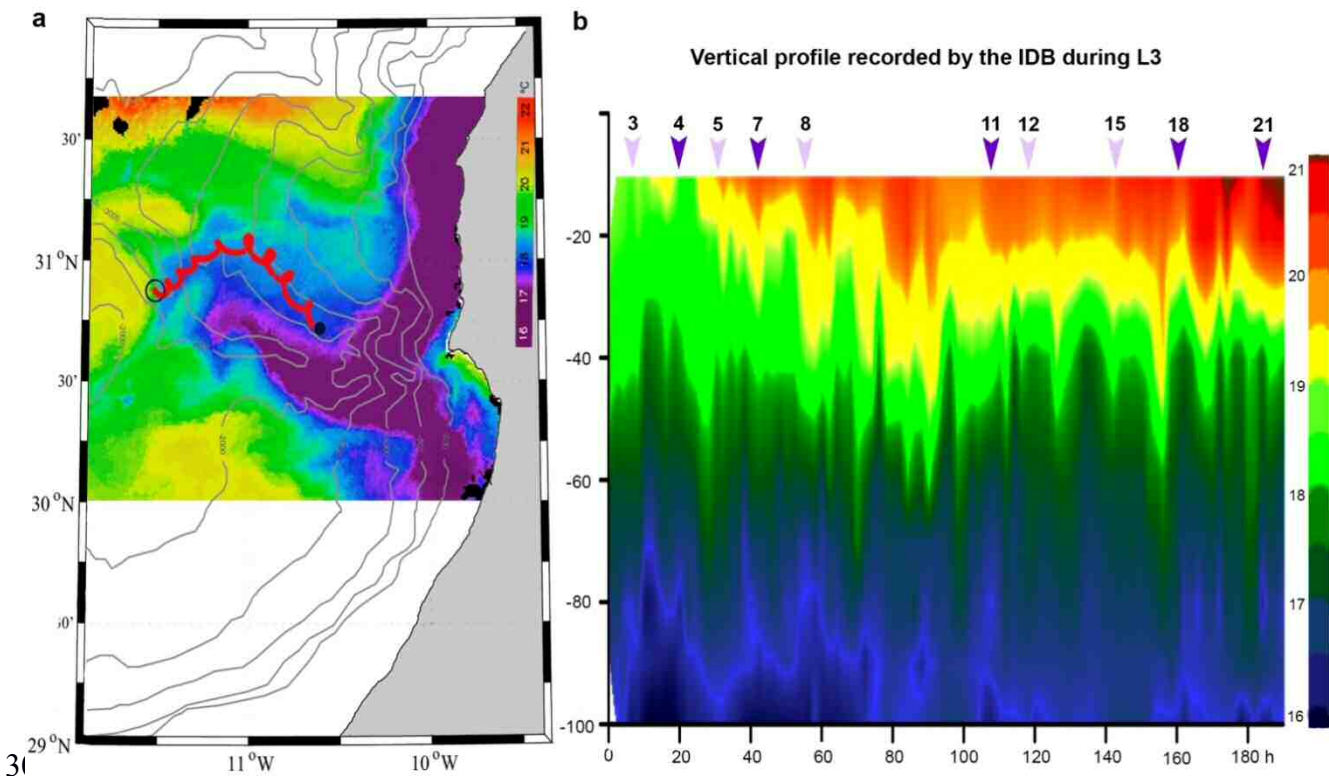
283 the end of L2 (July 20th). Temperature profiles recorded by the physical buoy during b) the
284 first (L1) and c) second Lagrangian experiment (L2) of CAIBEX-I. Zooplankton samplings
285 are shown with dark colour indicating night sampling.

286 3. Results

287 3.1 Oceanographic context

288 CAIBEX-I: The first Lagrangian experiment (L1, Fig. 2) was conducted over the
289 continental slope of Portugal with depths ranging between 2,667 and 3,105 m, under
290 northerly winds during the first half of the experiment and southerly winds onward. Wind
291 velocities were low and the IDB was displaced slowly south-westward (Fig. 2a). Vertical
292 temperature profiles recorded by the thermistor chain of the IDB showed that the water
293 column was strongly stratified with a small increase in temperature when the winds shifted
294 (Fig. 2b).

295 Experiment L2 was carried over the Iberian shelf of Galicia and Portugal between 90
296 and 150 m depth). Strong northerly winds during the first three days transported the IDB
297 equatorward and slightly offshore, but weakening and then reversal of the wind to southerly
298 in the last two days slowed and almost stopped the drift of the IDB (Fig. 2a). The temperature
299 recorded by the IDB (Fig. 2c) shows the presence of upwelled water over the shelf.



650
651
652
653
654
655
656
657
658
659
660
661
662
663
664
665
666
667
668
669
670
671
672
673
674
675
676
677
678
679
680
681
682
683
684
685
686
687
688
689
690
691
692
693
694
695
696
697
698
699
700
701
702
703
704
705
706
707
708

302 **Fig. 3.** a) Trajectory of the buoy during the third Lagrangian experiment in Moroccan waters
 303 (L3, August 23-31) overlaid on sea surface temperature (SST) at the beginning of the
 304 experiment (August 24th). b) Temperature profile recorded by the physical buoy during
 305 CAIBEX-III. Zooplankton samplings are shown, with dark colour indicating night sampling.

306 CAIBEX-III: Experiment L3 was carried out in waters 1540 – 3020 m deep. The IDB
 307 drifted north-westward offshore by 172 km within the core of an upwelling filament - 60 m
 308 deep and 25 km wide - and then shifted to the southwest (Fig. 3a). As recorded by the IDB
 309 thermistor chain, the drifters began in cold upwelled waters that were progressively heated by
 310 the warm and stratified surrounding oceanic waters (Fig. 3b). The temperature showed
 311 regular oscillation of the isotherms, at the local inertial period.

312 **3.2 Cephalopod paralarvae identification**

313 One adult (9.77 mm DML) and 134 cephalopod paralarvae and juveniles (ranging
 314 from 1.49 to 8.17 mm DML) were found in 48 samples from CAIBEX I (Table A.3). Four
 315 families were present. The most abundant were the octopods, with 99 paralarvae,
 316 representing 73.3% of the paralarvae collected, followed by the loliginids (16 paralarvae,
 317 11.9%), ommastrephids (15 rhynchoteuthion, 11.1%) and sepiolids (four juveniles and one
 318 adult, 3.7%). COI sequences were obtained for 124 paralarvae from a total of 135 individuals
 319 (91.9%, Fig. 4) with similarities higher than 99% against sequences present on GenBank
 320 (Table 1). All the octopods sequenced were *Octopus vulgaris* (88 paralarvae). Eleven octopod
 321 paralarvae did not amplify correctly, but were also identified as *O. vulgaris* based on
 322 similarities of morphology and chromatophore pattern. *Octopus vulgaris* was the only species
 323 that was found in both coastal and oceanic samples. All loliginid paralarvae were identified
 324 and were assigned to the following species: 10 *Alloteuthis subulata*, four *A. media* and two
 325 *Loligo vulgaris*. Sequence data were obtained for all ommastrephid paralarvae: 12 were
 326 identified as *Todaropsis eblanae*, two were *Todarodes sagittatus* and one was *Illex coindetii*.
 327 Finally, all five sepiolids were identified as *Sepiola tridens*.

Planktonic dispersal pattern	Species	Shelf (L2)				Ocean (L1)					
		day		night		day			night		
		100	5	100	5	500	100	5	500	100	5
Coastal	<i>Alloteuthis media</i>	2		1	1						
	<i>Alloteuthis subulata</i>			7	3						
	<i>Loligo vulgaris</i>			2							
	<i>Sepiola tridens</i>			5							
	<i>Illex coindetii</i>				1						
	<i>Todaropsis eblanae</i>	8		3	1						
Coast-ocean	<i>Octopus vulgaris</i>	7	4	12	28	5	4		14	10	15
Oceanic	<i>Todarodes sagittatus</i>						1			1	

	Ommastrephidae*										2		1			1							
	Onychoteuthidae*																				3		
	<i>Pyrotheuthis margaritifera</i>										1	1		1					4		2	2	
	Pyrotheutidae																		1			1*	1
	Sepiolidae*					1																	

Table 2. Cephalopod paralarvae present in the different locations sampled off Cape Ghir (31°N, W Morocco) during CAIBEX-III. Asterisk indicates species/taxa that lack genetic identification. Values 5, 100 and 500 indicate tow depth.

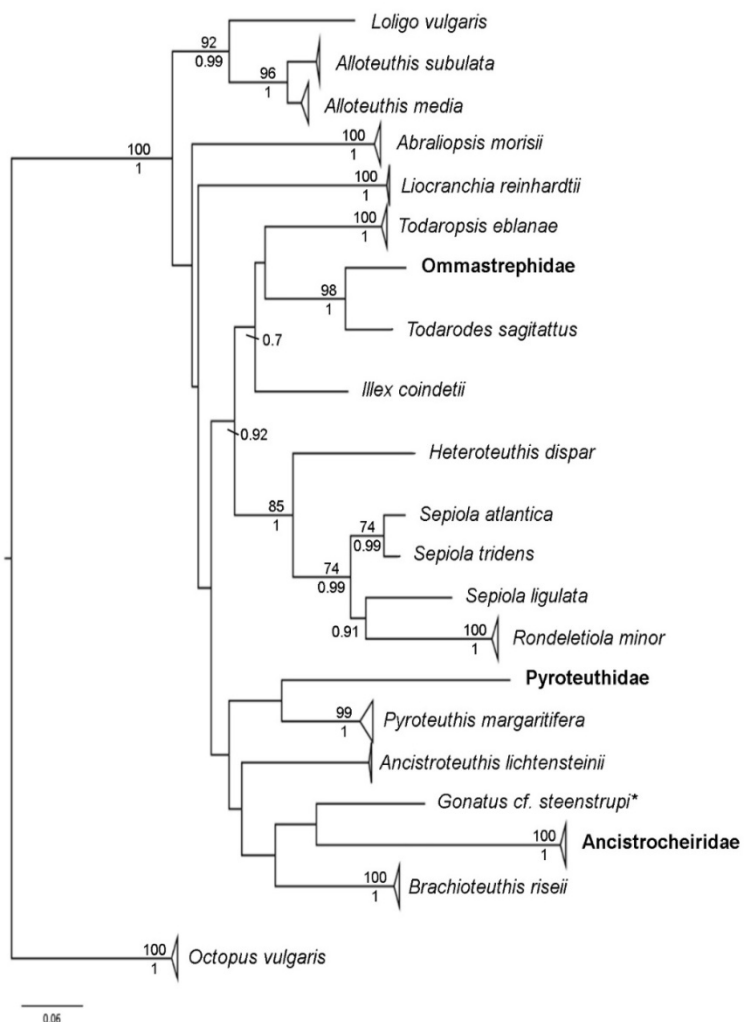
The oceanic families were present in the filament and oceanic samples. These included by order of abundance: onychoteuthids (n = 21, 8.5%) of which 18 were *Ancistroteuthis lichtensteini* (100% homology); brachioteuthids (n = 18, 7.3%) represented by *Brachioteuthis riisei* (100% homology); pyroteuthids (n = 14, 5.7%) with eleven sequences corresponding to *Pyroteuthis margaritifera* (99% homology) and two undefined Pyroteuthidae with the closest match being *P. addolux/Pterygioteuthis* (90%); ancistrocheirids (n = 12, 4.9%), with ten sequences showing homologies below 90% with *Ancistrocheirus lesueurii*; enoploteuthids (n = 11, 4.5%) eight identified as *Abraliopsis pfefferi* (99%), which is a junior synonym of *A. morisii* (Ropert and Jereb, 2010); cranchids (n = 3, 1.2%) two of which had 100% homologies with *Liocranchia reinhardti*; a gonatid with the closest match being with *Gonatopsis japonicus* (88.3%), however based on morphological characters and its distribution we identified it as *Gonatus steenstrupi*; an unidentified oegopsid (0.4%); and an adult mastigoteuthid morphologically identified as *Mastigopsis hjorti*.

The taxonomic status of the unknown ancistrocheirid paralarvae (n = 10) found on CAIBEX-III was further explored according to their position in a Kimura 2 parameter distance model tree constructed in BOLD (Fig. A.1). Our samples were grouped in a clade including two other ancistrocheirids, being 88.6 - 89.3% similar to seven sequences from an undefined Ancistrocheiridae collected in South Africa (private sequences) and 86.3 - 87.2% similar to available sequences of *Ancistrocheirus lesueurii* specimens from the Indo-Pacific. Accordingly, we suggest that the paralarvae collected off Morocco belong to *Ancistrocheirus* but represent a new species within the monotypic family Ancistrocheiridae. Our genetic results suggest that our species is different from that known from the Indo-Pacific - named *A. lesueurii* (d'Orbigny [in Férussac & d'Orbigny], 1842) -, therefore we would have to assign the ancistrocheirid paralarvae found off the coast of Morocco to the former species described

827
828
829
830
831
832
833
834
835
836
837
838
839
840
841
842
843
844
845
846
847
848
849
850
851
852
853
854
855
856
857
858
859
860
861
862
863
864
865
866
867
868
869
870
871
872
873
874
875
876
877
878
879
880
881
882
883
884
885

381 by V erany in the Mediterranean, which is *Ancistrocheirus alessandrini* (V erany, 1847), as
382 indicated in the discussion section.

383
384



385
386 **Fig. 4.** Maximum likelihood (ML) phylogenetic tree showing the different cephalopod
387 paralarvae species identified in the surveys CAIBEX-I and III. Species marked with an
388 asterisk are not present in the genetic database but were assigned to a species based on their
389 morphology. Species in bold represent those cephalopod paralarvae that are not present in the
390 genetic database and were not possible to identify morphologically. Bootstrap values >60
391 after 1,000 replications and posterior probabilities >0.6 are shown above and below the
392 nodes, respectively.

