



**HAL**  
open science

## **Molecular approach indicates consumption of jellyfish by commercially important fish species in a coastal Mediterranean lagoon**

Raquel M Marques, Audrey M. Darnaude, Sandrine Crochemore, Corinne Bouvier, Delphine Bonnet

### ► **To cite this version:**

Raquel M Marques, Audrey M. Darnaude, Sandrine Crochemore, Corinne Bouvier, Delphine Bonnet. Molecular approach indicates consumption of jellyfish by commercially important fish species in a coastal Mediterranean lagoon. *Marine Environmental Research*, 2019, 152, pp.104787. <10.1016/j.marenvres.2019.104787>. <hal-02371139>

**HAL Id: hal-02371139**

**<https://hal.science/hal-02371139v1>**

Submitted on 20 Jul 2022

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons CC BY-NC 4.0 - Attribution - Non-commercial use - International License

1 **Molecular approach indicate consumption of jellyfish by**  
2 **commercially important fish species in a coastal Mediterranean**  
3 **lagoon.**

4

5

6 *Raquel Marques<sup>1</sup>, Audrey M. Darnaude<sup>1</sup>, Sandrine Crochemore<sup>1</sup>, Corinne Bouvier<sup>1</sup>, Delphine*  
7 *Bonnet<sup>1</sup>*

8

9 <sup>1</sup> MARBEC, Univ Montpellier, CNRS, Ifremer, IRD, Montpellier, France

10

11

12 **Corresponding author:** raquel.fonseca-da-silva-marques@etu.umontpellier.fr

13 Tel: +33(0)769370312

14

15

16

17

18

19 **Key words:** Predation, *Aurelia coerulea*, Eel, Seabream, Polyps, Medusae, Quantitative PCR,  
20 Gut content, Thau lagoon

21

22

23 **Abstract**

24 Until recently, jellyfish have been ignored as an important source of food, due to their low nutritional  
25 value. Here, quantitative PCR was used to detect and quantify the DNA of the jellyfish *Aurelia*  
26 *coerulea* in the gut contents of commercially important fish species from the Thau Lagoon.  
27 Individuals from five fish species were collected during two different periods: the bloom period, when  
28 the pelagic stages of *A. coerulea* are abundant, and the post-bloom period, when only the benthic stage  
29 – polyps – is present in the lagoon. The DNA of *A. coerulea* was detected in the guts of 41.9% of the  
30 fish analysed, belonging to four different species. The eel *Anguilla anguilla* and the seabream *Sparus*  
31 *aurata* were important jellyfish consumers during the bloom and post-bloom periods, respectively.  
32 These results provide new insights on the potential control of jellyfish populations and on jellyfish  
33 importance as a food source for exploited fishes.

## 34 1. Introduction

35 Gelatinous organisms (scyphozoan, ctenophores, siphonophores, salps, pyrosomes and  
36 appendicularians, hereafter called jellyfish) are ubiquitous components of marine food webs and  
37 their noticeable outbreaks have been promoting recent research on their ecology. However,  
38 these studies have long focused on the drivers of jellyfish blooms (e.g. Purcell 2012) while little  
39 is known still on the causes of jellyfish mortality (Purcell and Arai 2001), although this  
40 information is fundamental though to understand their population dynamics.

41 So far, jellyfish were consistently considered as “dead ends” in marine food webs, due  
42 to their high water content and low nutritional value (e.g. 2.3-3.6 KJ.g.dry mass<sup>-1</sup> for *A. aurita*  
43 vs. 15.6 – 27.9 for various fishes, Doyle et al. 2007). They were largely believed to be ignored  
44 by most predators, with the exception of a few specialists, feeding exclusively on gelatinous  
45 organisms, such as the ocean sunfish (*Mola mola*), the butterfish (*Peprilus triacanthus*) and the  
46 leatherback turtle (*Dermochelys coriacea*) (Mianzan et al. 1996; Purcell and Arai 2001; Arai  
47 2005). Recent research though, has led to a shift in this paradigm (Hays et al. 2018).

48 Historically, diet assessments were performed by gut content analysis, which may  
49 provide biased information regarding jellyfish consumption as it gives excessive importance to  
50 hard prey that are more resistant to digestion (Hyslop 1980). Gelatinous organisms are digested  
51 rapidly and often destroyed or shrunk by preservative methods (Arai 2005). Although gut  
52 contents still provide new evidences of the importance of jellyfish as prey (Díaz Briz et al.  
53 2018), contemporary studies have been using new techniques to identify jellyfish predators,  
54 such as stable isotope analysis, animal-borne cameras, remotely operated vehicles and  
55 molecular analysis (Hays et al. 2018). Due to these modern techniques, the list of jellyfish  
56 predators has been growing and now includes commercially important fishes such as herring  
57 (*Clupea harengus*), whiting (*Merlangius merlangus*), bluefin tuna (*Thunnus thynnus*) and  
58 swordfish (*Xiphias gladius*) (Cardona et al. 2012; Lamb et al. 2017). Jellyfish were also shown  
59 to be of high importance in the diet of the larvae of a critically endangered fish, the European  
60 eel, *Anguilla anguilla* (Ayala et al. 2018) and to be ingested even by herbivorous fishes (Bos et  
61 al. 2016). Likewise, cephalopods, anemones, crabs, echinoderms and several species of birds  
62 have been reported to feed on jellyfish (Ates 2017; Hoving and Haddock 2017; McInnes et al.  
63 2017; Phillips et al. 2017; Thiebot et al. 2017). Jellyfish consumption apparently even occurs in  
64 deep benthic habitats of the Norwegian fjords, where mass falls of jellyfish carcasses can  
65 provide food for several scavengers, including the commercially exploited lobster *Nephrops*  
66 *norvegicus* (Sweetman et al. 2014; Dunlop et al. 2017).

67 The life cycle of many blooming jellyfish species (i.e. scyphozoans) is complex,  
68 though, comprising two pelagic stages (the young immature ephyrae and the sexually mature  
69 medusae) and an asexual reproductive benthic stage (the scyphistomae, hereafter called polyps).  
70 The magnitude of most jellyfish blooms is therefore dependent on the density of polyps and

71 ephyrae survival. Therefore, the mortality during these two early life stages may have a major  
72 effect on jellyfish population dynamics (Lucas et al. 2012; Fu et al. 2014). Insights on the  
73 predation on polyps and ephyrae and its potential impact on jellyfish outbreaks are still limited  
74 though (*e.g.* Ishii et al. 2004; Takao et al. 2014). In a recent laboratory experiment (Marques et  
75 al. 2016), the jellyfish *Aurelia coerulea* was found to be a potentially non-negligible source of  
76 food for an opportunistic fish, the gilthead seabream *Sparus aurata*. This fish was able to feed  
77 on all life stages of this jellyfish (including polyps) with potentially high ingestion rates.  
78 However, the predation of jellyfish by this fish has never been shown in the field, so far.

79 Jellyfish from the *Aurelia* Genus, are among the most common scyphozoans that form  
80 blooms (Dawson and Martin 2001; Mills 2001). They are widely distributed in coastal areas and  
81 semi-enclosed seas (Mills 2001). The Thau lagoon (NW Mediterranean, south coast of France)  
82 presents the rare particularity to harbour a completely resident population of *A. coerulea*,  
83 seemingly isolated from the Mediterranean Sea (Bonnet et al. 2012; Marques et al. 2015a). In  
84 this lagoon, *A. coerulea* ephyrae first appear in the early winter (in November - December) and  
85 grow during winter to give rise, at the beginning of spring (in March –April), to the adult  
86 medusae that form the annual bloom (Marques et al. 2015b). The medusae remain in the water  
87 column until the late spring (June), but disappear from the system afterwards. Polyps of *A.*  
88 *coerulea*, on the contrary, are found all year round in the lagoon (Marques et al. 2019), mainly  
89 settled on biofouling organisms, such as oysters and mussels (Marques et al. 2015a). Therefore,  
90 the Thau lagoon offers an ideal framework to investigate whether marine predators benefit from  
91 the jellyfish annual blooms and identify which life stages of *A. coerulea* are consumed in the  
92 field. This is imperative to address the role of predation in controlling jellyfish population  
93 dynamics and the potential importance of jellyfish as food for exploited fish species.