393
394 *3.3 Planktonic dispersal patterns and vertical behaviour*

395 The spatial distribution of cephalopod paralarvae in both upwelling systems showed a
396 similar trend (Fig. 5) and three different planktonic dispersal patterns were identified:

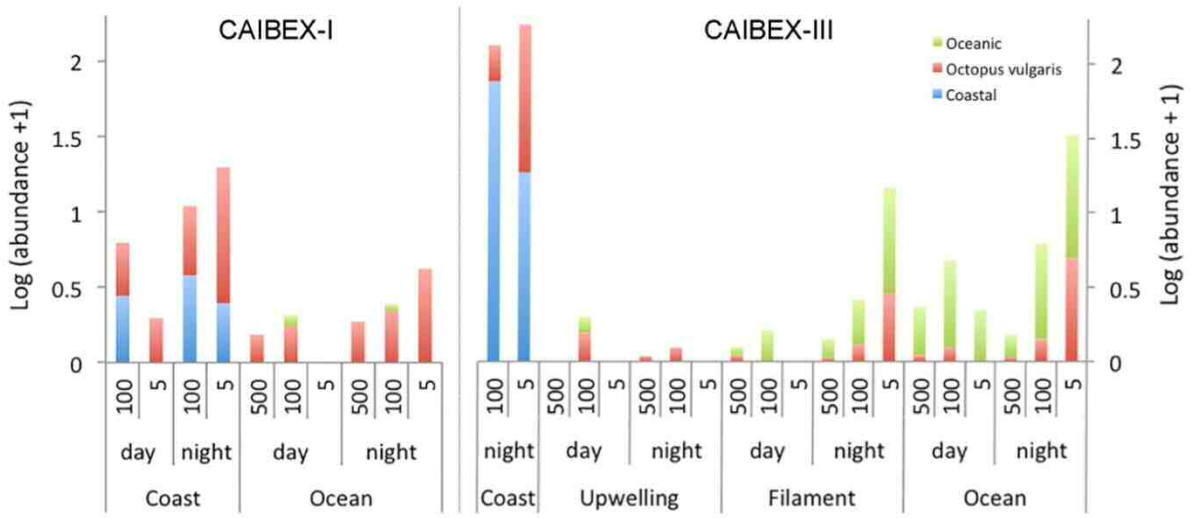
886
887
888
889
890
891
892
893
894
895
896
897
898
899
900
901
902
903
904
905
906
907
908
909
910
911
912
913
914
915
916
917
918
919
920
921
922
923
924
925
926
927
928
929
930
931
932
933
934
935
936
937
938
939
940
941
942
943
944

397 - A “coastal” dispersal pattern followed by the loliginids *Alloteuthis media*, *A.*
398 *subulata* and *L. vulgaris*; the sepiolids *Sepiola tridens*, *S. atlantica*, *S. ligulata* and
399 *Rondeletiola minor*; and the ommastrephids *Todaropsis eblanae* and *Illex coindetii*. The wide
400 size range measured - especially in loliginids and sepiolids (Table A.3) - provides evidence
401 for the retention of these paralarvae between the coast and the limit of the continental shelf
402 (200 m depth).

403 - A “coastal-oceanic” dispersal pattern followed exclusively by *O. vulgaris* in both
404 upwelling ecosystems, with early stages (3 suckers per arm) found over the continental shelf;
405 while advanced stages (between 3 to 15 suckers per arm) were found beyond the continental
406 shelf in the upwelling filament and the ocean (Tables 1-3, Fig. 5). A positive relationship was
407 recorded between distance from shore and *O. vulgaris* body size, showing that the paralarvae
408 are growing as they are transported by upwelling filaments into the ocean (Fig. 6).

409 - An “oceanic” dispersal pattern followed by ommastrephids like *Todarodes*
410 *sagitattus* in CAIBEX-I (Table1), oceanic sepiolids like *Heteroteuthis dispar* as well as true
411 mesopelagic squids only found in samples with bottom depths greater than 2,000 m off the
412 coast of Morocco (Table 2, Fig. 5). This mesopelagic squid assemblage is composed by
413 *Brachioteuthis riseii*, *Ancistroteuthis lichtensteinii*, *Abraliopsis morisii*, *Pyroteuthis*
414 *margaritifera*, *Ancistrocheirus alessandrini*, *Liocranchia reinhardtii*, *Gonatus steenstrupii*,
415 *Mastigopsis hjorti* and other undefined pyroteuthids, ommastrephids and oegopsids. Most of
416 these species include a wide range of sizes (Table A.3) and their abundance increases
417 progressively with the distance from the coast, especially in the filament and oceanic areas
418 (Fig. 5).

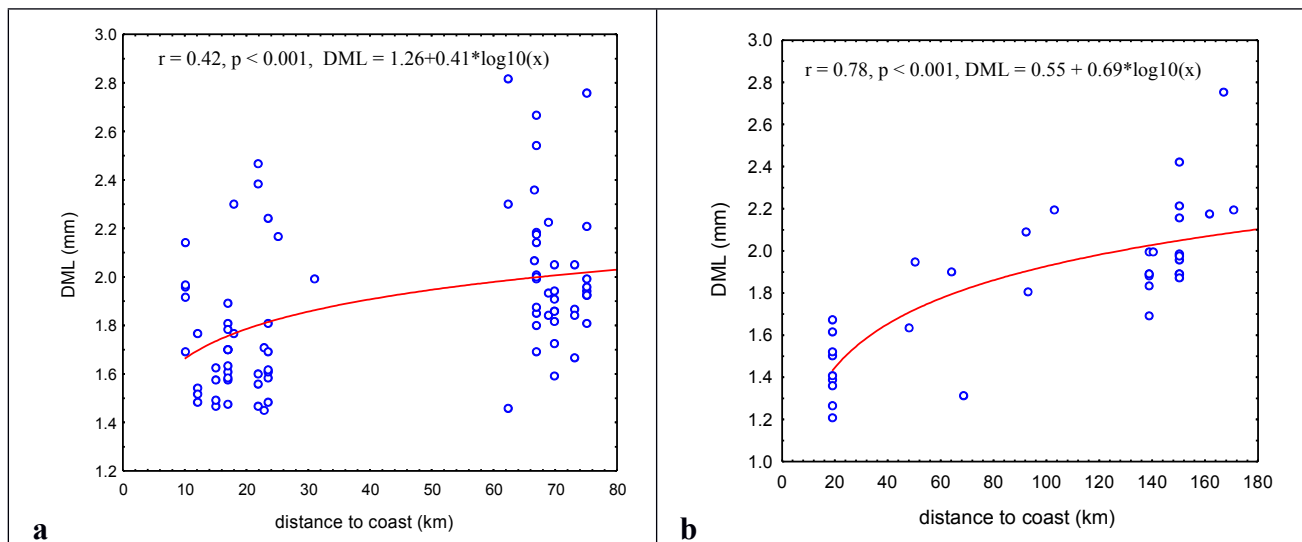
419



420

945
 946
 947 421 **Fig. 5.** Spatio-temporal distribution of the cephalopod paralarvae found in CAIBEX-I and III
 948 422 grouped according to the planktonic dispersal pattern and the location sampled. Paralarval
 949 423 abundances are calculated as individuals per 1,000 m³ and represented as log (abundance +
 950 424 1).
 951
 952
 953
 954 425

955 426 PERMANOVA analyses showed differences in the vertical behaviour between day
 956 427 and night for all planktonic dispersal patterns (p<0.001). Overall, cephalopod paralarvae were
 957 428 more abundant during the night, with increased abundance at the surface (p<0.01) decreasing
 958 429 gradually with depth (Fig. 5). In contrast, cephalopod paralarvae are absent from the surface
 960 430 in both coastal and oceanic environments during the day (with the exception of four very
 961 431 small octopus paralarvae collected in CAIBEX-I, Table 1, and two oceanic paralarvae
 962 432 collected in CAIBEX-III, Table 2) with the highest abundances recorded at 100 m.
 963
 964
 965
 966
 967
 968 434



969
 970
 971
 972
 973
 974
 975
 976
 977
 978
 979
 980
 981
 982
 983
 984 435
 985
 986 436 **Fig. 6.** Scatter plot showing the relationship between the distance to the coast (km) and the
 987 437 DML (mm) in *Octopus vulgaris* paralarvae collected during CAIBEX-I (a) and III (b).
 988
 989
 990
 991 439

992 440

Survey	Location	n	DML (mm)	Sucker n°	Depth (m)	Distance to coast (km)
CAIBEX I	Coast	51	1.75 ± 0.27	3 - 4	62 - 147	10 - 31
	Ocean	48	2.02 ± 0.28	3 - 5	1,940 - 3,105	62 - 75
CAIBEX III	Coast	9	1.44 ± 0.15	3	88 - 90	19
	Upwelling	4	1.66 ± 0.26	3 - 4	787 - 2,328	48 - 93
	Filament	10	1.96 ± 0.15	3 - 6	1,526 - 2,720	50 - 162
	Ocean	12	2.13 ± 0.33	4 - 15	2,418 - 3,110	140 - 171

999 444
 1000
 1001
 1002
 1003

1004
1005
1006 445 **Table 3.** Average dorsal mantle length range (DML \pm standard deviation) and sucker counts
1007 446 of *Octopus vulgaris* paralarvae found at the different locations sampled in CAIBEX I and III
1008 447 surveys. The range of depths and distances to coast is shown for the locations sampled.
1009 448
1010
1011

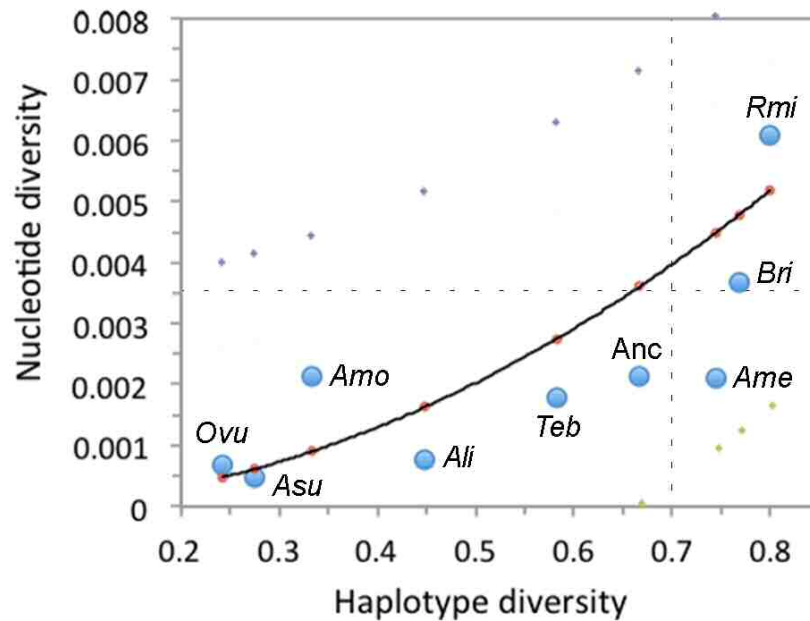
1012
1013 449 *3.4 Cephalopod paralarvae genetic diversity and population structure*
1014

1015 450 Only 11 out of 21 different putative species identified with COI gene met the criteria
1016 451 to study their genetic diversity (at least five individuals per species and \geq three haplotypes).
1017 452 The number of haplotypes per species ranged between 1 and 22, with a positive relationship
1018 453 with the number of sequences analysed ($R^2 = 0.63$). No diversity metrics could be calculated
1019 454 for *Sepiolo tridens* ($n = 5$) and *Pyroteuthis margaritifera* ($n = 6$) as all sequences represented
1020 455 the same haplotype. Nucleotide diversity for the 624 bp data set ranged from 0.00046 to
1021 456 0.00609 and haplotype diversity ranged from 0.243 to 0.8 (Table 4). Values for these COI
1022 457 diversity metrics were evenly distributed within the 95th percentile boundaries around the
1023 458 fitted model that relates these two variables (Fig. 7), thus indicating that there were no
1024 459 outliers despite the low number of sequences analysed for some species. According to the
1025 460 reference values calculated by Goodall-Copestake et al. (2012) as a cut-off for qualitative
1026 461 descriptions, three cephalopod species had high haplotype diversity ($h > 0.7013$): the coastal
1027 462 species *Alloteuthis media* and *Rondeletiola minor*, and the oceanic one *Brachioteuthis riseii*
1028 463 (Table 4). Conversely, only two species had high nucleotide diversity ($\pi > 0.00356$): *R. minor*
1029 464 and *B. riseii*. Significant differences ($p < 0.001$) were found among planktonic dispersal
1030 465 patterns, with high nucleotide and haplotype diversity for both coastal and oceanic patterns
1031 466 and low diversity for the coastal-oceanic distribution displayed by *Octopus vulgaris* (Fig. 8).
1032
1033
1034
1035

1036 467 Significant deviance from neutrality was revealed in both statistics analysed
1037 468 (Tajimas's D and Fu's Fs) for *Alloteuthis media*, *A. subulata* and *O. vulgaris* (Table 4). These
1038 469 species showed negative values of both statistics, suggesting that the sequences analysed
1039 470 were evolving under a non-random process, likely a recent population expansion (e.g. after a
1040 471 bottleneck or a selective sweep). A negative value for Fu's Fs provides evidence for an
1041 472 excess number of alleles relative to simulations, as would be expected from a recent
1042 473 population expansion or from genetic hitchhiking. Negative Tajima's D implies an excess of
1043 474 low frequency polymorphisms relative to expectation, also pointing in the direction of
1044 475 population size expansion and /or purifying selection. Remarkably, these three species were
1045 476 the only ones that were captured in both upwelling regions (Tables 1 and 2) and the observed
1046 477 significant deviances from neutrality tests could indicate that the sampled specimens belong
1047
1048
1049
1050
1051
1052
1053
1054
1055
1056
1057
1058
1059
1060
1061
1062

1063
1064
1065
1066
1067
1068
1069
1070
1071
1072
1073
1074
1075
1076
1077
1078
1079
1080
1081
1082
1083
1084
1085
1086
1087
1088
1089
1090
1091
1092
1093
1094
1095
1096
1097
1098
1099
1100
1101
1102
1103
1104
1105
1106
1107
1108
1109
1110
1111
1112
1113
1114
1115
1116
1117
1118
1119
1120
1121

478 to different populations. However, pairwise F_{st} tests revealed that such population structure
479 was not supported for *O. vulgaris* and *A. subulata* ($p > 0.05$), despite being separated by more
480 than 2,000 km. Although *A. media* was also captured in both upwelling ecosystems the four
481 sequences obtained from NW Iberian Peninsula were smaller than 624 bp and were not
482 included in the analysis.



484
485 **Fig. 7.** Nucleotide-haplotype diversity relationship obtained from a homologous 624-base
486 region of COI for the following cephalopod paralarvae: Anc, *Ancistrocheirus alessandrini*;
487 Ali, *Ancistroteuthis lichtensteinii*; Ame, *Alloteuthis media*; Amo, *Abraliopsis morisii*; Asu,
488 *Alloteuthis subulata*; Bri, *Brachioteuthis riseii*; Ovu, *Octopus vulgaris*; Rmi, *Rondeletiola*
489 *minor*; Teb, *Todaropsis eblanae*. Red dots represent the expected nucleotide diversity values
490 (π , with the 95th percentiles above and below) according to the model $\pi=0.0081h^2$ (Goodall-
491 Copestake et al. 2012), based on our measured haplotype diversity (h). Dashed lines represent
492 the median values of nucleotide and haplotype diversity obtained by Goodall-Copestake et al.
493 (2012) that are used to represent low and high diversity.

495 The haplotype networks calculated from the common 624-data set are shown in Fig. 9
496 grouped according to their dispersal pattern. The star-shaped haplotype network found in the
497 school of *A. media* (Fig. 9a) is in agreement with the recent population expansion event
498 detected with neutrality tests, where many unique haplotypes ($n = 16$) radiate from few
499 common haplotypes. The same star-shaped pattern is observed in *O. vulgaris* (Fig. 9b), but
500 with fewer number of different haplotypes despite greater number of individuals included (n
501 = 115). The case of *A. subulata*, for which there were fewer specimens analysed ($n = 14$),

1122
 1123
 1124
 1125
 1126
 1127
 1128
 1129
 1130
 1131
 1132
 1133
 1134
 1135
 1136
 1137
 1138
 1139
 1140
 1141
 1142
 1143
 1144
 1145
 1146
 1147
 1148
 1149
 1150
 1151
 1152
 1153
 1154
 1155
 1156
 1157
 1158
 1159
 1160
 1161
 1162
 1163
 1164
 1165
 1166
 1167
 1168
 1169
 1170
 1171
 1172
 1173
 1174
 1175
 1176
 1177
 1178
 1179
 1180

Dispersal pattern	Species	n	H	S	Hd	π	Tajima's D	Fu's Fs
Coastal	<i>Alloteuthis media</i>	94	22	24	0.745 ± 0.037	0.00209 ± 0.00024	- 2.16 (p=0.002)	- 19.94 (p<0.001)
	<i>A. subulata</i>	14	3	2	0.275 ± 0.148	0.00046 ± 0.00026	- 1.48 (p=0.048)	- 1.47 (p=0.007)
	<i>Todaropsis eblanae</i>	9	4	3	0.583 ± 0.183	0.00178 ± 0.00063	0.03 (p=0.577)	- 0.82 (p=0.158)
	<i>Sepiola tridens</i>	5	1	0	0	0	-	-
	<i>Rondeletiola minor</i>	5	3	7	0.800 ± 0.164	0.00609 ± 0.00133	0.91 (p=0.803)	1.78 (p=0.799)
Coastal - Oceanic	<i>Octopus vulgaris</i>	115	9	11	0.243 ± 0.053	0.00068 ± 0.00021	- 2.03 (p=0.001)	- 7.36 (p=0.001)
	<i>Brachioteuthis riisei</i>	14	6	8	0.769 ± 0.089	0.00368 ± 0.00048	- 0.33 (p=0.423)	- 0.60 (p=0.353)
Oceanic	<i>Ancistroteuthis lichtensteinii</i>	15	3	2	0.448 ± 0.134	0.00076 ± 0.00025	- 0.59 (p=0.299)	- 0.52 (p=0.261)
	<i>Pyroteuthis margaritifera</i> *	6	1	0	0	0	-	-
	<i>Abraliopsis morisii</i>	6	2	4	0.333 ± 0.215	0.00214 ± 0.00138	- 1.29 (p=0.064)	2.14 (p=0.82)
	<i>Ancistrocheirus alessandrinii</i>	7	3	3	0.667 ± 0.16	0.00214 ± 0.00051	0.40 (p=0.688)	0.54 (p=0.543)

502 also showed evidence of non-neutral processes, which were not evident from the calculated
 503 haplotype network (Fig. 9a). Contrarily, in other haplotype network topologies – such as
 504 *Rondeletiola minor*, *Brachioteuthis riisei* and the new species of Ancistrocheiridae (Figs. 9a,
 505 c) - the observed haplotypes seem to be derived from an ancestral haplotype that may have
 506 not been sampled or may have been lost because of a recent population bottleneck.

507
 508 **Table 4.** Genetic diversity calculated from a fragment of the COI gene of 624 bp from those
 509 cephalopod paralarvae (n) with at least 5 sequences per species: number of haplotypes (H),
 510 number of polymorphic sites (S), haplotype diversity ($h \pm SD$), nucleotide diversity per site (π
 511 $\pm SD$). Tajima's D and Fu's Fs tests of neutrality were calculated and its significance was
 512 obtained after 1,000 simulated samples (p-value).

513 4. Discussion

514 The multidisciplinary seascape approach sheds light on the processes that shape
 515 genetic diversity/population structure of planktonic cephalopod paralarvae according to their
 516 different life dispersal patterns. We used COI to identify cephalopod paralarvae and to
 517 explore the genetic diversity within species and among planktonic dispersal patterns. This
 518 barcoding approach revealed 21 different species and was particularly useful to identify early
 519 stages of cephalopod paralarvae <4 mm, where conspicuous taxonomic features were absent
 520 or not yet formed and would have limited the taxonomic identification above family level,
 521 especially within the families Loliginidae, Sepiolidae, Enoploteuthidae and Ommastrephidae.

522 4.1 Cephalopod paralarval diversity

523 Eight cephalopod species were identified in the upwelling ecosystems of NW Iberian
 524 Peninsula (CAIBEX-I) and 16 in W Morocco (CAIBEX-III), showing a marked latitudinal
 525 change in the cephalopod assemblage. The main reasons for the absence of tropical and sub-

1181
1182
1183
1184
1185
1186
1187
1188
1189
1190
1191
1192
1193
1194
1195
1196
1197
1198
1199
1200
1201
1202
1203
1204
1205
1206
1207
1208
1209
1210
1211
1212
1213
1214
1215
1216
1217
1218
1219
1220
1221
1222
1223
1224
1225
1226
1227
1228
1229
1230
1231
1232
1233
1234
1235
1236
1237
1238
1239

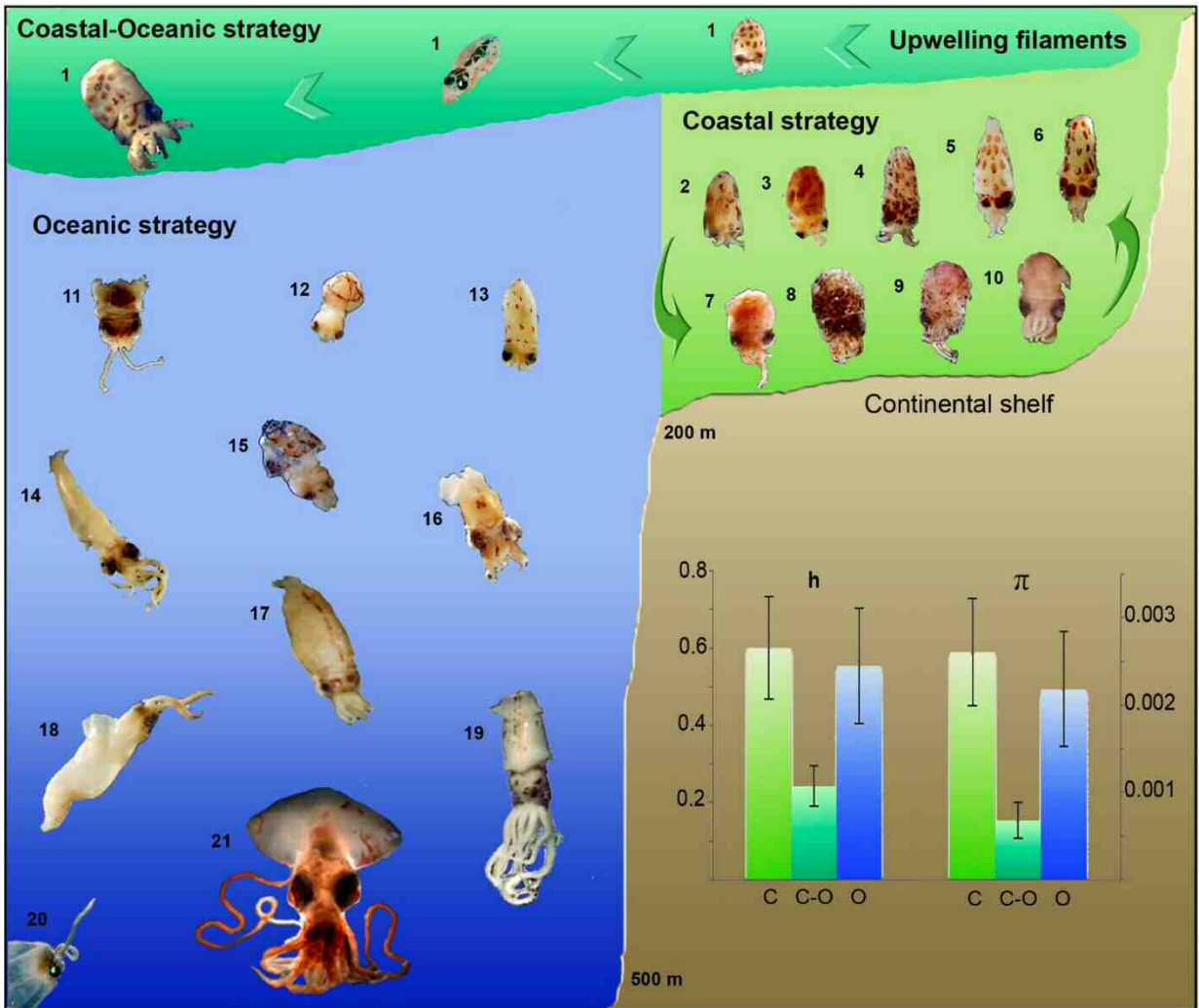
526 tropical oceanic cephalopod paralarvae during CAIBEX-I (between 41°15'N and 42°6'N)
527 may be the persistent fronts between the shelf and deep waters close to Cape S. Vicente
528 (37°N), which represent a temperature boundary to poleward dispersion (Peliz et al., 2005;
529 Moreno et al., 2009). In fact, Moreno et al. (2009) found only 22 oceanic paralarvae
530 belonging to four different species north of 40°N in 57 surveys carried from 1986 to 2004. Of
531 these, three were tropical species (belonging to two families; Onychoteuthidae and
532 Mastigoteuthidae) found from January to June and *Teuthowenia megalops*, a sub-Arctic and
533 northern temperate Atlantic species found only in winter months (Collins et al., 2001;
534 Moreno et al., 2009). The only record of a tropical species found north of 40°N is of
535 *Brachioteuthis riseii* paralarvae, a cosmopolitan oceanic species found west of the British
536 Isles between March and July (Collins et al., 2001). Accordingly, the absence of oceanic
537 squid paralarvae during CAIBEX-I was a normal result for the area sampled, which is north
538 of their spawning grounds off southern Portugal, and for the month sampled (July), which is
539 too warm for northern temperate Atlantic species.

540 An unexpected finding of CAIBEX-I paralarval barcoding was the presence of
541 *Sepiola tridens* in samples collected off the Portuguese shelf at night in bottom depths
542 ranging from 100 to 148 m. This species, described from specimens collected in the North
543 Sea, Ireland and NW Spain (de Heij and Goud, 2010), has been recently identified in the Ría
544 de Vigo (Olmos-Pérez et al., 2017). Its presence off the Portuguese coast increases the
545 southern limit of its range to 41°23'N. Morphologically, juveniles of this species cannot be
546 separated with certainty from *S. atlantica*; however, *S. tridens* inhabits deeper waters
547 (average depth 81.8m) than *S. atlantica* (average depth 37.4 m, de Heij and Goud, 2010).