94 To this end, commercial fish species were sampled at different periods of the year and  
95 molecular analyses of their gut content were used to study their consumption of *A. coerulea*,  
96 during and after its local pelagic bloom.

97

## 98 **2. Material and Methods**

### 99 *2.1. Sampling and samples preparation*

100 Fishing is the oldest economic activity in the Thau lagoon, which is mainly performed  
101 by small enterprises (50 to 65 fishing boats operating in the lagoon), who target different  
102 species of fish using different fishing gears (CÉPRALMAR 2006). Different fish species were  
103 collected in the Thau lagoon by a professional fisherman. Fishes were collected during the  
104 annual bloom of *A. coerulea* (hereafter called bloom period, between April and June; Bonnet et  
105 al. 2012; Marques et al. 2015b) in 2012, 2013 and 2018, and during a period (from September to  
106 November) in 2018, when the pelagic stages of *A. coerulea* are not present in the lagoon  
107 (hereafter called post-bloom period, Bonnet et al. 2012; Marques et al. 2015b). During the

108 bloom period, fishes were collected by trammel nets, with an active fishing effort of maximum  
109 3h. During the post-bloom period, the traditional ‘capéchade’, which is the most used fishing  
110 technique in Thau (Crespi 2002), was used to collect fish for this study. The ‘capéchade’ is a  
111 fishing trap gear, placed at the same location for several days. Fishes are collected when the sun  
112 rises, after 24h of fishing effort. The number of species and individuals collected were therefore  
113 dependent on their occurrence in the nets. Immediately after collection, the fish were placed in  
114 separate plastic bags by the fisherman in order to avoid possible loss (or mixing) gut contents  
115 during sampling. Bags were then filled with absolute ethanol and stored in individual  
116 containers. Once in the laboratory, the fish were weighted (Total weight in g), dissected and  
117 their entire gut contents were removed and preserved at -30°C until DNA extraction. For  
118 positive DNA templates, samples of both the pelagic (medusae) and benthic (polyps) stages of  
119 *A. coerulea* were collected in the lagoon. Medusae were collected by a hand net and immediately  
120 preserved in absolute ethanol. Polyps attached to mussel shells were collected by SCUBA  
121 divers and transported in sea water to the laboratory. The samples were examined under a  
122 dissecting microscope (Olympus SZ40; Olympus KL 1500 LCD) and individual polyps were  
123 collected using needles. Fifty polyps were pooled per sample, frozen in liquid nitrogen and  
124 maintained at -30°C until DNA extraction.

125

## 126 2.2. DNA extraction

127 After thawing, the fish gut contents were mechanically ground in a mixer mill (MM400,  
128 Retsch). Three subsamples of 25 mg were collected from each gut content (when possible) and  
129 DNA was extracted using DNeasy blood and tissue kit (QIAGEN) (Stopar et al. 2010). The  
130 extraction was performed according to the instructions of the manufacturer, with an extra lysis  
131 step, performed overnight at 56°C. The same protocol was used for *A. coerulea* medusae  
132 samples, which were previously washed with pure molecular MilliQ water for ethanol removal.  
133 This protocol was, however, inefficient for the extraction of the polyp’s DNA, and therefore,  
134 their DNA was extracted by nucleic acid purification automated Maxwell ® instrument  
135 (Promega) and 16 LEV Blood DNA kit (Promega), with a modification of the lysis procedure,  
136 which was performed overnight at 56°C, using 30 µl of Proteinase K (Promega). In all cases, the  
137 extracted DNA was quantified in Nanodrop (NanoDrop One, Thermo Scientific).

138

## 139 2.3. Detection of *A. coerulea* DNA

140 Detection and quantification of *A. coerulea* DNA in the fish gut contents was performed  
141 by quantitative PCR (qPCR). This technique has been employed to detect and quantify the DNA  
142 of a specific prey in gut contents and faecal pellets, when traditional visual methods fails to do  
143 so (Matejusová et al. 2008; Nejstgaard et al. 2008; Töbe et al. 2010). This sensitive approach  
144 allows the detection and quantification of very small amounts of DNA so even highly digested

145 jellyfish can still be detected. All amplifications reactions were analysed using a Roche  
146 LightCycler 480 Real-Time thermocycler (qPHD-Montpellier GenomiX platform, Montpellier  
147 University, France). The total qPCR reaction volume was 1.5  $\mu\text{l}$  and consisted of 0.5  $\mu\text{l}$  DNA  
148 and 1  $\mu\text{l}$  LightCycler 480 SYBR Green I Master mix (Roche) with 0.6  $\mu\text{M}$  PCR primer  
149 (Eurofins Genomics). A 245 base pair gene fragment (partial sequences of mt-16S rDNA) was  
150 amplified by the species-specific (*A. coerulea*) primers AS3-F (5'-  
151 ATTGGTGACTGGAATGAATG - 3') and AS3-R (5'- TATGACAGCCCTTAGAGTTC - 3')  
152 designed by Wang et al. (2013). The best suited primer concentration (0.4, 0.6 or 0.8  $\mu\text{M}$ ) was  
153 determined in preliminary tests on three samples of *A. coerulea* polyps and medusae. A Labcyte  
154 Acoustic Automated Liquid Handling Platform (ECHO) was used for pipetting each component  
155 of the reaction mixture into a 384-well plate (Roche). The qPCR program consisted in an  
156 enzyme activation step at 95°C for 2 min, followed by 45 cycles of denaturation at 95°C for 10  
157 s, hybridization at 60°C for 10s and elongation at 72°C for 10 s. A final melting temperature  
158 curve ( $T_m$ ) of the amplicon was performed (95°C for 5 s and 65°C for 1 min), in order to ensure  
159 the specificity of the primers. The same amplification conditions and reaction concentrations  
160 were used in all assays performed in this study.

161         The efficiency and specificity of target gene detection by the primer was tested on a 2-  
162 fold dilution series of *A. coerulea* medusae and polyps positive templates. Triplicate reactions  
163 were performed at each dilution in order to generate the standard curves for each template. An  
164 ANCOVA analysis was performed, in order to assess if the efficiency (*i.e.* the slopes) of the two  
165 standard curves were significantly different. Absolute quantification of *A. coerulea* DNA in the  
166 fish gut contents was estimated using the regression equation of the standard curve obtained for  
167 the polyps positive template. The observed  $C_p$  values of each dilution of the positive template  
168 were plotted against its known DNA concentration to obtain the regression equation. The  $C_p$   
169 value is defined as the cycle when the sample fluorescence exceeds the threshold above the  
170 background fluorescence. The  $C_p$  value is therefore related with the amount of DNA present in  
171 the sample (Dorak 2006).

172         The quantification of DNA in fish gut contents was only performed in the samples  
173 showing  $C_p$  values below 31, which was found to be the  $C_p$  correspondent to the minimum  
174 quantifiable concentration ( $1.37 \times 10^{-4} \text{ ng } \mu\text{L}^{-1}$ ). Samples with the same  $T_m$  values as the  
175 positive templates ( $T_m$  peak at 81.5) and  $C_p$  values between 31 and 32.62 (maximum  $C_p$   
176 observed for the positive templates) were considered as positive detection but non-quantifiable.  
177 Samples showing  $C_p$  values above 32.62 were considered as negative detection. For each gut  
178 content sample, a minimum of 9 replicates (3 experimental replicates, *i.e.* for DNA extraction,  
179 of the same gut content and 3 technical replicates for each experimental replicate), were  
180 performed, except for some samples with very low material, from which only one experimental

181 replicate was collected. Only gut contents samples that showed positive detection in at least two  
182 technical replicates were considered to contain *A. coerulea* DNA.