548 Analyses based on morphological characters and identification guides (González et
549 al. 2010) indicated that the loliginid paralarvae present in the Ría de Vigo was *Loligo*
550 *vulgaris*. However, this deduction is likely incorrect, because in our genetic sampling off the
551 coast of the NW Iberian Peninsula only 2 out of 16 loliginids were *L. vulgaris* (Table 1).
552 Moreover, in a recent study carried out by Olmos-Pérez (2018) in the Ría de Vigo between
553 2012 and 2014, the most abundant loliginid was *A. media* (57.5%), followed by *A. subulata*
554 (21%) and *L. vulgaris* (14.5%). Although adult specimens of *Alloteuthis* can be differentiated
555 by the size of the central club sucker (Anderson FG et al. 2008), this characteristic is not yet
556 present in paralarvae. They can be identified only on the basis of the chromatophore pattern
557 of fresh individuals (Sweeney et al. 1992), which is a delicate character lost in fixed
558 specimens. Our morphological study showed *A. subulata* paralarvae to be significantly bigger
559 for all the body lengths measured than *A. media* (Table A.3). Recent morphological studies

1240
1241
1242
1243
1244
1245
1246
1247
1248
1249
1250
1251
1252
1253
1254
1255
1256
1257
1258
1259
1260
1261
1262
1263
1264
1265
1266
1267
1268
1269
1270
1271
1272
1273
1274
1275
1276
1277
1278
1279
1280
1281
1282
1283
1284
1285
1286
1287
1288
1289
1290
1291
1292
1293
1294
1295
1296
1297
1298

560 found that tentacle length may be a good character to discriminate between both *Alloteuthis*
561 species (Olmos-Pérez, 2018).
562



563
564 **Fig. 8.** Cephalopod paralarvae and their different planktonic dispersal patterns in the Iberian-
565 Canary Upwelling System: coastal (C, light green); coastal-oceanic (C-O, dark green) and
566 oceanic (O, blue). Average haplotype (h) and nucleotide (π) diversity estimated for the
567 different planktonic dispersal patterns. Bars indicate standard deviations. 1 *Octopus vulgaris*;
568 2 *Illex coindetii*; 3 *Todaropsis eblanae*; 4 *Loligo vulgaris*; 5 *Alloteuthis subulata*; 6 *A. media*;
569 7 *Rondeletiola minor*; 8 *Sepiola tridens*; 9 *S. atlantica*; 10 *S. ligulata*; 11 *Heteroteuthis*
570 *dispar*; 12 Ommastrephidae; 13 *Todarodes sagittatus*; 14 *Gonatus steenstrupi*; 15
571 *Ancistrocheirus alessandrini*; 16 *Pyroteuthis margaritifera*; 17 *Ancistroteuthis lichtensteinii*;
572 *Brachioteuthis riisei*; 19 *Abraliopsis morisii*; 20 *Liocranchia reinhardtii*; 21 *Mastigopsis*
573 *hjorti*.
574

575 The adults of most of the cephalopod paralarvae found during CAIBEX-III are
576 common inhabitants of the tropical and subtropical waters of the Atlantic Ocean (Roper and

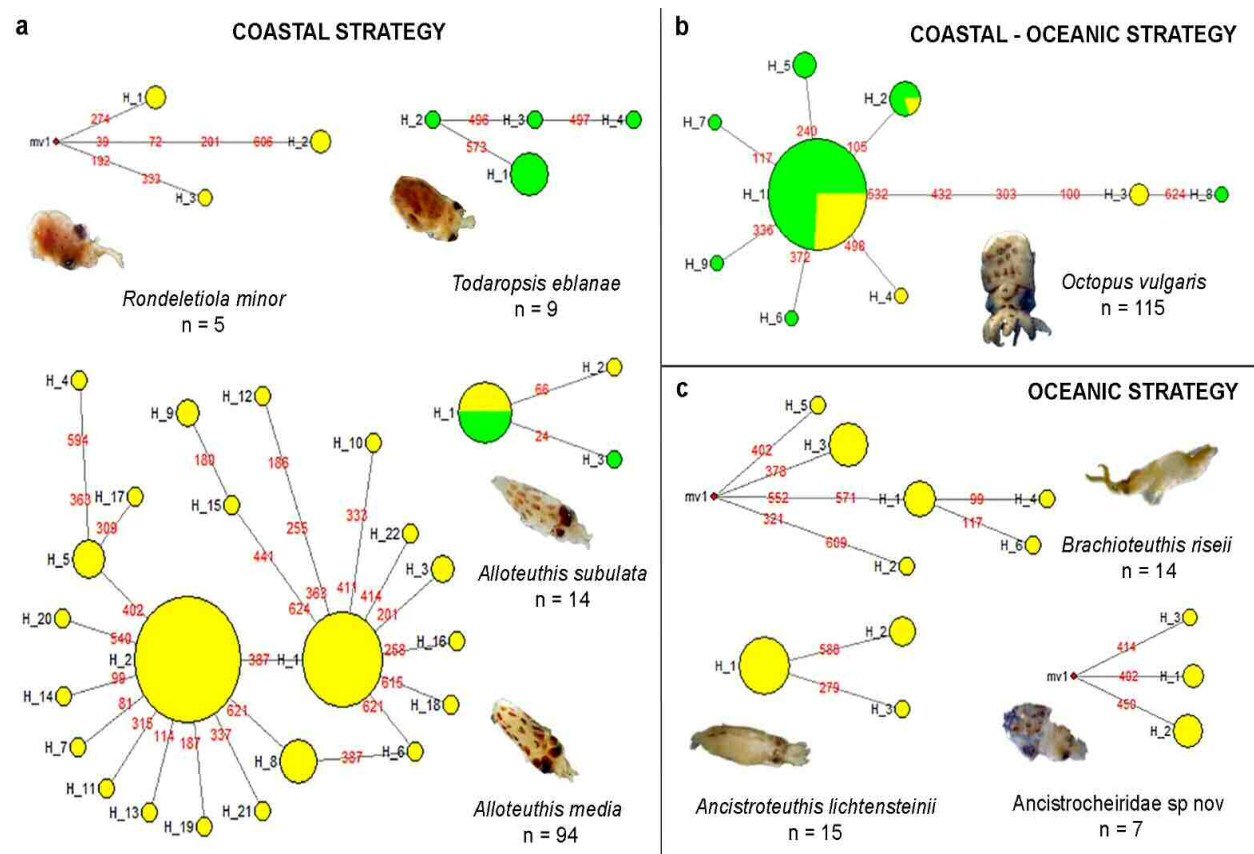
1299
1300
1301 577 Young, 1975; Collins et al., 2001; Diekmann and Piatkowski, 2004; Clarke, 2006; Jereb and
1302 Roper, 2010). Nonetheless, there were records new for the eastern Atlantic, like the juvenile
1303 578 of *Sepiolo ligulata*, collected on the Moroccan shelf (31°0.02'N, 10°0.78'W) at 80 m depth.
1304 579 *Sepiolo ligulata* is a species that, to the best of our knowledge, was only found in the
1305 580 Mediterranean Sea (Jereb and Roper, 2010). Moreover, the specimen of *S. atlantica* found in
1306 581 the coastal sample off Cape Ghir extends the southern limit of this species to 31°N. To our
1307 582 knowledge, the early life stages of *Ancistroteuthis lichtensteinii* (n = 18, 1.7 - 10.1 mm DML)
1308 583 collected during CAIBEX-III are the smallest ever found in the Atlantic (Vecchione et al.,
1309 584 2010) and their vertical distribution demonstrate that this species displays vertical migration
1310 585 at least during their early life stages (Table 2).
1311 586

1312 587 *Ancistrocheirus lesueurii* is presently deemed to be panoceanic and the only
1313 588 representative of the family Ancistrocheiridae (e.g. Roper and Jereb, 2010), whereas in the
1314 589 past two different species were ascribed to that family, namely *A. lesueurii* and
1315 590 *Thelidioteuthis alessandrinii* (e.g. Clarke, 1966). In the 1980s *A. lesueurii* was believed to be
1316 591 a cosmopolitan animal and *T. alessandrinii* was suspected to be the juvenile form of the same
1317 592 species (Roper et al., 1984). Since some ambiguities existed about the correct name to
1318 593 indicate the supposed unique ancistrocheirid species, Bello (1992) showed that it was
1319 594 *Ancistrocheirus lesueurii* (d'Orbigny [in Férussac & d'Orbigny], 1842), with type locality the
1320 595 Indo-Pacific Ocean. Based on the present genetic data, we suggest that the paralarvae
1321 596 collected in Morocco may represent a new species congeneric to, but different from, *A.*
1322 597 *lesueurii*, within the hitherto monotypic Ancistrocheiridae family. This suggestion is in
1323 598 agreement with differences in paralarval morphology between Atlantic and Pacific specimens
1324 599 that suggest that more than one species likely exist (Young et al., 1998), and also with the
1325 600 phylogenetic tree shown in Fig. A1 that shows a third species collected in South Africa
1326 601 within the Ancistrocheiridae.

1327 602 According to the geographical origin of the ancistrocheirid paralarvae collected in this
1328 603 study, i.e. close to the entrance to the Mediterranean, we can safely suppose that these
1329 604 paralarvae belong to the same ancistrocheirid species that occurs in the Mediterranean (all
1330 605 Mediterranean oegopsid squids are also distributed in the eastern Atlantic; cf. Jereb & Roper,
1331 606 2010). *A. lesueurii* was described by Vérany (1847), who named it *Loligo alessandrinii*,
1332 607 based on a paralarval specimen collected in the harbour of Messina (type locality: Strait of
1333 608 Messina, western Mediterranean). More than a century later, its presence was confirmed by
1334 609 the finding of the first adult female in the type locality, which was identified as *A. lesueurii*
1335 610 (Bello et al., 1994). Our genetic results suggest that the eastern Atlantic-Mediterranean
1336
1337
1338
1339
1340
1341
1342
1343
1344
1345
1346
1347
1348
1349
1350
1351
1352
1353
1354
1355
1356
1357

1358
 1359
 1360
 1361
 1362
 1363
 1364
 1365
 1366
 1367
 1368
 1369
 1370
 1371
 1372
 1373
 1374
 1375
 1376
 1377
 1378
 1379
 1380
 1381
 1382
 1383
 1384
 1385
 1386
 1387
 1388
 1389
 1390
 1391
 1392
 1393
 1394
 1395
 1396
 1397
 1398
 1399
 1400
 1401
 1402
 1403
 1404
 1405
 1406
 1407
 1408
 1409
 1410
 1411
 1412
 1413
 1414
 1415
 1416

611 species is different from that known from the Indo-Pacific (namely *A. lesueurii*, Fig. A.1),
 612 therefore we would have to assign the ancistrocheirid paralarvae found off the coast of
 613 Morocco to the former species that is *Ancistrocheirus alessandrinii* (Vérany, 1847).
 614 However, a morphological and genetic study with adults needs to be undertaken to uncover
 615 the true diversity within the Ancistrocheiridae family.



616 **Fig. 9.** Haplotype networks obtained for those cephalopod species with more than 5
 617 individuals and more than 3 different haplotypes, grouped according to their dispersal pattern.
 618 Colours represent the upwelling regions sampled: green, cape Silleiro (NW Iberian
 619 Peninsula); yellow, Cape Ghir (W Morocco).
 620

621
 622 According to the geographical origin of our paralarvae, i.e. close to the entrance to the
 623 Mediterranean, we can safely suppose that these paralarvae belong to the same
 624 ancistrocheirid species that occurs in the Mediterranean (all Mediterranean oegopsid squids
 625 are also distributed in the eastern Atlantic; cf. Jereb & Roper, 2010). *A. lesueurii* was
 626 described by Vérany (1847), who named it *Loligo alessandrinii*, based on a paralarval
 627 specimen collected in the harbour of Messina (type locality: Straits of Messina, western
 628 Mediterranean). More than a century later, its presence was confirmed by the finding of the

1417
1418
1419 629 first adult female in the type locality, which was identified as *A. lesueurii* (Bello et al., 1994).
1420
1421 630 Our genetic results suggest that the eastern Atlantic-Mediterranean species is different from
1422
1423 631 that known from the Indo-Pacific (namely *A. lesueurii*, Fig. A.1), therefore we would have to
1424
1425 632 assign the ancistrocheirid paralarvae found off the coast of Morocco to the former species
1426
1427 633 that is *Ancistrocheirus alessandrini* (Vérany, 1847).
1428

1429 635 4.2 Planktonic dispersal patterns in coastal upwelling systems: interplay between 1430 1431 636 oceanography and vertical behaviour

1432 637
1433
1434 638 Changes in the vertical behaviour of the cephalopod paralarvae under the same
1435
1436 639 oceanographic conditions in the upwelling regions of NE Atlantic lead to different
1437
1438 640 dispersal/retention capabilities that likely affect the patterns of genetic diversity detected.
1439
1440 641 Three major life dispersal patterns were identified for the planktonic stage of cephalopods:
1441
1442 642 coastal, coastal-oceanic and oceanic (Fig. 8). The coastal dispersal pattern is followed by
1443
1444 643 members of the families Loliginidae, Sepiolidae and Ommastrephidae, distributed from the
1445
1446 644 coast to the edge of the continental shelf (200m). This distribution was only detected in
1447
1448 645 *Octopus vulgaris*, with hatchings found in the coastal-shelf area and older paralarvae in the
1449
1450 646 open ocean. The oceanic dispersal pattern was observed for certain species of coastal families
1451
1452 647 like Sepiolidae (*Heteroteuthis dispar*) and Ommastrephidae (*Todarodes sagittatus*) together
1453
1454 648 with oceanic midwater families including Onychoteuthidae, Brachioteuthidae, Pyroteuthidae,
1455
1456 649 Ancistrocheiridae, Enoploteuthidae, Gonatidae, Mastigoteuthidae and Cranchiidae.

1457
1458 650 Cephalopod paralarvae with a coastal dispersal pattern are characterised by retention
1459
1460 651 of different developmental stages, which is likely achieved by adjusting their vertical
1461
1462 652 behaviour in accordance with the oceanographic conditions (Roura et al., 2016). Such
1463
1464 653 retention was supported by the schooling behaviour of the loliginid paralarvae (*Alloteuthis*
1465
1466 654 *media* and *A. subulata*, n = 99 that included specimens with TL ranging from 1.85 - 13.2 mm,
1467
1468 655 Table A.3) found in the water-column sample off the coast of Morocco (Table 2). Other
1469
1470 656 loliginid paralarvae (*A. media*, n = 16) were also found in the same coastal sample at the
1471
1472 657 surface and, therefore, would be expected to occur within the filament as it flowed seaward.
1473
1474 658 However, the lack of any loliginid paralarvae beyond the continental shelf in both upwelling
1475
1476 659 ecosystems (observed in this study and previous ones, e.g. Rocha et al., 1999; Moreno et al.,
1477
1478 660 2009) can be explained through an active behaviour controlling their vertical position - in this
1479
1480 661 case moving downward to evade the offshore surface flow - effectively limiting offshore
1481
1482 662 dispersal while favouring alongshore retention, as suggested by other studies (Otero et al.

1476
1477
1478
1479
1480
1481
1482
1483
1484
1485
1486
1487
1488
1489
1490
1491
1492
1493
1494
1495
1496
1497
1498
1499
1500
1501
1502
1503
1504
1505
1506
1507
1508
1509
1510
1511
1512
1513
1514
1515
1516
1517
1518
1519
1520
1521
1522
1523
1524
1525
1526
1527
1528
1529
1530
1531
1532
1533
1534

663 2009; Roura et al. 2016). Such behaviour has also been observed in crustacean larvae in the
664 ICC (Queiroga and Blanton, 2004; Queiroga et al., 2007) as well as in other upwelling
665 ecosystems (Roberts, 2005; Shanks and Shearman, 2009; Morgan and Fisher, 2010).
666 Consequently, the spatio-temporal aggregation of different life stages from different origins
667 likely contributes to high nucleotide (π) and haplotype (h) diversities for cephalopod
668 paralarvae displaying coastal dispersal patterns (Fig. 8). A similar effect has been observed in
669 the cardinal reef fish (*Ostorhinchus doederleini*) which has strong homing behaviour – high
670 aggregation and low dispersal – and displayed the highest genetic structure recorded to date
671 for relatively small scales (<20 km) as pointed out by Gerlach et al. (2007). The exception to
672 this general trend is represented by *A. subulata*, which inhabits deeper waters than its
673 congener *A. media*. The low π and h diversity might reflect the small number of sequences
674 obtained for this species, but a similar haplotype network with only three haplotypes was
675 obtained from 37 specimens collected in the Ría de Vigo between 2012 and 2014 (Olmos-
676 Pérez, 2018).

677 Contrary to the coastal dispersal pattern, the coastal-oceanic migration of *Octopus*
678 *vulgaris* requires a tight coupling of their vertical behaviour with the upwelled surface waters
679 to be transported offshore far from the coastal area, as suggested by Roura et al. (2016). One
680 of the mechanisms proposed for such migration would be offshore transport in the oceanward
681 upwelling filaments, as was observed for neritic ichthyoplankton larvae south of the Canary
682 Islands (Rodríguez et al., 1999). Despite the limitations of interpreting vertical data collected
683 with bongo nets, our results support the proposed migration mechanism, because octopus
684 paralarvae were mainly present at the surface at night and absent from the surface layer
685 during the day (Tables 1-2). Detailed vertical studies carried out in the Ría de Vigo (NW
686 Iberian Peninsula) with a Multinet between 2012 and 2014 revealed that *O. vulgaris*
687 paralarvae are mostly found during the day between 5 - 20 m and between 0 to 5 m at night
688 (Olmos-Pérez, 2018). Such positioning would allow octopus paralarvae to be transported
689 seaward within the upwelling filaments, which in our study was 60 m deep and 25 km wide
690 (Sangrá et al. 2015). In fact, *O. vulgaris* was the only cephalopod paralarvae that was found
691 in all the locations sampled in this study – *i. e.* the coastal area, the upwelling zone, the
692 filament and open ocean (Tables 1-2) – as a result of this offshore transport and dispersal.
693 Supporting this migratory behaviour is the positive relationship found between the distance to
694 coast and the size of the paralarvae (Fig. 6), showing that the paralarvae are feeding correctly
695 during this migration and are growing towards the open ocean. Species with large hatchlings

1535
1536
1537 696 such as *Enteroctopus dofleini* have also been found distributed in both shallow and oceanic
1538
1539 697 waters in the north Pacific Ocean (Villanueva and Norman, 2008).

1540 698 Moreover, age estimations based on beak ring increments showed that octopus
1541
1542 699 paralarvae collected from the open ocean were older (up to 28 days, Perales-Raya et al.,
1543
1544 700 2017) than those collected close to the coast (less than 8 days, Garrido et al., 2016; Perales-
1545
1546 701 Raya et al., 2017). Additional evidence of the seaward migration is that the 3,739 *O. vulgaris*
1547 702 paralarvae found in the ICC to date have been collected close to the coast over the continental
1548
1549 703 shelf with only three suckers per arm (Rocha et al., 1999; González et al., 2005; Otero et al.,
1550
1551 704 2008; Moreno et al., 2009; Garrido et al., 2016; Roura et al., 2013; Lourenço et al., 2017).
1552 705 However, up to 74 *O. vulgaris* individuals were collected beyond the edge of the continental
1553
1554 706 shelf during CAIBEX-I and -III (bottom depths ranging from 787 to 3110 m) and 58 of them
1555 707 had more than three suckers per arm. These results support the hypothesis that *O. vulgaris*
1556
1557 708 hatchlings are advected oceanward by coupling their vertical distribution with the prevailing
1558
1559 709 oceanographic conditions to leave the coastal area, because paralarvae with more than 3
1560 710 suckers have only been found far from the continental shelf.

1561 711 The genetic data provided further evidence of the mechanism underpinning such
1562
1563 712 dispersal in *O. vulgaris* paralarvae. Despite being found in all the locations sampled through a
1564
1565 713 coastal-oceanic distance of more than 171 km and an alongshore extent covering more than
1566
1567 714 2,000 km (from 31° to 42°N), the spatial genetic diversity obtained for *O. vulgaris*, compared
1568
1569 715 with the rest of cephalopod paralarvae (Fig. 8), can only be explained by means of within-
1570
1571 716 cohort sampling. This pattern of relatedness / low genetic diversity may result from
1572
1573 717 paralarvae with similar origins remaining together throughout their dispersal stage, in this
1574
1575 718 case in the upwelling waters and filament. Planes et al. (2002) revealed that as recruit cohorts
1576
1577 719 of the blue spine unicorn fish (*Naso unicornis*) aged, their internal mean relatedness
1578
1579 720 decreased. However, the large spatial scale that separates the two upwelling ecosystems
1580
1581 721 (~2,000 km) and the different haplotypes detected clearly indicates that different hatching
1582
1583 722 events were sampled. Low levels of population differentiation were also observed in the
1584
1585 723 velvet swimming crab around the Iberian Peninsula (Sotelo et al. 2009). This species also has
1586
1587 724 a pelagic stage of around two months suggesting that gene flow could be also operating in
1588
1589 725 this area to maintain the genetic homogeneity observed.

1585 726 In order to investigate whether the genetic patterns detected in CAIBEX-I and III
1586
1587 727 resulted from schooling behaviour of larval *O. vulgaris*, the genetic diversity of the
1588
1589 728 paralarvae collected in this study was compared to that of the species across its distribution
1590
1591 729 range. COI samples were selected from GenBank based on the distribution of *O. vulgaris*
1592
1593

1594
1595
1596 730 *sensu stricto* (Amor et al. 2014; 2017): three from Galicia (NW Spain: DQ683221-683223),
1597
1598 731 two from Portugal (KF844042-844043), three from Senegal (DQ683224-683226), two from
1599
1600 732 west Mediterranean (France: DQ683227, KF774311) and two from east Mediterranean
1601 733 (Turkey: KC311412, KC789315). The obtained sequences of *O. vulgaris sensu stricto* had a
1602
1603 734 common region of 482 bp and the same fragment was trimmed in our sequences for direct
1604
1605 735 comparison. Genetic diversities for *O. vulgaris sensu stricto* were $h = 0.530 \pm 0.076$ and $\pi =$
1606 736 0.0033 ± 0.00048 , while for our paralarvae were $h = 0.166 \pm 0.047$ and $\pi = 0.00057 \pm$
1607
1608 737 0.00019 . Such marked difference, suggest that the genetic pattern observed in the paralarvae
1609
1610 738 was the result of sampling closely related individuals that were transported within the same
1611 739 water mass. This is a clear example of how a multidisciplinary approach that combines
1612
1613 740 oceanography, behaviour, morphometry and genetics can shed light to explain low diversity
1614 741 values that would be otherwise inexplicable (Selkoe et al. 2008).

1615
1616 742 The stability and age of the oceanic pelagic ecosystem has enabled the evolution of
1617 743 many species, with subtle niche differences (Hopkins and Sutton, 1998). The oceanic
1618
1619 744 dispersal pattern was followed by oceanic squids and oceanic species of coastal families like
1620
1621 745 the Sepiolidae. These oceanic paralarvae were present within samples with bottom depths
1622 746 above 2000 m and were absent from the samples collected in the coastal and upwelling areas.
1623
1624 747 All the oceanic species found in this study complete their development in the oceanic realm
1625 748 and have been observed elsewhere in the Atlantic (Diekmann and Piatkowski, 2004,
1626
1627 749 Vecchione et al. 2010). Genetic diversity in these paralarvae was also high, as a result of
1628
1629 750 sampling different specimens from different developmental stages and different origins.
1630 751 Mesopelagic cephalopods, together with fishes and decapods, are key zooplankton predators
1631
1632 752 in the ocean that display diel vertical migration to feed close to the surface at night
1633 753 (Passarella and Hopkins, 1991). Interestingly, onychoteuthid and ancistrocheirid paralarvae
1634
1635 754 were concentrated in the surface waters of the samples collected within the filament at night
1636 755 (Table 2), likely attracted by the increased zooplankton biomass (Hernández-León et al.,
1637
1638 756 2002), as observed in other mesopelagic predators like shrimps or fishes (Hopkins and
1639
1640 757 Sutton, 1998).

1641 758 New comprehensive studies are needed to investigate the coupling of larval
1642
1643 759 abundance in the plankton with settlement and post-settlement mortality on the shore for
1644
1645 760 diverse taxa and locations across upwelling coasts to determine the underlying mechanisms
1646 761 responsible for the observed spatial and temporal patterns of recruitment. Such investigations
1647
1648
1649
1650
1651
1652

1653
1654
1655 762 are essential for further advancing our understanding of processes that regulate marine
1656 763 populations and communities.

1659 764 **5. Conclusions**

1662 765 - The multidisciplinary approach undertaken in this study sheds light on the processes that
1663 766 shape genetic diversity/population structure of planktonic cephalopod paralarvae in two
1664 767 upwelling ecosystems of the Canary Current eastern boundary upwelling ecosystem.

1667 768 - Eight cephalopod species were genetically identified in the upwelling ecosystem of NW
1668 769 Iberian Peninsula and 16 in W Morocco, showing a marked latitudinal change in the
1670 770 cephalopod assemblage.

1672 771 - An undescribed species within the monotypic Ancistrocheiridae family was genetically
1673 772 identified in Moroccan waters and named *Ancistrocheirus alessandrinii* (Vérany, 1847).

1675 773 - Genetic diversity in the COI gene revealed no genetic structure between the two upwelling
1676 774 regions for *Alloteuthis subulata* and *Octopus vulgaris*.

1678 775 - Changes in the vertical behaviour of the cephalopod paralarvae under the same
1679 776 oceanographic conditions lead to different dispersal/retention capabilities and three different
1680 777 planktonic dispersal patterns were identified: coastal, coastal-oceanic and oceanic.

1683 778 - Coastal and oceanic dispersal patterns displayed high levels of nucleotide and haplotype
1684 779 diversity as a result of spatio-temporal retention of different life stages, while the low levels
1685 780 registered for the coastal-oceanic dispersal pattern resulted from the advection of closely
1686 781 related specimens within upwelled waters / filaments into the ocean.

1689 782 - *Octopus vulgaris* is the only cephalopod that display a coastal-oceanic dispersal pattern in
1690 783 the ICC as shown by the presence of paralarvae with more than three suckers per arm far
1691 784 from the continental shelf, the significant positive correlation between size and distance to
1692 785 coast and the low genetic diversity measured.

1697 786 **6. Acknowledgements**

1699 787 We are indebted to the captain, crew and technicians of R/V “Sarmiento de Gamboa”,
1700 788 for their assistance in collecting the zooplankton samples and hydrographical data. We
1701 789 acknowledge the help of Félix Álvarez sorting the paralarvae, as well as Mariana Rivas for
1702 790 preparing the cephalopod samples for barcoding. Special thanks to Dr. Dirk Steinke, who
1703 791 offered the facilities of the Barcoding of Life Data (BOLD) to sequence the cephalopods.
1704 792 Furthermore, we thank Marcos Regueira and Rocío Graña for their help preparing figures 1,
1705
1706
1707
1708
1709
1710
1711

1712
1713
1714
1715
1716
1717
1718
1719
1720
1721
1722
1723
1724
1725
1726
1727
1728
1729
1730
1731
1732
1733
1734
1735
1736
1737
1738
1739
1740
1741
1742
1743
1744
1745
1746
1747
1748
1749
1750
1751
1752
1753
1754
1755
1756
1757
1758
1759
1760
1761
1762
1763
1764
1765
1766
1767
1768
1769
1770

793 and 2 and 3, respectively. This study was supported by the project CAIBEX (Spanish
794 Ministry of Innovation and Science CTM2007-66408-C02) and the first author by a
795 “Fundación Barrié de la Maza” postdoctoral fellowship (3003197/2013) and a Securing Food,
796 Water and the Environment Research Focus Area grant (La Trobe University) during writing.