183 The specificity of the primers and the detection of *A. coerulea* DNA was further  
184 confirmed by sequencing the positive templates and the qPCR product of 16 samples with  
185 positive amplifications. For that, 10  $\mu$ L of molecular MilliQ water was added to the qPCR  
186 products. The DNA was purified using a commercial kit (QIAquick PCR Purification kit,  
187 QIAGEN), following the manufacturer protocol. The purified DNA was amplified by traditional  
188 PCR using PCR kit illustra puretaq ready to go (GE Healthcare), with 5  $\mu$ L of purified DNA  
189 sample, 0.6  $\mu$ M of each forward and reverse primers (primer pair AS3) and 27.5 $\mu$ L of molecular  
190 MilliQ water. The thermal profile for the PCR reaction was composed by 3 min at 95°C, 35  
191 cycles of 1 min at 95°C, 1 min at 60°C and 90 s at 72°C, followed by 1min at 72°C (Stopar et  
192 al. 2010; Ramšak et al. 2012). The products of PCR reactions were analysed through  
193 electrophoresis (Mupid-One; Advance) at 100V for 30min in 0.5 X TAE buffer (Euromedex).  
194 An aliquot of 3  $\mu$ L of samples were load on Agarose gel 1.5%, using loading buffer  
195 (AppliChem, Panreac) and 1Kb DNA ladder (Euromedex). Gels were stained with GelRed ®  
196 Nucleic Acid Gel Stain (Biotium), visualized and photographed on UV table using Molecular  
197 Imager Gel Doc™ XR System (Bio-Rad) for quality control of DNA amplification. Sequencing  
198 was performed at the genotyping and sequencing facilities in Montpellier University. The  
199 purification of PCR products was performed by magnetic beads, using the CleanPCR kit  
200 (GCBiotech), performed by an automated liquid handler (Biomek 4000, Beckman Coulter).  
201 Sequencing was then performed with 55-60 ng of DNA using the BigDye Terminator Cycle  
202 sequencing v3.1 kit (Life Technologies), with the following PCR program: 3 min at 96°C, 25  
203 cycles of 10 sec at 96°C, 5 sec at 50°C and 4min at 60°C. The products of the sequencing  
204 reaction were purified using magnetic beads, following the same protocol as previously  
205 described. The purified products were then analysed on an ABI 3500 xL capillary sequencer  
206 (Applied Biosystems, Darmstadt, Germany). A BLAST analysis of the resulting sequences  
207 against the GenBank nucleotide database was performed.

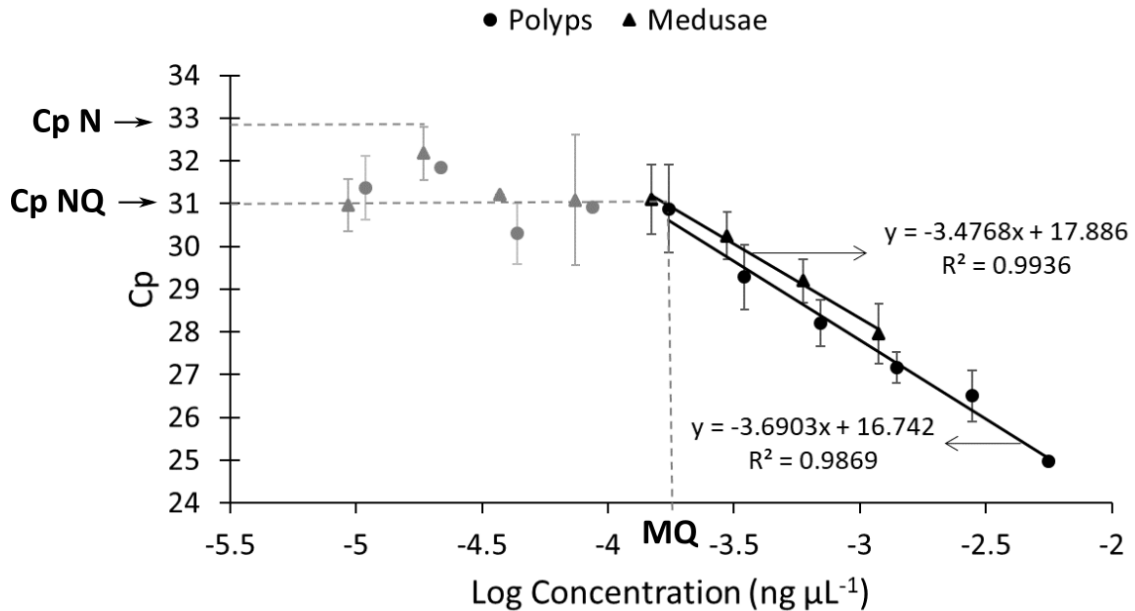
208

### 209 **3. Results**

#### 210 *3.1. Standard curve and DNA quantification*

211 Both positive templates (polyps and medusae) were identified as *A. coerulea*, after the  
212 BLAST analysis. The standard curves of the qPCR assay (Fig. 1), determined with polyps and  
213 medusae positive templates, showed high assay efficiencies (86.64 and 93.80 %, respectively)  
214 and high correlation coefficient,  $R^2$  (both 99%). The slope of both positive templates did not  
215 differ significantly (ANCOVA;  $F = 0.03$ ,  $P = 0.85$ ). However, the initial template concentration  
216 of polyps was higher (55.79 ng  $\mu$ L<sup>-1</sup>) than that of medusae (11.89 ng  $\mu$ L<sup>-1</sup>) and therefore, more  
217 dilution steps showed  $C_p$  values below 31. In consequence, the standard curve of polyps

218 comprises more dilution steps and wider range of quantifiable template concentration,  
 219 increasing the accuracy of the regression fit. For this reason, its correspondent equation ( $y = -$   
 220  $3.69 x + 16.74$ ) was selected to estimate the concentration of *A. coerulea* DNA in the fish gut  
 221 content.  
 222



223  
 224 **Fig. 1:** Standard curves determined from 2-fold dilutions of polyps (circles) and medusae (triangles)  
 225 positive templates. The dilutions included in the standard curve of each template are represented in black,  
 226 while the dilutions excluded from the standard curve are in grey. In each case, the standard curve equation  
 227 is shown, but only that of the polyp's template was used to estimate DNA concentration in fish gut  
 228 contents (efficiency of 86.63%). The minimum quantifiable concentration ( $MQ = 1.37 \times 10^{-4} \text{ ng } \mu\text{L}^{-1}$ )  
 229 corresponded to a Cp of 31 (*i.e.* the threshold for quantification; Cp NQ). Samples with Cp values  
 230 between 31 (Cp NQ) and 32.62 (*i.e.* the threshold for detectability; Cp N) were considered positive but  
 231 Non-quantifiable. Samples with Cp values above Cp N were considered negative (see methods section for  
 232 further information). Error bars are standard deviations.

233  
 234 Although the more diluted samples of the positive template for *A. coerulea* polyps and  
 235 medusae were positive (*i.e.* with proper melting curves), they showed low Cp values, indicating  
 236 that their DNA concentrations were too low to be accurately quantified. Therefore, those  
 237 dilutions were excluded from the standard curve. Among the three technical replicates analysed  
 238 for each dilution sample, six false negatives (*i.e.* deviated  $T_m$  peak values) were observed.  
 239 Although non-quantifiable, *A. coerulea* DNA was still detected at a maximum Cp of 32.62,  
 240 which was therefore considered as the threshold of detectability.

241 The BLAST analysis revealed that all sequenced qPCR products of gut contents  
 242 samples matched the previously designated *Aurelia* sp.1 (> 96.7% similarity), recently accepted  
 243 as *A. coerulea* (Scorrano et al. 2016).

244

245 *3.2. Fish ingestion of A. coerulea*

246 During the period of *A. coerulea* bloom (from April to June) 50 fish individuals were  
 247 provided by the fisherman. They belonged to five different species: the European eel (*Anguilla*  
 248 *anguilla*, Linnaeus, 1758), the sand smelt (*Atherina boyeri*, Risso, 1810), the golden mullet  
 249 (*Liza aurata*, Risso, 1810), the salema (*Sarpa salpa*, Linnaeus, 1758) and the gilthead sea bream  
 250 (*Sparus aurata*, Linnaeus, 1758) (Table 1). During the post-bloom period, when only polyps are  
 251 present in the lagoon (September to November), only 12 individuals could be collected for this  
 252 work. They belonged to three different species: the golden mullet (*L. aurata*), the salema (*S.*  
 253 *salpa*) and the gilthead sea bream (*S. aurata*).