797 **7. References**

- 798 1. Akaike, H. 1974. A new look at the statistical model identification. IEEE Trans Automat
799 Contr 19: 716-723. doi: 10.1109/TAC.1974.1100705.
- 800 2. Álvarez-Salgado, X.A., Aristegui, J., Barton, E.D., Hansell, D.A., 2007. Contribution of
801 upwelling filaments to offshore carbon export in the subtropical Northeast Atlantic Ocean.
802 Limnol Oceanogr. 52: 1287-1292. Doi: 10.4319/lo.2007.52.3.1287.
- 803 3. Amor, M.D., Norman, M.D., Cameron, H.E., Strugnell, J.M. 2014. Allopatric speciation
804 within a cryptic species complex of Australasian octopuses. PLoS ONE, 9, e98982. doi:
805 10.1371/journal.pone.0098982.
- 806 4. Amor, M.D., Norman, M.D., Roura, A., Leite, T.S., Gleadall, I.G., Reid, A., et al. 2017.
807 Morphological assessment of the *Octopus vulgaris* species complex evaluated in light of
808 molecular-based phylogenetic inferences. Zool Scripta, 46: 275-288. doi:
809 10.1111/zsc.12207.
- 810 5. Anderson, M.J., Gorley, R.N., Clarke, K.R., 2008. PERMANOVA+ for PRIMER: Guide
811 to Software and Statistical Methods. PRIMER-E, Plymouth, UK.
- 812 6. Anderson, FE, Pilsits, A, Clutts, S, Laptikhovsky, V, Bello, G, Balguerías, E, Lipinski, M,
813 et al. 2008. Systematics of *Alloteuthis* (Cephalopoda: Loliginidae) based on molecular and
814 morphometric data. J Exp Mar Biol Ecol. 364: 99-109. doi: 10.1016/j.jembe.2008.07.026
- 815 7. Aristegui, J., Barton, E.D., Álvarez-Salgado, X.A., Santos, A.M.P., Figueiras, F.G.,
816 Kifani, S., Hernández-León, S., Mason, E., Machú, E., Demarcq, H. 2009. Sub-regional
817 ecosystem variability in the Canary Current upwelling. Prog Oceanogr. 83: 33-48. Doi:
818 10.1016/j.pocean.2009.07.031
- 819 8. Bandelt H.J., Forster P., Röhl A. 1999. Median-joining networks for inferring intraspecific
820 phylogenies. Mol Biol Evol. 16: 37-48. Doi: 10.1093/oxfordjournals.molbev.a026036
- 821 9. Barton, E.D. 1998. Eastern boundary of the North Atlantic: Northwest Africa and Iberia.
822 Coastal segment (18,E). In *The Sea*, Vol. 11, Robinson, A. R. and K. H. Brink, eds, John
823 Wiley and Sons, Inc., New York, pp. 633-657.

1771
1772
1773
1774
1775
1776
1777
1778
1779
1780
1781
1782
1783
1784
1785
1786
1787
1788
1789
1790
1791
1792
1793
1794
1795
1796
1797
1798
1799
1800
1801
1802
1803
1804
1805
1806
1807
1808
1809
1810
1811
1812
1813
1814
1815
1816
1817
1818
1819
1820
1821
1822
1823
1824
1825
1826
1827
1828
1829

- 824 10. Barton, E.D., Inall, M., Sherwin, T.J., Torres, R. 2001. Vertical structure, turbulent mixing
825 and fluxes during Lagrangian observations of an upwelling filament system off Northwest
826 Iberia. *Prog Oceanogr.* 51: 249-267. Doi: 10.1016/S0079-6611(01)00069-6
- 827 11. Bécognée, P., Almeida, C., Barrera, A., Hernández-Guerra, A., Hernández-León, S. 2006.
828 Annual cycle of clupeiform larvae around Gran Canaria Island, Canary Islands. *Fish*
829 *Oceanogr.* 15: 293-300. Doi: 10.1111/j.1365-2419.2005.00390.x
- 830 12. Bello, G. 1992. On the validity, authorship, and publication date of the specific name
831 *Ancistrocheirus lesueurii* (Cephalopoda: Ancistrocheiridae). *Veliger*, 35:141-145.
- 832 13. Bello, G., Potoschi, A. Berdar, A. 1994. Adult of *Ancistrocheirus lesueurii* caught in the
833 straits of Messina (Cephalopoda: Ancistrocheiridae). *Boll Malacol.* 29: 259-266. ISSN:
834 0394-7149.
- 835 14. Bohonak, A.J. 1999. Dispersal, gene flow, and population structure. *Q Rev Biol* 74: 21–
836 45. Doi: 10.1086/392950
- 837 15. Boletzky, S.v. 2003. Biology of early life stages in cephalopod molluscs. *Adv Mar Biol.*
838 44: 143-203. Doi: 10.1016/S0065-2881(03)44003-0
- 839 16. Boyle, P.R., Boletsky, S.V. 1996. Populations: definitions and dynamics. In *The role of*
840 *cephalopods in the world's oceans.* Edited by M.R. Clarke. *Phil Trans R Soc Lond B.* 351:
841 985-1002. Doi: 10.1098/rstb.1996.0089
- 842 17. Clarke, K.R., Green, R.H. 1988. Statistical design and analysis for a "biological effects"
843 study. *Mar Ecol Prog Ser.* 46: 213-226. Doi: 10.3354/meps046213
- 844 18. Clarke, M.R. 1966. A review of the systematics and ecology of oceanic squids. *Adv Mar*
845 *Biol.* 4: 91-300. Doi: 10.1016/S0065-2881(08)60314-4
- 846 19. Clarke, M.R. 2006. Oceanic cephalopod distribution and species diversity in the eastern
847 north Atlantic. *Arquipélago. Life and Marine Sciences.* 23A: 27-46.
- 848 20. Collins, M.A., Yau, C., Allcock, A.L., Thurston, M.H. 2001. Distribution of deep-water
849 benthic and benthopelagic cephalopods from the north-east Atlantic. *J Mar Biol Assoc*
850 *U.K.* 81: 105-117. Doi: 10.1017/S0025315401003459
- 851 21. Cordeiro, N., Nolasco, R., Cordeiro-Pires, A., Barton, E.D., Dubert, J. 2015. Filaments on
852 the Western Iberian Margin: a modeling study. *J Geophys Res Oc.* 120,
853 doi:10.1002/2014JC010688.
- 854 22. Cordeiro, N., Dubert, J., Nolasco, R., Barton, E.D. 2018. Transient response of the
855 Northwestern Iberian upwelling regime. *PLoS ONE* 13(5): e0197627. [https://doi.org/](https://doi.org/10.1371/journal.pone.0197627)
856 [10.1371/journal.pone.0197627](https://doi.org/10.1371/journal.pone.0197627)

1830
1831
1832
1833
1834
1835
1836
1837
1838
1839
1840
1841
1842
1843
1844
1845
1846
1847
1848
1849
1850
1851
1852
1853
1854
1855
1856
1857
1858
1859
1860
1861
1862
1863
1864
1865
1866
1867
1868
1869
1870
1871
1872
1873
1874
1875
1876
1877
1878
1879
1880
1881
1882
1883
1884
1885
1886
1887
1888

- 857 23. Diekmann, R., Piatkowski, U. 2004. Species composition and distribution patterns of early
858 life stages of cephalopods at Great Meteor Seamount (subtropical North-east Atlantic).
859 Arch Fish Mar Res. 51: 115-131. Doi: 0944-1921/2004/51/1-3-115
- 860 24. Excoffier, L., Laval, G., Schneider, S. 2005. ARLEQUIN v.3.0: an integrated software
861 package for population genetics data analysis. Evol Bioinform Online. 1: 47-50.
- 862 25. Faure, V., Inejih, A.C., Demarcq, H., Cury, P. 2000. The importance of retention
863 processes in upwelling areas for recruitment of *Octopus vulgaris*: the example of the
864 Arguin Bank (Mauritania). Fish Oceanogr. 9: 343-355. Doi: 10.1046/j.1365-
865 2419.2000.00149.x
- 866 26. Fernández-Álvarez, F.A., Martins, C.P.P., Vidal, E.A.G., Villanueva, R. 2016. Towards
867 the identification of the ommastrephid squid paralarvae (Mollusca: Cephalopoda):
868 Morphological description of three species and a key to the north-east Atlantic species.
869 Zool J Linn Soc. doi: 10.1111/zoj.12496
- 870 27. Garrido, D., Navarro, J.C., Perales-Raya, C., Nande, M., Martín, M.V., Iglesias, J., et al.
871 2016. Fatty acid composition and age estimation of wild *Octopus vulgaris* paralarvae.
872 Aquaculture. 464: 564-569. Doi: 10.1016/j.aquaculture.2016.07.034
- 873 28. Gerlach, G., Atema, J., Kingsford, M.J., Black, K.P., Miller-Sims, V. 2007. Smelling
874 home can prevent dispersal of reef fish larvae. Proc Natl Acad Sci U S A. 104: 858-863.
875 Doi: 10.1073/pnas.0606777104
- 876 29. Goodall-Copestake, W., Tarling, G.A., Murphy, E.J. 2012. On the comparison of
877 population-level estimates of haplotype and nucleotide diversity: a case study using the
878 gene *cox1* in animals. Heredity, 109: 50-56. Doi: 10.1038/hdy.2012.12
- 879 30. González, A.F., Otero, J., Guerra, A., Prego, R., Rocha, F., Dale, A.W. 2005. Distribution
880 of common octopus and common squid paralarvae in a wind-driven upwelling area (Ría of
881 Vigo, northwestern Spain). J Plankton Res. 27: 271-277. Doi: 10.1093/plankt/fbi001
- 882 31. González, A.F., Otero, J., Pierce, G.J., Guerra, A. 2010. Age, growth, and mortality of
883 *Loligo vulgaris* wild paralarvae: implications for understanding of the life cycle and
884 longevity. ICES J Mar Sci. 67: 1119-1127. Doi: 10.1093/icesjms/fsq014
- 885 32. Guindon, S., Dufayard, J.F., Lefort, V., Anisimova, M., Hordijk, W., et al. 2010. New
886 algorithms and methods to estimate Maximum-Likelihood phylogenies: Assessing the
887 performance of PhyML 3.0. Syst Biol. 59: 307-321. Doi: 10.1093/sysbio/syq010
- 888 33. Hebert, P.D.N., Cywinska, A., Ball, S.L., de Waard, J.R. 2003. Biological identifications
889 through DNA barcodes. Proc R Soc Lond B Biol Sci, 270: 313-321. Doi:
890 10.1098/rspb.2002.2218

1889
1890
1891
1892
1893
1894
1895
1896
1897
1898
1899
1900
1901
1902
1903
1904
1905
1906
1907
1908
1909
1910
1911
1912
1913
1914
1915
1916
1917
1918
1919
1920
1921
1922
1923
1924
1925
1926
1927
1928
1929
1930
1931
1932
1933
1934
1935
1936
1937
1938
1939
1940
1941
1942
1943
1944
1945
1946
1947

- 891 34. de Heij, A., Goud, J. 2010. *Sepiola tridens* spec. nov., an overlooked species
892 (Cephalopoda, Sepiolidae) living in the North Sea and north-eastern Atlantic Ocean.
893 *Basteria*. 74: 51-62.
- 894 35. Hernández-León, S., Almeida, C., Portillo-Hahnefeld, A., Gómez, M., Rodríguez, J.M.,
895 Aristegui, J., 2002. Zooplankton biomass and indices of feeding and metabolism in
896 relation to a filament off the Northwest African Upwelling zone. *J Mar Res.* 60: 327-346.
897 10.1357/00222400260497516
- 898 36. Hopkins, T.L., Sutton, T.T. 1998. Midwater fishes and shrimps as competitors and
899 resource partitioning in low latitude oligotrophic ecosystems. *Mar Ecol Prog Ser.* 164: 37-
900 45. Doi: 10.3354/meps164037
- 901 37. Jereb, P., Roper, C.F.E. 2010. Cephalopods of the world. An annotated and illustrated
902 catalogue of cephalopod species known to date. Volume 2. Myopsid and Oegopsid Squids.
903 FAO Species Catalogue for Fishery Purposes. No. 4, Vol. 2. Rome, FAO. 605p. Doi: 978-
904 92-5-106720-8
- 905 38. Joint, I., Inall, M., Torres, R., Figueiras, F.G., Álvarez-Salgado, X.A., Rees, A.P.,
906 Woodward, E.M.S. 2001. Two lagrangian experiments in the Iberian upwelling system:
907 tracking an upwelling event and an off-shore filament. *Prog Oceanogr.* 51: 221-248. Doi:
908 10.1016/S0079-6611(01)00068-4
- 909 39. Librado, P., Rozas, J. 2009. DnaSP v5: A software for comprehensive analysis of DNA
910 polymorphism data. *Bioinformatics.* 25: 1451-1452. Doi: 10.1093/bioinformatics/btp187
- 911 40. Lourenço, S., Roura, Á., Fernández-Reiriz, M.-J., Narciso, L., González, Á.F. 2017.
912 Feeding relationship between *Octopus vulgaris* (Cuvier, 1797) early life-cycle stages and
913 their prey in the Western Iberian upwelling system: correlation of reciprocal lipid and fatty
914 acid contents. *Front Physiol*, 8: 467. Doi: 10.3389/fphys.2017.00467
- 915 41. Moreno, A., dos Santos, A., Piatkowski, U., Santos, A.M.P., Cabral, H. 2009. Distribution
916 of cephalopod paralarvae in relation to the regional oceanography of the western Iberia. *J*
917 *Plankton Res.* 31: 73-91. Doi: 10.1093/plankt/fbn103
- 918 42. Morgan, S.G., Fisher, J.L. 2010. Larval behavior regulates nearshore retention and
919 offshore migration in an upwelling shadow and along the open coast. *Mar Ecol Prog Ser.*
920 404: 109–126. Doi: 10.3354/meps08476
- 921 43. Navarro-Pérez, E., Barton, E.D. 1998. The physical structure of an upwelling filament off
922 the north-west African coast during August 1993. In: Benguela dynamics: impacts of
923 variability of shelf – sea environments and their living resources, S.C. Pillar, C.L.

1948
1949
1950
1951
1952
1953
1954
1955
1956
1957
1958
1959
1960
1961
1962
1963
1964
1965
1966
1967
1968
1969
1970
1971
1972
1973
1974
1975
1976
1977
1978
1979
1980
1981
1982
1983
1984
1985
1986
1987
1988
1989
1990
1991
1992
1993
1994
1995
1996
1997
1998
1999
2000
2001
2002
2003
2004
2005
2006

- 924 Moloney, A.I.L. Payne and F.A. Shillington (eds.). S Afr J Mar Sci. 19: 61-74. Doi:
925 10.2989/025776198784126827
- 926 44. Olmos-Pérez, L., Roura, Á., Pierce, G.J., González, Á.F. 2017a. Sepiolid paralarval
927 diversity in a regional upwelling area of the NE Atlantic. Hydrobiologia. 808: 57-70. doi:
928 10.1007/s10750-017-3186-3
- 929 45. Olmos-Pérez, L., Roura, Á., Pierce, G.J., Boyer, S., González, Á.F. 2017b. Diet
930 composition and variability of wild *Octopus vulgaris* and *Alloteuthis media* (Cephalopoda)
931 paralarvae through a metagenomic lens. Front Physiol. 8: 321. Doi:
932 10.3389/fphys.2017.00321
- 933 46. Olmos-Pérez, L. Ecology of cephalopod paralarvae in a seasonal upwelling system. PhD.
934 Universidad de Vigo, 2018. <http://hdl.handle.net/10261/161653>
- 935 47. Otero, J., Álvarez-Salgado, X.A., González, A.F., Miranda, A., Groom, S.B., Cabanas,
936 J.M., et al. 2008. Bottom-up control of common octopus *Octopus vulgaris* in the Galician
937 upwelling system, northeast Atlantic Ocean. Mar Ecol Prog Ser. 362: 181-192. Doi:
938 10.3354/meps07437
- 939 48. Passarella, K.C., Hopkins, T.L. 1991. Species composition and food habits of the
940 micronektonic cephalopod assemblage in the eastern Gulf of México. Bull Mar Sci 49:
941 638-659.
- 942 49. Peliz, Á., Dubert, J., Santos, A.M.P., Oliveira, P.B., Le Cann, B. 2005. Winter upper
943 ocean circulation in the Western Iberian Basin -Fronts, Eddies and Poleward Flows: an
944 overview. Deep Sea Res Part I Oceanogr Res Pap. 52: 621-646. Doi:
945 10.1016/j.dsr.2004.11.005
- 946 50. Peliz, A., Marchesiello, P., Dubert, J., Marta-Almeida, M., Roy, C., & Queiroga, H. 2007.
947 A study of crab larvae dispersal on the Western Iberian Shelf: Physical processes. J Mar
948 Syst, 68: 215–236. Doi: 10.1016/j.jmarsys.2006.11.007
- 949 51. Posada, D. 2008. jModelTest: Phylogenetic model averaging. Mol Biol Evol. 25: 1253-
950 1256. Doi: 10.1093/molbev/msn083
- 951 52. Queiroga, H., Blanton, J. 2004. Interactions between behaviour and physical forcing in the
952 control of horizontal transport of decapod crustacean larvae. Adv Mar Biol. 47: 107-214.
953 Doi: 10.1016/S0065-2881(04)47002-3
- 954 53. Queiroga, H., Cruz, T., dos Santos, A., Dubert, J., González-Gordillo, J.I., Paula, J., Peliz,
955 Á., Santos, A.M.P. 2007. Oceanographic and behavioural processes affecting invertebrate
956 larval dispersal and supply in the western Iberia upwelling ecosystem. Prog Oceanogr. 74:
957 174-191. Doi: 10.1016/j.pocean.2007.04.007

2007
2008
2009
2010
2011
2012
2013
2014
2015
2016
2017
2018
2019
2020
2021
2022
2023
2024
2025
2026
2027
2028
2029
2030
2031
2032
2033
2034
2035
2036
2037
2038
2039
2040
2041
2042
2043
2044
2045
2046
2047
2048
2049
2050
2051
2052
2053
2054
2055
2056
2057
2058
2059
2060
2061
2062
2063
2064
2065

- 958 54. Perales-Raya, C., Nande, M., Roura, Á., Bartolomé, A., Gestal, C., Otero, J. J., et al. 2017.
959 Comparative study of age estimation in wild and cultured *Octopus vulgaris* paralarvae:
960 Effect of temperature and diet. Mar Ecol Prog Ser. doi: 10.3354/meps12218.
- 961 55. Planes, S., Lecaillon, G., Lenfant, P. and Meekan, M. 2002. Genetic and demographic
962 variation in new recruits of *Naso unicornis*. J Fish Biol. 61: 1033-1049. 10.1111/j.1095-
963 8649.2002.tb01861.x
- 964 56. Rambaut, A., Drummond, A.J. 2003. Tracer 1.3 Oxford University. Available:
965 <http://tree.bio.ed.ac.uk/software/tracer>.
- 966 57. Ratnasingham, S., Hebert, P.D.N. 2007. BOLD: The Barcode of Life Data System
967 (www.barcodinglife.org). Mol Ecol Notes. 7: 355-364. Doi: 10.1111/j.1471-
968 8286.2007.01678.x
- 969 58. Roberts, M.J. 2005. Chokka squid (*Loligo vulgaris reynaudii*) abundance linked to
970 changes in South Africa's Agulhas Bank ecosystem during spawning and the early life
971 cycle. ICES J Mar Sci. 62: 33-55. Doi: 10.1016/j.icesjms.2004.10.002
- 972 59. Rocha, F., Guerra, A., Prego, R., Piatkowski, U. 1999. Cephalopod paralarvae and
973 upwelling conditions off Galician waters (NW Spain). J Plankton Res. 21: 21-33.
974 <http://hdl.handle.net/10261/26345>
- 975 60. Rodríguez, J.M., Hernández-León, S., Barton, E.D. 1999. Mesoscale distribution of fish
976 larvae in relation to an upwelling filament off northwest Africa. Deep-Sea Res. 46: 1969-
977 1984. Doi: 10.1016/S0967-0637(99)00036-9
- 978 61. Ronquist F, Huelsenbeck JP (2003) MrBayes 3: Bayesian phylogenetic inference under
979 mixed models. Bioinformatics. 19: 1572-1574. Doi: 10.1093/bioinformatics/btg180
- 980 62. Roper, C.F.E., Young, R.E. 1975. Vertical distribution of pelagic cephalopods. Smithsonian
981 Contributions to Zoology. 209, 49 pp.
- 982 63. Roper, C.F.E., Sweeney, M.J. Nauen, C.E. 1984. FAO species catalogue. Vol. 3.
983 Cephalopods of the world. FAO Fish Synop 125: 1-277.
- 984 64. Roper, C.F.E., Jereb, P. 2010. Family Enoploteuthidae. In P. Jereb & C.F.E. Roper, eds.
985 Cephalopods of the world. An annotated and illustrated catalogue of species known to
986 date. Volume 2. Myopsid and Oegopsid Squids. FAO Species Catalogue for Fishery
987 Purposes. No. 4, Vol. 2. Rome, FAO. pp. 183-200.
- 988 65. Roura, Á., González, Á.F., Redd, K., Guerra, Á. 2012. Molecular prey identification in
989 wild *Octopus vulgaris* paralarvae. Mar Biol. 159: 1335-1345. Doi: 10.1007/s00227-012-
990 1914-9

2066
2067
2068
2069
2070
2071
2072
2073
2074
2075
2076
2077
2078
2079
2080
2081
2082
2083
2084
2085
2086
2087
2088
2089
2090
2091
2092
2093
2094
2095
2096
2097
2098
2099
2100
2101
2102
2103
2104
2105
2106
2107
2108
2109
2110
2111
2112
2113
2114
2115
2116
2117
2118
2119
2120
2121
2122
2123
2124

- 991 66. Roura, Á., Álvarez-Salgado, X.A., González, A.F., Gregori, M., Rosón, G., Guerra, A.
992 2013. Short-term meso-scale variability of mesozooplankton communities in a coastal
993 upwelling system (NW Spain). *Prog Oceanogr.* 109: 18-32. Doi:
994 10.1016/j.pocean.2012.09.003
- 995 67. Roura, Á., Álvarez-Salgado, X.A., González, A.F., Gregori, M., Rosón, G., Otero, J.,
996 Guerra, A. 2016. Life dispersal patterns of cephalopod paralarvae in a coastal upwelling
997 system (NW Iberian Peninsula): insights from zooplankton community and spatio-
998 temporal analyses. *Fish Oceanogr.* 25: 241-258. Doi: 10.1111/fog.12151
- 999 68. Roura, Á., Doyle, S.R., Nande, M., Strugnell, J.M. 2017. You are what you eat: a
1000 genomic analysis of the gut microbiome of captive and wild *Octopus vulgaris* paralarvae
1001 and their zooplankton prey. *Front Physiol.* 8: 362. Doi: 10.3389/fphys.2017.00362
- 1002 69. Sangrá, P., Troupin, C., Barreiro-González, B., Barton, E.D., Orbi, A., Arístegui, J. 2015.
1003 The Cape Ghir filament system in August 2009 (NW Africa). *J Geophys Res Oceans.* 120:
1004 4516-4533. Doi: 10.1002/2014JC010514
- 1005 70. Selkoe, K.A., Henzler, C.M., Gaines, S.D. 2008. Seascape genetics and the spatial ecology
1006 of marine populations. *Fish Fish.* 9: 363-377. Doi: 10.1111/j.1467-2979.2008.00300.x
- 1007 71. Shanks, A.L., Eckert, G.L. 2005. Population persistence of California Current fishes and
1008 benthic crustaceans: a marine drift paradox. *Ecol Monograph.* 75: 505-524. Doi:
1009 10.1890/05-0309
- 1010 72. Shanks, A.L., Shearman, R.K. 2009. Paradigm lost? Cross-shelf distributions of intertidal
1011 invertebrate larvae are unaffected by upwelling or downwelling. *Mar Ecol Prog Ser.* 385:
1012 189-204. Doi: 10.3354/meps08043
- 1013 73. Shea, E.K., Vecchione, M. 2010. Ontogenic changes in diel vertical migration patterns
1014 compared with known allometric changes in three mesopelagic squid species suggest an
1015 expanded definition of a paralarva. *ICES J Mar Sci.* 67: 1436-1443. Doi:
1016 10.1093/icesjms/fsq104
- 1017 74. Siegel, D.A., Kinlan, B.P., Gaylord, B., Gaines, S.D. 2003. Lagrangian descriptions of
1018 marine larval dispersion. *Mar Ecol Prog Ser.* 260: 83-96. Doi: 10.3354/meps260083
- 1019 75. Sotelo, G., Posada, D., Morán, P. 2009. Low-mitochondrial diversity and lack of structure
1020 in the velvet swimming crab *Necora puber* along the Galician coast. *Mar Biol.* 156: 1039-
1021 1048. Doi: 10.1007/s00227-009-1148-7
- 1022 76. Sweeney, M.J., Roper, C.F.E., Mangold, K., Clarke, M.R., Boletzky, S.V. 1992. 'Larval'
1023 and juvenile cephalopods: a manual for their identification. *Smithsonian Contributions to*
1024 *Zoology*, Washington DC. Doi: 10.5479/si.00810282.513

2125
2126
2127 1025 77. Teske PR, Sandoval-Castillo J, Sebille Ev, Waters J, Beheregaray LB. 2015. On-shelf
2128 larval retention limits population connectivity in a coastal broadcast spawner. *Mar Ecol*
2129 1026 *Prog Ser.* 532: 1-12. Doi: 10.3354/meps11362
2130
2131 1027
2132 1028 78. Troupin, C., Mason, E., Beckers, J.M., Sangrà, P. 2012. Generation of the Cape Ghir
2133 upwelling filament: A numerical study. *Ocean Model.* 41: 1-15. Doi:
2134 1029 10.1016/j.ocemod.2011.09.001
2135 1030
2136
2137 1031 79. Van Camp, L., Nykjaer, L., Mittelstaedt, E., Schlittenhardt, P. 1991. Upwelling and
2138 boundary circulation off Northwest Africa as depicted by infrared and visible satellite
2139 1032 observations. *Prog Oceanogr.* 26: 357-402. Doi: 10.1016/0079-6611(91)90012-B
2140 1033
2141
2142 1034 80. Vance, R.R. 1973. On reproductive dispersal patterns of marine benthic invertebrates.
2143 1035 *Amer Natural.* 107: 339-352. <http://www.jstor.org/stable/2459535>
2144
2145 1036 81. Vecchione, M., Roper, C.F.E., Sweeney, M.J., Lu, C.C. 2001. Distribution, relative
2146 abundance and developmental morphology of paralarval cephalopods in the Western
2147 1037 North Atlantic Ocean. NOAA Tech Report NMFS. 152, 54 pp.
2148 1038
2149
2150 1039 82. Vecchione, M., Young, R.E., Piatkowski, U. 2010. Cephalopods of the northern Mid-
2151 1040 Atlantic Ridge. *Mar Biol Res.* 6: 25-52. Doi: 10.1080/17451000902810751
2152
2153 1041 83. Vérany, J.B. 1847. [On six new species from the Mediterranean]. *Atti della Ottava*
2154 1042 *Reunion degli Scienziati Italiani*, Genova, 14-28 September 1846, pp. 512-514.
2155
2156 1043 84. Villanueva, R., Norman, M.D. 2008. Biology of the planktonic stages of benthic
2157 octopuses. *Oceanogr Mar Biol Annu Rev.* 46: 105-202. Doi: 10.1201/9781420065756.ch4
2158 1044
2159 1045 85. Villanueva, R., Vidal, E., Fernández-Álvarez, F., Nabhitabhata, J. 2016. Early mode of life
2160 and hatchling size in cephalopod molluscs: Influence on the species distributional ranges.
2161 1046 *PloS One*, 11: 1-26. Doi: 10.1371/journal.pone.0165334
2162 1047
2163
2164 1048 86. Weersing, K., Toonen, R.J. 2009. Population genetics, larval dispersal, and connectivity in
2165 marine systems. *Mar Ecol Prog Ser.* 393: 1-12. Doi: 10.3354/meps08287
2166 1049
2167 1050 87. Young, R.E., Harman, R.F. 1988. "Larva", "paralarva" and "subadult" in cephalopod
2168 terminology. *Malacologia.* 29: 201-207.
2169 1051
2170 1052 88. Young, R.E., Burgess L.A., Roper, C.F.E., Sweeney, M.J., Stephen, S.J. 1998.
2171 Classification of the Enoploteuthidae, Pyroteuthidae and Ancistrocheiridae. *Smithsonian*
2172 1053 *Contributions to Zoology*, 586: 239-255.
2173 1054
2174
2175 1055
2176
2177
2178
2179
2180
2181
2182
2183