254

255 **Table 1:** Numbers of fish gut contents analysed (N) and of fish guts with positive detection of *A. coerulea*  
 256 DNA (N Positives). In each case, the species, the range of weight and length of the sampled fish are  
 257 indicated with the sampling period, from April to June (bloom) and from July to November (Post-bloom).

Period	Common name	Fish Species	Weight (g)	Length (mm)	N	N Positives (%)
Bloom	European eel	<i>Anguilla anguilla</i>	4.8 <sup>a</sup>	150 <sup>b</sup>	10	10 (100 %)
Bloom	Sand smelt	<i>Atherina boyeri</i>	0.41 - 8.1 <sup>c</sup>	40 - 99 <sup>c</sup>	5	0 (0 %)
Bloom	Golden mullet	<i>Liza aurata</i>	251.2 - 900	306.0 - 488.4 <sup>a</sup>	12	4 (33.3 %)
Bloom	Salema	<i>Sarpa salpa</i>	260.6 - 650	263.7 - 360.2 <sup>a</sup>	11	1 (9.1 %)
Bloom	Gilthead sea bream	<i>Sparus aurata</i>	133.6 - 300	95.5 - 126.7 <sup>a</sup>	12	4 (33.3 %)
Post-bloom	Golden mullet	<i>Liza aurata</i>	219 - 660.7	291.0 - 436.1 <sup>a</sup>	3	1 (33.3 %)
Post-bloom	Salema	<i>Sarpa salpa</i>	219.5 - 324.1	248.7 - 284.1 <sup>a</sup>	2	1 (50 %)
Post-bloom	Gilthead sea bream	<i>Sparus aurata</i>	159.9 - 234.6	101.7 - 116.3 <sup>a</sup>	7	5 (71.4 %)

258

<sup>a</sup> Calculated from length-weight relationships (Melià et al. 2006; Crec'hriou et al. 2012)

259

<sup>b</sup> Data not collected during the study, an approximate length of the individuals is provided.

260

<sup>c</sup> Data not collected during the study, but data from individuals collected in the Thau lagoon during the same period of the year is provided.

261

262

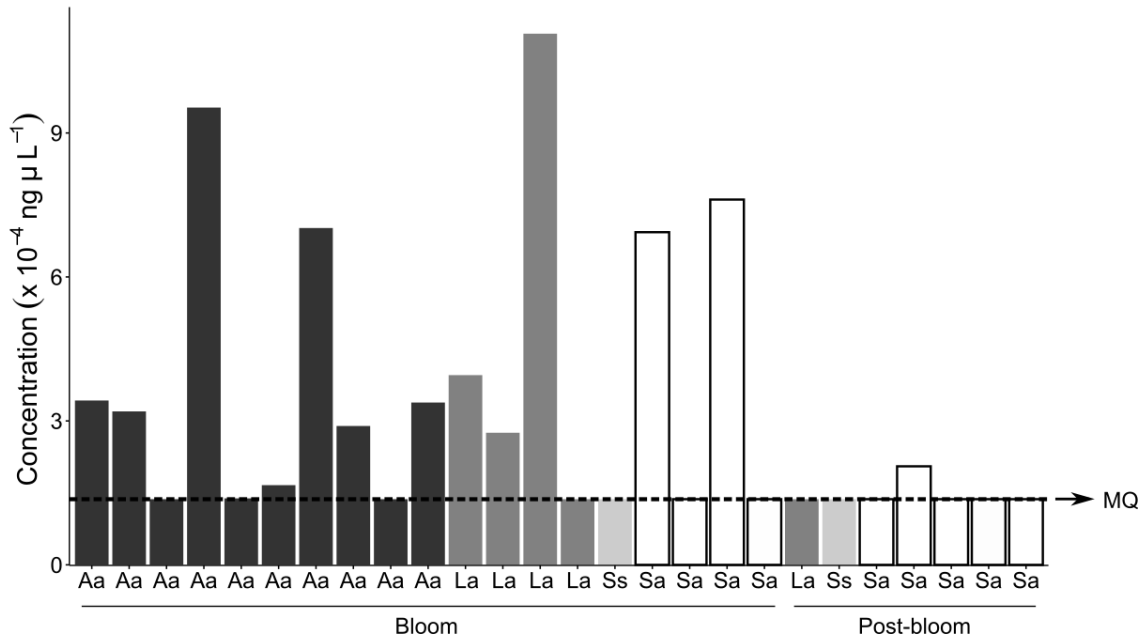
263 In total, DNA from *A. coerulea* was detected in the gut content of 26 fish (41.9 % of the  
 264 62 individuals analysed), among which 73% had been collected during the bloom period and  
 265 27% during the post-bloom period (Table 1). With the exception of the sand smelt, all species  
 266 were shown to have consumed *A. coerulea*, irrespective of the period of sampling. During the  
 267 bloom period, medusae DNA was detected in the gut contents of all the eels collected (10  
 268 individuals). One third of the golden mullets and gilthead sea breams analysed were also shown  
 269 to have consumed *A. coerulea*, while positive detection was only observed in one individual of  
 270 salema (9.1%). During the post-bloom period, *A. coerulea* was detected in only one golden  
 271 mullet, one salema, and in 5 (71.4 %) gilthead sea bream.

272

273 The concentration of DNA in the fish gut contents was higher during the bloom than in  
 274 the post-bloom period (Fig. 2). At this time of the year 63.2 % of the jellyfish consumers had a  
 sufficient amount of DNA to be quantified (*i.e.* > 1.37 x 10<sup>-4</sup> ng µL<sup>-1</sup>). The maximum

275 concentration ( $11.1 \times 10^{-4} \text{ ng } \mu\text{L}^{-1}$ ) was detected in a golden mullet, but four other fish (two eels  
 276 and two gilthead sea breams) showed DNA concentrations above  $4 \times 10^{-4} \text{ ng } \mu\text{L}^{-1}$  in their gut  
 277 contents. During the post-bloom period, the concentration of DNA in the guts was very low and,  
 278 in most cases, non-quantifiable (Fig. 2).

279  
 280



281

282 **Fig. 2:** Estimated concentration of *A. coerulea* DNA in the gut contents of the fish with positive  
 283 detection: (Aa) European eel (*Anguilla anguilla*), (La) Golden mullet (*Liza aurata*), (Ss) Salema (*Sarpa*  
 284 *salpa*), (Sa) Gilthead sea bream (*Sparus aurata*). The horizontal dashed line (MQ) indicate the minimum  
 285 quantifiable DNA concentration ( $1.37 \times 10^{-4} \text{ ng } \mu\text{L}^{-1}$ , *i.e.* Cp = 31; see methods section for further  
 286 information), below which the detection was positive but non-quantifiable.

287

#### 288 4. Discussion

289 The present work brings new insights on the prey-predator relationships between fish  
 290 and jellyfish in the Thau lagoon. Indeed, four of the five fish species analysed in this study were  
 291 found to feed on *A. coerulea*. For some species, all the individuals tested had *A. coerulea* DNA  
 292 in their gut, suggesting that this jellyfish might be a non-negligible source of food for  
 293 commercial fish in the Thau lagoon.

294 The concentration of the target DNA in the gut contents was frequently low, with many  
 295 individuals showing non-quantifiable DNA concentrations. This is not very surprising because  
 296 jellyfish are rapidly digested in fish guts, compared to other prey (*e.g.* > 93% of the jellyfish  
 297 biomass can be digested within 1h in controlled laboratory studies, Arai et al. 2003). In  
 298 particular, due to the fishing method used, most of the fish captured during the post-bloom  
 299 period, probably had largely digested their prey during their prolonged captivity in the net.  
 300 Therefore, we consider our results to be conservative and likely to underestimate *A. coerulea*

301 consumption by commercial fish in the Thau lagoon. However, since only few individuals of  
302 each species were analysed, especially during the post-bloom period, additional studies are  
303 needed to confirm the actual importance of *A. coerulea* as a source of food for fishes in the  
304 Thau lagoon.

305 During *A. coerulea* bloom periods, the only fish species which did not seem to consume  
306 jellyfish was the sand smelt. The diet of this species is opportunistic but mainly based on  
307 pelagic organisms such as zooplankton, phytoplankton, arachnids, insects and fish larvae  
308 (Vizzini and Mazzola 2005, Dias et al 2014, Yagci et al 2018). However, due to the small size  
309 of the specimens examined (< 7cm), they might have avoid jellyfish blooms, since they may  
310 become prey for large jellyfish medusae.

311 In contrast, the European eel was shown to be a potential important consumer of  
312 jellyfish in the Thau lagoon. All the individuals tested showed positive detection of *A. coerulea*  
313 in their gut contents, with relatively high DNA concentrations. The consumption of gelatinous  
314 organisms by eels was also previously reported for their larvae (leptocephali) in the Sargasso  
315 Sea (Riemann et al. 2010; Ayala et al. 2018). Although there is a progressive ontogenic change  
316 in the diet of the eels (Costa et al. 1992; Provan and Reynolds 2000), it is not surprising that  
317 they retain the ability to feed on gelatinous organisms. After their migration from their  
318 spawning areas in Sargasso Sea (Ginneken and Maes 2005), European eels reach the  
319 Mediterranean lagoons as glass eels, with an average length of ca. 60–65 mm (Melià et al.  
320 2006). At this stage, eels are considered to be non-feeding, starting to feed only when reaching  
321 the elver eel stage (Tesch et al. 2003). The individuals collected in this study were all at the  
322 elver stage (*i.e.* pigmented with a length of ca. 150 mm). The diet at this stage is mostly based  
323 on small sized prey like amphipods, isopods, mysids and insects (Costa et al. 1992; Provan and  
324 Reynolds 2000) but it depends highly on prey availability (Costa et al. 1992; Bouchereau et al.  
325 2006). Therefore, it is likely that the high abundances of jellyfish during the bloom might result  
326 in their ingestion by this opportunistic species.

327 The rate of standard metabolism of an European eel at 25°C (*in situ* temperature in June  
328 2013 was  $20 \pm 0.7$  °C) is  $83.3 \text{ J g}^{-1} \text{ day}^{-1}$  (Owen et al. 1998). Assuming a similar energy  
329 requirement for the individuals collected in Thau, an eel with 4.72 g (estimated for a 15 cm eel,  
330 from length-mass relationships; Carss et al. 1999), would require  $0.4 \text{ KJ d}^{-1}$  of energy. One  
331 gram (wet weight) of *Aurelia* sp. provides 0.1 KJ of energy (Arai 1997 in Doyle et al. 2007,  
332 after wet weight estimation according to Lucas 1994). Therefore, one eel would require 3.8 g of  
333 medusae wet weight per day to meet its energy requirements. The eels analysed in this study  
334 were collected in June 2013, when the abundance of medusae was at its highest ( $75.5 \text{ ind } 100 \text{ m}^{-3}$ ;  
335 Marques et al. 2015b). Because medusae are big in this time of the year ( $16.4 \pm 2.8 \text{ cm}$ ;  
336 Marques et al. 2015b, which corresponds to 195.1 g of wet weight, estimated after Hirst and  
337 Lucas 1998, at the same salinity conditions), it is possible that the eels had bitten their

338 umbrellas, taking advantage of the soft consistency of jellyfish body. Indeed, many jellyfish  
339 predators do not ingest the whole medusae, but instead, they bite the umbrella margins and/or  
340 select particular parts of the medusae with higher nutritional values, such as gonadal tissue  
341 (Milisenda et al. 2014; Marques et al. 2016; Dunlop et al. 2017; Hoving and Haddock 2017).  
342 Therefore, in theory, one medusae could provide enough energy to sustain the standard  
343 metabolism of one eel for 51 days. Even though a large amount of jellyfish consumption is  
344 needed to meet such energy requirements (*i.e.* 80.5 % of the eel weight per day), the rapid  
345 digestion and gut clearance rates (Arai et al. 2003) allow the fish to increase its ingestion rates.  
346 Similar results were also reported for the leptocephali stage of the European eel (Ayala et al.  
347 2018) and for other commercially important organisms (*e.g.* fish top predators, eel larvae,  
348 lobsters, deep water octopus), which, during blooms, jellyfish are able to meet and maybe  
349 overcome the entire energy requirements of these predators (Cardona et al. 2012; Dunlop et al.  
350 2017; Hoving and Haddock 2017). Here we confirm the potential important role of jellyfish as  
351 food for young stages of the European eel. These results are of great importance since the  
352 European eel is listed as a critically endangered species by IUCN (Freyhof and Kottelat 2010)  
353 and information regarding its diet is still limited.

354         The consumption of *A. coerulea* during its bloom period was also recorded for the  
355 gilthead sea bream and the golden mullet, both species showing, in some cases, high *A. coerulea*  
356 DNA concentration in their gut contents. This result is not surprising for sea bream as this  
357 species has been shown to prey on all life stages of *A. coerulea* in laboratory experiments, with  
358 high ingestion rates of polyps and small medusae (Marques et al. 2016). The sea breams with  
359 the highest concentrations of the target DNA in their guts were collected in April 2013, when  
360 medusae bell diameter is < 3 cm (Marques et al. 2015b). In the laboratory, small medusae (1  
361 cm bell diameter) were preferred by this fish, but larger ones (up to 8 cm bell diameter) were  
362 also preyed upon, by taking several bites on the edge of their umbrella (Marques et al. 2016).  
363 Therefore, our results provide evidence of a possible active predation of sea bream individuals  
364 on pelagic jellyfish in the field. Jellyfish, though, were not selected in the laboratory when prey  
365 with higher nutritional value were equally available (Marques et al. 2016). In the field, gilthead  
366 seabreams prey mainly on polychaetes, small fishes, crustaceans, gastropods and bivalves but  
367 adapt their diet to local prey availability (Pita et al. 2002; Escalas et al. 2015). Therefore, we  
368 suspect that the high abundance and accessibility of *A. coerulea* medusae during the bloom  
369 periods, benefit this opportunistic predator by providing a suitable source of food when its  
370 preferred prey are less accessible (Marques et al. 2016; Díaz Briz et al. 2018).

371         More surprisingly, one third of the golden mullet specimens analysed had the target  
372 DNA in their gut contents. Mulletts are detritivores, eating a mixture of sand, detritus,  
373 microphytobenthos, macroalgae, zooplankton and benthic macrofauna (Laffaille et al. 2002;  
374 Almeida 2003). To our knowledge, the consumption of jellyfish by this species has never been

375 described so far. It is possible that *A. coerulea* was consumed unintentionally, since dead  
376 medusae are occasionally found decomposing on soft bottoms (Marques, personal observation)  
377 and the resulting organic matter may be incorporated in the surface sediment layer. One  
378 individual of this species, though, showed high concentration of the target DNA in its gut  
379 content (the maximum concentration recorded in this study). Although, the active predation of  
380 jellyfish by the golden mullet cannot be excluded, this particular individual was collected during  
381 the peak of *A. coerulea* abundance (in May 2018), when high biomass of jellyfish was also  
382 caught in the fishing nets (J. Fabrice, personal communication). Therefore, the high  
383 concentration of *A. coerulea* DNA in its gut contents might have been the consequence of its  
384 unintentional ingestion of medusae in the fishing net during sampling.

385         The ingestion of *A. coerulea* by salema might also be unintentional. Indeed, this species  
386 has been described so far as a true herbivore, with a diet largely based on seagrass leaves  
387 (Havelange et al. 1997). Because decaying medusae are also occasionally observed entangled  
388 among the seagrass leaves in the Thau lagoon (R. Marques, personal observation), they might  
389 have been ingested together with the target seagrass leaves. However, recent observations have  
390 shown that even herbivorous fish may actively prey on jellyfish (Bos et al. 2016), which cannot  
391 be excluded here. Still, additional individuals of this species should be analysed to test this  
392 hypothesis.