2184
2185
2186
2187
2188
2189
2190
2191
2192
2193
2194
2195
2196
2197
2198
2199
2200
2201
2202
2203
2204
2205
2206
2207
2208
2209
2210
2211
2212
2213
2214
2215
2216
2217
2218
2219
2220
2221
2222
2223
2224
2225
2226
2227
2228
2229
2230
2231
2232
2233
2234
2235
2236
2237
2238
2239
2240
2241
2242

1056 **Appendices**

1057

1058 **Table A.1.** Detailed information about the mesozooplankton samplings carried out during

1059 CAIBEX-I. Abbreviation: first (L1) and second (L2) Lagrangian experiments.

1060

Sample	Date	CTD	Latitude	Longitude	Depth (m)	Volume (m ³)	Station	Day/night
S1	10-7-09	CS017	41°24.52'N	9°31.04'W	2667	6159	L1	night
S2	11-7-09	CS021	41°21.16'N	9°33.44'W	2885	5881	L1	day
S3	11-7-09	CS026	41°19.87'N	9°32.78'W	2784	7144	L1	night
S4	12-7-09	CS029	41°19.86'N	9°34.26'W	2294	6481	L1	day
S5	12-7-09	CS035	41°17.18'N	9°34.14'W	2924	6050	L1	night
S6	13-7-09	CS038	41°17.38'N	9°35.96'W	3100	5014	L1	day
S7	13-7-09	CS045	41°15.8'N	9°37.29'W	3105	4840	L1	night
S8	14-7-09	CS047	41°30.01'N	8°54.89'W	62	1019	Shelf	day
S9	14-7-09	CS050	41°30'N	9°10'W	134	2192	Shelf	day
S10	14-7-09	CS055	41°29.94'N	9°35.09'W	1940	5039	Ocean	night
S11	15-7-09	CS067	42°5.99'N	9°5.04'W	141	975	Shelf	night
S12	16-7-09	CS083	41°59.92'N	9°11.94'W	147	1796	Shelf	night
S13	17-7-09	CS089	42°0.15'N	9°0.35'W	108	1997	L2	day
S14	17-7-09	CS094	41°55.17'N	9°1.58'W	108	1335	L2	night
S15	18-7-09	CS096	41°50.47'N	9°3.57'W	114	1268	L2	day
S16	18-7-09	CS106	41°42.05'N	9°4.06'W	106	2616	L2	night
S17	19-7-09	CS108	41°36.35'N	9°2.16'W	96	1390	L2	day
S18	19-7-09	CS119	41°29.17'N	9°3.1'W	90	1496	L2	night
S19	20-7-09	CS121	41°26.2'N	9°3.54'W	90	1898	L2	day
S20	20-7-09	CS131	41°23.18'N	9°4.05'W	102	2261	L2	night

1061

1062

2243
 2244
 2245
 2246
 2247
 2248
 2249
 2250
 2251
 2252
 2253
 2254
 2255
 2256
 2257
 2258
 2259
 2260
 2261
 2262
 2263
 2264
 2265
 2266
 2267
 2268
 2269
 2270
 2271
 2272
 2273
 2274
 2275
 2276
 2277
 2278
 2279
 2280
 2281
 2282
 2283
 2284
 2285
 2286
 2287
 2288
 2289
 2290
 2291
 2292
 2293
 2294
 2295
 2296
 2297
 2298
 2299
 2300
 2301

1063 **Table A.2.** Detailed information about the mesozooplankton samplings carried out during
 1064 CAIBEX III. Most of the samples were obtained from the core of the filament during the
 1065 third Lagrangian experiment (L3).
 1066

Sample	Date	CTD	Latitude	Longitude	Depth (m)	Volume (m ³)	Station	Day/night
G1	20-8-09	FIL7	10°19.99'N	30°50.06'W	1437	7501	upwelling	day
G2	21-8-09	FIL10	10°0.78'N	31°0.02'W	90	2247	coast	night
G3	23-8-09	FIL13	10°36.01'N	30°43.32'W	1837	5509	L3	day
G4	24-8-09	FIL17	10°43'N	30°49.20'W	1532	4423	L3	night
G5	24-8-09	FIL18	10°48.12'N	30°54.20'W	1671	7007	L3	day
G6	24-8-09	FIL20	10°30.10'N	30°59.93'W	808	6436	upwelling	night
G7	25-8-09	FIL21	10°46.77'N	30°55.82'W	1823	5057	L3	night
G8	25-8-09	FIL23	10°54.47'N	30°59.75'W	1640	6325	L3	day
G9	26-8-09	FIL26	10°19.92'N	30°49.53'W	1104	6592	upwelling	night
G10	27-8-09	FIL40	10°36.02'N	30°13.75'W	1974	5648	upwelling	night
G11	28-8-09	FIL41	11°16.75'N	31°0.37'W	2095	5290	L3	night
G12	28-8-09	FIL42	11°18.48'N	31°0.38'W	2200	5107	L3	day
G13	28-8-09	FIL44	11°31.33'N	31°26.42'W	2472	4965	ocean	day
G14	29-8-09	FIL44	11°22.50'N	30°57.27'W	2561	7735	ocean	night
G15	29-8-09	FIL46	11°25'N	30°57.25'W	2410	5534	L3	day
G16	29-8-09	FIL47	11°35.90'N	31°8.15'W	3042	5243	ocean	day
G17	29-8-09	FIL48	11°51.77'N	31°8.15'W	2688	6384	ocean	night
G18	30-8-09	FIL49	11°27.10'N	30°54.57'W	2505	6518	L3	night
G19	30-8-09	FIL51	11°36.18'N	31°8.27'W	2888	5473	ocean	day
G20	30-8-09	FIL52	11°54.11'N	31°9.89'W	2983	7214	ocean	night
G21	31-8-09	FIL53	11°30.88'N	30°52.70'W	2891	6188	L3	night
G22	3-9-09	FIL55	10°36.35'N	30°37.43'W	2006	7774	upwelling	day

1069 **Table A.3.** Morphological variables measured on the cephalopod paralarvae collected during
 1070 CAIBEX-I and III that were identified genetically: total length (TL), width (W), dorsal
 1071 mantle length (DML) and tentacle length (TeL).

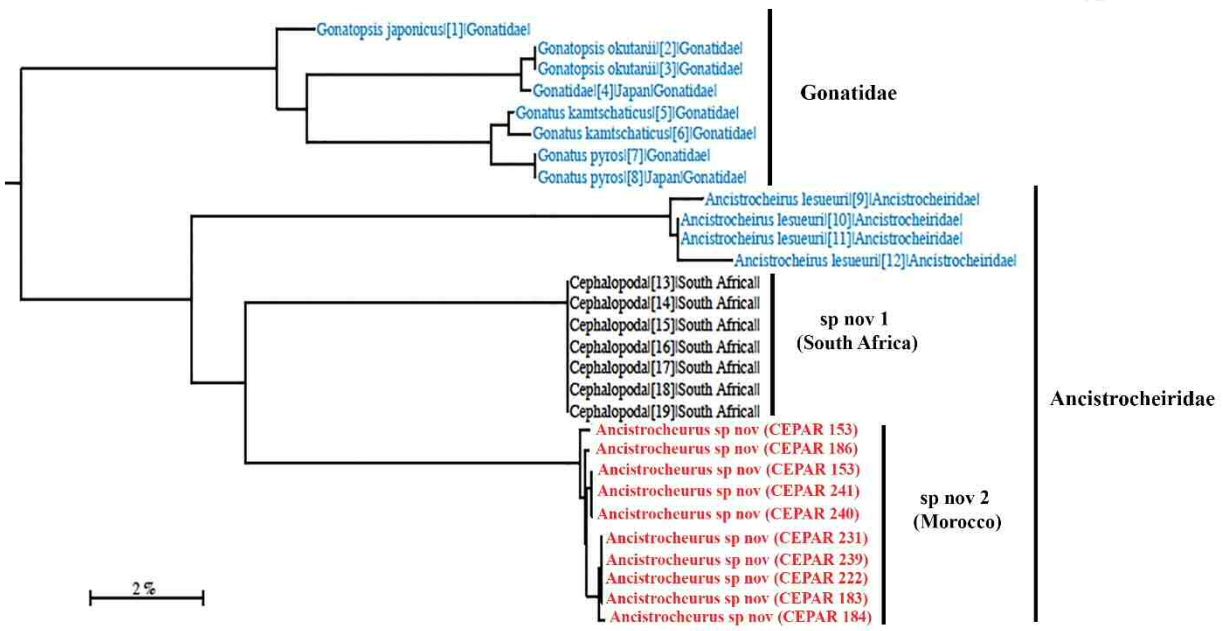
Survey	Species	n	TL	W	DML	TeL
CAIBEX I	<i>Alloteuthis media</i>	4	4.52 (2.88 – 5.52)	1.52 (1.13 – 1.9)	3.13 (2.11 – 3.66)	1.13 (0.61 – 1.61)
	<i>Alloteuthis subulata</i>	10	8.55 (3.59 – 12.22)	2.70 (1.52 – 3.77)	5.93 (2.67 – 8.17)	2.15 (0.67 – 3.43)
	<i>Illex coindetii</i>	1	3.66	1.63	2.72	0.90
	<i>Loligo vulgaris</i>	2	5.99 (5.18 – 6.79)	2.04 (1.84 – 2.25)	4.10 (3.69 – 4.51)	1.58 (1.34 – 1.81)
	<i>Octopus vulgaris</i>	88	2.58 (1.99 – 3.58)	1.42 (1.14 – 1.85)	1.89 (1.45 – 2.76)	
	<i>Sepiola tridens</i>	5	8.49 (4.97 – 18.18)	4.64 (3.22 – 9.32)	4.93 (3.22 – 9.77)	2.76 (1.17 – 7.09)
	<i>Todarodes sagittatus</i>	2	4.28 (2.48 – 6.08)	1.89 (1.53 – 2.26)	3.53 (2 – 5.07)	0.77 (0.58 – 0.97)
	<i>Todaropsis eblanae</i>	12	2.98 (2.37 – 5.36)	1.52 (0.98 – 2.88)	2.34 (1.65 – 3.88)	0.59 (0.35 – 1.2)
CAIBEX III	<i>Abraliopsis morisii</i>	8	7.03 (3.01 – 15.65)	1.34 (0.38 – 2.03)	3.25 (1.04 – 6.08)	4.00 (1.32 – 8.77)
	<i>Alloteuthis media</i>	100	5.45 (1.85 – 13.2)	1.59 (0.55 – 3.59)	3.69 (1.27 – 9.05)	1.30 (0.33 – 2.83)
	<i>Alloteuthis subulata</i>	8	7.42 (3.9 – 12.29)	2.38 (1.33 – 3.73)	5.25 (2.97 – 8.71)	1.54 (0.68 – 2.72)
	Ancistrocheiridae	10	4.97 (2.77 – 9.51)	1.58 (0.84 – 3.13)	3.07 (1.69 – 6.45)	1.71 (0.48 – 3.75)
	<i>Ancistroteuthis lichtensteinii</i>	18	4.62 (2.47 – 10.62)	1.33 (0.62 – 2.74)	3.43 (1.78 – 8.1)	1.00 (0.44 – 1.96)
	<i>Brachiooteuthis riisei</i>	18	7.39 (4.05 – 16.09)	1.8 (1.14 – 3.01)	5.19 (2.43 – 10.05)	1.89 (0.75 – 5.53)
	<i>Gonatopsis japonicus</i>	1	14.36	2.54	8.07	5.37
	<i>Heteroteuthis dispar</i>	2	7.31 (6.7 – 7.91)	3.13 (3.01 – 3.24)	2.65 (2.34 – 2.95)	3.88 (3.8 – 3.96)
	<i>Liocranchia reinhardtii</i>	2	14.33 (5.91 – 22.76)	2.41 (0.98 – 3.83)	8.74 (3.67 – 13.82)	5.48 (2.19 – 8.77)
	<i>Octopus vulgaris</i>	33	2.59 (1.66 – 5)	1.39 (0.99 – 2.74)	1.85 (1.21 – 3.04)	
	Ommastrephidae	1	1.79	0.51	0.93	0.69
	Pyroteuthidae	2	6.57 (3.88 – 9.27)	3.03 (1.51 – 4.66)	3.95 (2.26 – 5.63)	2.31 (1.31 – 3.31)
	<i>Pyroteuthis margaritifera</i>	11	5.87 (1.4 – 13.37)	2.45 (0.51 – 9.24)	3.28 (0.97 – 6.55)	1.66 (0.37 – 4.01)
	<i>Rondeletiola minor</i>	5	4.11 (1.93 – 5.97)	1.34 (0.58 – 2.12)	1.90 (0.98 – 2.69)	1.83 (0.78 – 2.9)
	<i>Sepiola atlantica</i>	1	5.26	2.01	3.01	1.73
<i>Sepiola ligulata</i>	1	5.25	2.40	2.90	1.72	

1072

1073

2361
2362
2363
2364
2365
2366
2367
2368
2369
2370
2371
2372
2373
2374
2375
2376
2377
2378
2379
2380
2381
2382
2383
2384
2385
2386
2387
2388
2389
2390
2391
2392
2393
2394
2395
2396
2397
2398
2399
2400
2401
2402
2403
2404
2405
2406
2407
2408
2409
2410
2411
2412
2413
2414
2415
2416
2417
2418
2419

1074 **Fig. A.1.** Kimura 2 parameter distance model tree obtained from BOLD database to assess
1075 the phylogenetic position of the ten Ancistrocheiridae paralarvae (in red) collected on
1076 CAIBEX-III. Sequences in black are private and cannot be accessed while sequences in blue
1077 are publicly available.



1079
1080
1081