393         The consumption of *A. coerulea* was also observed when its pelagic stages were absent,  
394 which suggests that polyps might also be ingested by commercial fish species in Thau. The  
395 most important consumer of polyps in our study was the gilthead seabream (71.4% of the  
396 individuals showed positive detection of *A. coerulea* DNA in their gut contents). In previous  
397 laboratory experiments (Marques et al. 2016), the sea bream was shown to consume polyps,  
398 likely in an indirect way. In this recent study, it was suggested that the actual target of the fish  
399 during the predation experiments, could have been the settling substrate of polyps (*i.e.* living  
400 mussels), rather on the polyps themselves. In Thau, most of the polyps of *A. coerulea* are found  
401 fixed on oysters or mussel shells (Marques et al. 2015a). Because bivalves are highly important  
402 in the diet of adult sea bream (Pita et al. 2002; Tancioni et al. 2003; Russo et al. 2007) and very  
403 abundant in Thau lagoon, we hypothesize that the consumption of *A. coerulea* polyps in the  
404 field results from an indirect ingestion, when fishes are preying on their settling substrates. In  
405 addition, pieces of mussel shells were recurrently observed in the sea bream gut contents. These  
406 findings highlight the likely underestimated impact of sea bream predation on the regulation of  
407 the benthic population of *A. coerulea* in the lagoon, potentially contributing to the reduction of  
408 medusae abundances and to the magnitude of jellyfish blooms in this lagoon.

409         *A. coerulea* polyps consumption was also detected for both the salema and the golden  
410 mullet. This is surprising, since polyps generally settle on the underside surface of hard  
411 substrates and have never been found on soft sediments or fixed to seagrass leaves (Marques et

412 al. 2015a). Even though unintentional consumption can not be excluded, very few individuals of  
413 these two species were analysed in this study. Therefore, the importance of the consumption of  
414 *A. coerulea* polyps by their individuals in the lagoon still needs further investigation.

415

## 416 **5. Conclusion**

417 Our results demonstrate that the jellyfish *A. coerulea* is ingested by several  
418 commercially important fishes in Thau lagoon during its bloom period, when the abundance and  
419 biomass of its pelagic stages are high, but also in post-bloom periods, when only *A. coerulea*  
420 polyps are present in this semi-enclosed ecosystem. This provides evidence that the  
421 vulnerability of jellyfish to fish predation has been underestimated in the lagoon but also,  
422 potentially, elsewhere. Indeed, predation pressure by a large number of fish species with broad  
423 diets is more ecologically important than that by a few specialized ones (Purcell and Arai 2001;  
424 Arai 2005). Here we highlight potential ecological implications for both fish and jellyfish  
425 ecology. On the one hand, direct predation on jellyfish pelagic stages or indirect predation on  
426 polyps might contribute to control jellyfish blooms, through top-down regulation. In this sense,  
427 the overexploitation of fish stocks might contribute to the increase of jellyfish outbreaks, by  
428 releasing the predation pressure over jellyfish populations (Roux et al. 2013). On the other hand,  
429 the availability and accessibility of jellyfish during their blooms provide an alternative food  
430 source for fish populations, that might actively consume jellyfish when their primary prey are  
431 less available (Diaz Briz et al. 2018; Mianzan et al 2001).

432

433 **6. Acknowledgment:**

434 We thank the fisherman Jean Fabrice for his collaboration in fish collection and samples  
435 preparation in the field and Dr. Jean Antoine Tomasini for helping in fish dissection. A special  
436 thanks to Dr. Julien de Lorgeril for his expertise and contribution in qPCR sample and data  
437 analysis. We also thank Philippe Claire and Jean Luc Rolland for their technical support and  
438 expertise during qPCR analysis and DNA purification procedures. All qPCR analyses were  
439 performed at the qPHd platform, University of Montpellier/ Montpellier Genomix. This work  
440 has benefited from the facilities and expertise of the MARBEC Technical Pole. The sequencing  
441 analysis was performed at the genotyping and sequencing facilities of ISEM (Institut des  
442 Sciences de l'Evolution-Montpellier) and Labex Centre Méditerranéen Environnement  
443 Biodiversité. This project was funded by the EC2CO project AO2014\_911620 Dynamo (P.I.:  
444 Dephine Bonnet). Finally, we would like to thank the two reviewers for their comments and  
445 suggestions during the revision process.

446

447 **7. Declaration of interest: none**

448

449

450 **8. References:**

- 451 Almeida PR (2003) Feeding ecology of *Liza ramada* (Risso, 1810) (Pisces, Mugilidae) in a  
452 south-western estuary of Portugal. *Estuar Coast Shelf Sci* 57:313–323. doi:  
453 10.1016/S0272-7714(02)00357-8
- 454 Arai MN (2005) Predation on pelagic coelenterates: a review. *J Mar Biol Assoc UK* 85:523–  
455 536. doi: 10.1017/S0025315405011458
- 456 Arai MN, Welch DW, Dunsmuir AL, Jacobs MC, Ladouceur AR (2003) Digestion of pelagic  
457 Ctenophora and Cnidaria by fish. *Can J Fish Aquat Sci* 60:825–829. doi: 10.1139/f03-071
- 458 Ates RML (2017) Benthic scavengers and predators of jellyfish , material for a review. *Plankt*  
459 *Benthos Res* 12:71–77. doi: <https://doi.org/10.3800/pbr.12.71>
- 460 Ayala DJ, Munk P, Lundgreen RBC, Traving SJ, Jaspers C, Jørgensen TS, Hansen LH,  
461 Riemann L (2018) Gelatinous plankton is important in the diet of European eel (*Anguilla*  
462 *anguilla*) larvae in the Sargasso Sea. *Sci Rep* 8:6156:1–10. doi: 10.1038/s41598-018-  
463 24388-x
- 464 Bonnet D, Molinero J-C, Schohn T, Daly-Yahia MN (2012) Seasonal changes in the population  
465 dynamics of *Aurelia aurita* in Thau lagoon. *Cah Biol Mar* 53:343–347.
- 466 Bos AR, Cruz-Rivera E, Sanad AM (2016) Herbivorous fishes *Siganus rivulatus* (Siganidae)  
467 and *Zebrasoma desjardini* (Acanthuridae) feed on Ctenophora and Scyphozoa in the Red  
468 Sea. *Mar Biodivers* 47:243–246. doi: 10.1007/s12526-016-0454-9
- 469 Bouchereau J, Marques C, Pereira P, Guelorget O, Vergne Y (2006) Trophic characterization of  
470 the Prévost lagoon (Mediterranean Sea) by the feeding habits of the European eel *Anguilla*  
471 *anguilla*. *Cah Biol Mar* 47:133–142. doi: 10.21411/CBM.A.DAEAA449
- 472 Cardona L, de Quevedo IÁ, Borrell A, Aguilar A (2012) Massive consumption of gelatinous  
473 plankton by mediterranean apex predators. *PLoS One*. doi: 10.1371/journal.pone.0031329
- 474 Carss DN, Elston DA, Nelson KC, Kuuk H (1999) Spatial and temporal trends in unexploited  
475 yellow eel stocks in two shallow lakes and associated streams. *J Fish Biol* 55:636–654.  
476 doi: <https://doi.org/10.1111/j.1095-8649.1999.tb00704.x>
- 477 CÉPRALMAR (2006) Suivi de la pêche aux petits métiers. Languedoc Roussillon, France
- 478 Costa JL, Assis CA, Almeida PR, Moreira FM, Costa MJ (1992) On the food of the European  
479 eel, *Anguilla anguilla* (L.), in the upper zone of the Tagus estuary, Portugal. *J Fish Biol*  
480 41:841–850. doi: <https://doi.org/10.1111/j.1095-8649.1992.tb02712.x>
- 481 Crec’hriou BR, Neveu R, Lenfant P (2012) Technical contribution. Length – weight relationship  
482 of main commercial fishes from the French Catalan coast. *J Appl Ichthyol* 105:1–2. doi:  
483 10.1111/j.1439-0426.2012.02030.x
- 484 Crespi V (2002) Recent evolution of the fishing exploitation in the Thau lagoon, France. *Fish*  
485 *Manag Ecol* 19–29. doi: <https://doi.org/10.1046/j.1365-2400.2002.00275.x>
- 486 Dawson MN, Martin DL (2001) Geographic variation and ecological adaptation in *Aurelia*

487 *aurita* (Scyphozoa, Semaestomeae): some implications from molecular phylogenetics.  
488 *Hydrobiologia* 451:259–273. doi: <https://doi.org/10.1023/A:1011869215330>

489 Díaz Briz L, Sánchez F, Marí N, Genzano G (2018) Seasonal variation of gelatinous plankton  
490 consumption by fish in the South-western Atlantic Ocean : a question of strategy. *Mar Biol*  
491 *Res.* doi: 10.1080/17451000.2018.1508847

492 Dorak MT (2006) *Real-time PCR*. Taylor & Francis Group, Newcastle-upon-Tyne, UK, ISBN:  
493 0-4153-7734-X

494 Doyle TK, Houghton JDR, McDevitt R, Davenport J, Hays GC (2007) The energy density of  
495 jellyfish: Estimates from bomb-calorimetry and proximate-composition. *J Exp Mar Bio*  
496 *Ecol* 343:239–252. doi: 10.1016/j.jembe.2006.12.010

497 Dunlop KM, Jones DOB, Sweetman AK (2017) Direct evidence of an efficient energy transfer  
498 pathway from jellyfish carcasses to a commercially important deep-water species. *Sci Rep*  
499 7:17455. doi: 10.1038/s41598-017-17557-x

500 Escalas A, Ferraton F, Paillon C, Vidy G, Carcaillet F, Salen-Picard C, Le Loc'h F, Richard P,  
501 Darnaude AM (2015) Spatial variations in dietary organic matter sources modulate the  
502 size and condition of fish juveniles in temperate lagoon nursery sites. *Estuar Coast Shelf*  
503 *Sci* 152:78–90. doi: 10.1016/j.ecss.2014.11.021

504 Freyhof J, Kottelat M (2010) *Anguilla anguilla*. The IUCN Red List of Threatened Species  
505 2010: e.T60344A45833138.DOI: <http://dx.doi.org/10.2305/IUCN.UK.2014->  
506 1.RLTS.T60344A45833138.en. Accessed on 24 May 2019.

507 Fu Z, Shibata M, Makabe R, Ikeda H, Uye SI (2014) Body size reduction under starvation, and  
508 the point of no return, in ephyrae of the moon jellyfish *Aurelia aurita*. *Mar Ecol Prog Ser*  
509 510:255–263. doi: 10.3354/meps10799

510 Ginneken VJT van, Maes GE (2005) The European eel (*Anguilla anguilla*, Linnaeus), its  
511 lifecycle, evolution and reproduction: a literature review. *Rev Fish Biol Fish* 15:367–398.  
512 doi: 10.1007/s11160-006-0005-8

513 Havelange S, Lepoint G, Dauby P, J-M B (1997) Feeding of the Sparid Fish *Sarpa salpa* in a  
514 Seagrass Ecosystem : Diet and Carbon Flux. *Mar Ecol* 18:289–297. doi:  
515 <https://doi.org/10.1111/j.1439-0485.1997.tb00443.x>

516 Hays GC, Doyle TK, Houghton JDR (2018) A Paradigm Shift in the Trophic Importance of  
517 Jellyfish? *Trends Ecol Evol* 33:874–884. doi: 10.1016/j.tree.2018.09.001

518 Hirst AG, Lucas CH (1998) Salinity influences body weight quantification in the scyphomedusa  
519 *Aurelia aurita*: Important implications for body weight determination in gelatinous  
520 zooplankton. *Mar Ecol Prog Ser* 165:259–269. doi: 10.3354/meps165259

521 Hoving HJT, Haddock SHD (2017) The giant deep-sea octopus *Haliphron atlanticus* forages on  
522 gelatinous fauna. *Sci Rep* 7:44952:1–4. doi: 10.1038/srep44952

523 Hyslop EJ (1980) Stomach contents analysis-a review of methods and their application. *J Fish*

524 Biol 17:411–429. doi: <https://doi.org/10.1111/j.1095-8649.1980.tb02775.x>

525 Ishii H, Kojima S, Tanaka Y (2004) Survivorship and production of *Aurelia aurita* ephyrae in  
526 the innermost part of Tokyo Bay, Japan. *Plankt Biol Ecol* 51:26–35.

527 Laffaille P, Feunteun E, Lefebvre C, Radureau A, Sagan G, Laffaille P, Feunteun E, Lefebvre  
528 C, Radureau A, Sagan G (2002) Can thin-lipped mullet directly exploit the primary and  
529 detritic production of European macrotidal salt marshes? *Estuar Coast Shelf Sci* 54:729–  
530 736. doi: 10.1006/ecss.2001.0855

531 Lamb PD, Hunter E, Pinnegar JK, Creer S, Davies RG, Taylor MI (2017) Jellyfish on the menu:  
532 mtDNA assay reveals scyphozoan predation in the Irish Sea. *R Soc Open Sci* 4:171421.  
533 doi: 10.1098/rsos.171421

534 Lucas CH (1994) Biochemical composition of *Aurelia aurita* in relation to age and sexual  
535 maturity. *J Exp Mar Bio Ecol* 183:179–192. doi: 10.1016/0022-0981(94)90086-8

536 Lucas CH, Graham WM, Widmer C (2012) Jellyfish Life Histories: Role of polyps in forming  
537 and maintaining Scyphomedusa populations, 1st edn. Elsevier Ltd.

538 Marques R, Cantou M, Soriano S, Molinero JC, Bonnet D (2015a) Mapping distribution and  
539 habitats of *Aurelia* sp. polyps in Thau lagoon, north-western Mediterranean sea (France).  
540 *Mar Biol* 162:1441–1449. doi: 10.1007/s00227-015-2680-2

541 Marques R, Albouy-Boyer S, Delpy F, Carré C, Le Floc'H É, Roques C, Molinero JC, Bonnet  
542 D (2015b) Pelagic population dynamics of *Aurelia* sp. in French Mediterranean lagoons. *J*  
543 *Plankton Res* 37:1019–1035. doi: 10.1093/plankt/fbv059

544 Marques R, Bouvier C, Darnaude AM, Molinero J-C, Przybyla C, Soriano S, Tomasini J-A,  
545 Bonnet D (2016) Jellyfish as an alternative source of food for opportunistic fishes. *J Exp*  
546 *Mar Bio Ecol*. doi: 10.1016/j.jembe.2016.08.008

547 Marques R, Darnaude AM, Schiariti A, Tremblay Y, Carlos J, Soriano S, Hatey E, Colantoni S,  
548 Bonnet D (2019) Dynamics and asexual reproduction of the jellyfish *Aurelia coerulea*  
549 benthic life stage in the Thau lagoon (Northwestern Mediterranean ). *Mar Biol* 166:74:1–  
550 14. doi: 10.1007/s00227-019-3522-4

551 Matejusová I, Doig F, Middlemas SJ, Mackay S, Douglas A, Armstrong JD, Cunningham CO,  
552 Snow M (2008) Using quantitative real-time PCR to detect salmonid prey in scats of grey  
553 *Halichoerus grypus* and harbour *Phoca vitulina* seals in Scotland – an experimental and  
554 field study. *J Appl Ecol* 45:632–640. doi: 10.1111/j.1365-2664.2007.0

555 McInnes JC, Alderman R, Lea MA, Raymond B, Deagle BE, Phillips RA, Stanworth A,  
556 Thompson DR, Catry P, Weimerskirch H, Suazo CG, Gras M, Jarman SN (2017) High  
557 occurrence of jellyfish predation by black-browed and Campbell albatross identified by  
558 DNA metabarcoding. *Mol Ecol* 26:4831–4845. doi: 10.1111/mec.14245

559 Melià P, Bevacqua D, Crivelli A, Leo G De, Panfili J, Gatto M (2006) Age and growth of  
560 *Anguilla anguilla* in the Camargue lagoons. *J Fish Biol* 68:876–890. doi: 10.1111/j.1095-

561 8649.2006.00975.x

562 Mianzan HW, Mari N, Prenski B, Sanchez F (1996) Fish predation on neritic ctenophores from  
563 the Argentine continental shelf: A neglected food resource? *Fish Res* 27:69–79. doi:  
564 10.1016/0165-7836(95)00459-9

565 Milisenda G, Rosa S, Fuentes VL, Boero F, Guglielmo L, Purcell JE, Piraino S (2014) Jellyfish  
566 as prey: Frequency of predation and selective foraging of *Boops boops* (vertebrata,  
567 actinopterygii) on the mauve stinger *Pelagia noctiluca* (cnidaria, scyphozoa). *PLoS One*  
568 9:1–10. doi: 10.1371/journal.pone.0094600

569 Mills C (2001) Are population increasing globally in response to changing ocean conditions?  
570 *Hydrobiologia* 451:55–68. doi: <https://doi.org/10.1023/A:1011888006302>

571 Nejstgaard JC, Frischer ME, Simonelli P, Troedsson C, Brakel M, Adiyaman F, Sazhin AF,  
572 Artigas LF (2008) Quantitative PCR to estimate copepod feeding. *Mar Biol* 153:565–577.  
573 doi: 10.1007/s00227-007-0830-x

574 Owen SF, Houlihan DF, Rennie MJ, Weerd JH Van (1998) Bioenergetics and nitrogen balance  
575 of the European eel (*Anguilla anguilla*) fed at high and low ration levels. *Can J Fish Aquat*  
576 *Sci* 55:2365–2375. doi: <https://doi.org/10.1139/f98-119>

577 Phillips N, Eagling L, Harrod C, Reid N, Cappanera V, Houghton J (2017) Quacks snack on  
578 smacks : mallard ducks (*Anas platyrhynchos*) observed feeding on hydrozoans (*Velella*  
579 *velella*). *Plankt Benthos Res* 12:143–144. doi: <https://doi.org/10.3800/pbr.12.143>

580 Pita BC, Gamito S, Erzini K (2002) Feeding habits of the gilthead seabream (*Sparus aurata*)  
581 from the Ria Formosa (southern Portugal) as compared to the black seabream  
582 (*Spondyliosoma cantharus*) and the annular seabream (*Diplodus annularis*). *J Appl*  
583 *Ichthyol* 18:81–86. doi: <https://doi.org/10.1046/j.1439-0426.2002.00336.x>

584 Proman JM, Reynolds JD (2000) Differences in head shape of the European eel , *Anguilla*  
585 *anguilla* (L.). *Fish Manag Ecol* 7:349–355. doi: [https://doi.org/10.1046/j.1365-](https://doi.org/10.1046/j.1365-2400.2000.007004349.x)  
586 [2400.2000.007004349.x](https://doi.org/10.1046/j.1365-2400.2000.007004349.x)

587 Purcell JE (2012) Jellyfish and Ctenophore Blooms Coincide with Human Proliferations and  
588 Environmental Perturbations. *Ann Rev Mar Sci* 4:209–235. doi: 10.1146/annurev-marine-  
589 120709-142751

590 Purcell JE, Arai MN (2001) Interactions of pelagic cnidarians and ctenophores with fish: a  
591 review. *Hydrobiologia* 451:27–44. doi: 10.1023/A

592 Ramšak A, Stopar K, Malej A (2012) Comparative phylogeography of meroplanktonic species,  
593 *Aurelia* spp. and *Rhizostoma pulmo* (Cnidaria: Scyphozoa) in European Seas.  
594 *Hydrobiologia* 690:69–80. doi: 10.1007/s10750-012-1053-9

595 Riemann L, Alfredsson H, Hansen M, Als TD, Nielsen TG, Munk P, Aarestrup K, Maes GE,  
596 Sparholt H, Petersen MI, Bachler M, Castonguay M (2010) Qualitative assessment of the  
597 diet of European eel larvae in the Sargasso Sea resolved by DNA barcoding. *Biol Lett*

598 6:819–822. doi: <https://doi.org/10.1098/rsbl.2010.0411>

599 Roux JP, Van Der Lingen CD, Gibbons MJ, Moroff NE, Shannon LJ, Smith ADM, Cury PM  
600 (2013) Jellyfication of marine ecosystems as a likely consequence of overfishing small  
601 pelagic fishes: Lessons from the Benguela. *Bull Mar Sci* 89:249–284. doi:  
602 10.5343/bms.2011.1145

603 Russo T, Costa C, Cataudella S (2007) Correspondence between shape and feeding habit  
604 changes throughout ontogeny of gilthead sea bream *Sparus aurata* L., 1758. *J Fish Biol*  
605 71:629–656. doi: 10.1111/j.1095-8649.2007.01528.x

606 Scorrano S, Aglieri G, Boero F, Dawson MN, Piraino S (2016) Unmasking *Aurelia* species in  
607 the Mediterranean Sea: An integrative morphometric and molecular approach. *Zool J Linn*  
608 *Soc* 1:25. doi: 10.1111/zoj.12494

609 Stopar K, Ramšak A, Trontelj P, Malej A (2010) Lack of genetic structure in the jellyfish  
610 *Pelagia noctiluca* (Cnidaria: Scyphozoa: Semaestomeae) across European seas. *Mol*  
611 *Phylogenet Evol* 57:417–428. doi: 10.1016/j.ympev.2010.07.004

612 Sweetman AK, Smith CR, Dale T, Jones DOB (2014) Rapid scavenging of jellyfish carcasses  
613 reveals the importance of gelatinous material to deep-sea food webs. *Proc R Soc B Biol*  
614 *Sci* 281:20142210–20142210. doi: 10.1098/rspb.2014.2210

615 Takao M, Okawachi H, Uye S (2014) Natural predators of polyps of *Aurelia aurita* s.l.  
616 (Cnidaria: Scyphozoa: Semaestomeae) and their predation rates. *Plankt Benthos Res*  
617 9:105–113. doi: 10.3800/pbr.9.105

618 Tancioni L, Mariani S, Maccaroni A, Mariani A (2003) Locality-specific variation in the  
619 feeding of *Sparus aurata* L.: evidence from two Mediterranean lagoon systems. 57:469–  
620 474. doi: 10.1016/S0272-7714(02)00376-1

621 Tesch F, Bartsch P, Berg R, Gabriel O, Henderson IW, Kamstra A, Kloppmann M, Reimer LW,  
622 Wirth T (2003) *The Eel*, Third edit. Blackwell Science Ltd

623 Thiebot JB, Arnould JPY, Gómez-Laich A, Ito K, Kato A, Mattern T, Mitamura H, Noda T,  
624 Poupart T, Quintana F, Raclot T, Ropert-Coudert Y, Sala JE, Seddon PJ, Sutton GJ, Yoda  
625 K, Takahashi A (2017) Jellyfish and other gelata as food for four penguin species –  
626 insights from predator-borne videos. *Front Ecol Environ* 15:437–441. doi:  
627 10.1002/fee.1529

628 Töbe K, Meyer B, Fuentes V (2010) Detection of zooplankton items in the stomach and gut  
629 content of larval krill, *Euphausia superba*, using a molecular approach. *Polar Biol*  
630 33:407–414. doi: 10.1007/s00300-009-0714-2

631 Vizzini S, Mazzola A (2005) Feeding ecology of the sand smelt *Atherina boyeri* (Risso 1810)  
632 (Osteichthyes, Atherinidae) in the western Mediterranean: evidence for spatial variability  
633 based on stable carbon and nitrogen isotopes. *Environ Biol Fishes* 72:259–266. doi:  
634 <https://doi.org/10.1007/s10641-004-2586-1>

635 Wang J, Zhen Y, Wang G, M I T, Y U Z (2013) Molecular identification and detection of moon  
636 jellyfish (*Aurelia* sp. ) based on partial sequencing of mitochondrial 16S rDNA and COI.  
637 Chinese J Appl Ecol 24:847–852.  
638