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## Developing a sclerochronology network in the Adriatic Sea: Growth synchrony among populations of *Callista chione*

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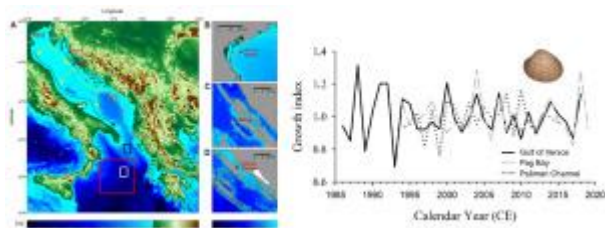
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### Abstract :

*Callista chione* samples were live collected at three localities in the Adriatic Sea: Gulf of Venice, Italy, Pag Bay and Pašman Channel, Croatia. Acetate peel replicas were prepared and the Image Pro Primer program was used to measure increment widths in samples that had clear boundaries. Visual cross-dating of the images was performed using list-year method and validated using COFECHA, while the R package *dplR* was used to construct the chronologies. The age of analysed shells ranged from 15 to 46 years (N=32;  $29,9 \pm 8,8$  years) at the Gulf of Venice, from 14 to 41 years (N=63;  $26,0 \pm 5,1$  years) at Pag Bay, and from 14 to 41 years (N=33;  $26,8 \pm 4,8$  years) at the Pašman Channel. Final statistically robust standard master chronologies spanned from 1986 to 2018 (Gulf of Venice), from 1994 to 2019 (Pag Bay) and from 1994 to 2015 (Pašman Channel). Statistically significant correlation was obtained only between the Gulf of Venice and Pag Bay residual master chronologies (1994–2018;  $r = 0.607$ ;  $p < 0.001$ ). Although there are some coincidences between all three localities, suggesting a common signal among populations, the growth chronology in the Pašman Channel recorded a very different signal potentially linked to differing oceanographic influences. The Pašman Channel locality is more affected by the inflow of warmer and saline waters coming from the south than the two other sites which are located in geographically and hydrographically isolated Adriatic areas. No correlation was found between any shell master chronologies and surface seawater temperature. Also, no correlations between the shell *C. chione* master chronologies and parameters describing the Adriatic-Ionian Bimodal Oscillating System (BiOS) were observed. Correlation maps indicate different relationships between shell master chronology and chlorophyll a concentration in the region. Significant correlations were mostly limited to March and April and showed spatial variations.

## Graphical abstract



**Keywords** : Mediterranean Sea, dpIR, Veneridae, shell growth, clams, sclerochronology

## 1. Introduction

Coastal and shelf areas are dynamic and complex multifunctional systems at the interface between land and sea (Salgado-Hernanz *et al.*, 2022). These areas are subject to a variety of stressors that overlap, including the effects of climate change, land-based pollution, and commercial fisheries (Ramírez *et al.*, 2018). Semi-enclosed seas, such as the Mediterranean Sea, are particularly vulnerable to disturbance due to high surface-to-volume ratios and strong pressures from various human activities (Halpern *et al.*, 2015; Schroede *et al.*, 2015; Piroddi *et al.*, 2017). Anthropogenic pressures on Mediterranean marine ecosystems are predicted to increase in the future (Coll *et al.*, 2010), particularly due to habitat destruction, resource exploitation, and climate change (Giorgi and Lionello, 2008; Coll *et al.*, 2012; Adloff *et al.*, 2015).

To understand the significance of potential future changes, knowledge of present and past environmental variability and its influence on marine organisms is critical. This requires detailed records of oceanic conditions over broad temporal and spatial scales (Grebmeier, 2012). As observational multidecadal or multicentennial records are often lacking, scientists have been exploring possibilities for reconstructing environmental conditions from growth increments in hard tissues of various marine organisms including bivalve shells, fish otoliths, coralline algae and corals (Gröcke and Gilikin, 2008; Oschmann, 2009; Butler *et al.*, 2019). Bivalves are often used in sclerochronological research because they continuously deposit shell material throughout their lifetimes and populations may exhibit a common response to environmental forcing, exhibiting synchronous shell growth rates (e.g., Wanamaker *et al.*, 2012; DeLong *et al.*, 2014; Reynolds *et al.*, 2016). Measuring of growth increment widths enables the development of absolutely dated, annually resolved, and environmentally sensitive proxy records ranging from several decades to a few millennia (e.g., Schöne, 2008; Black *et al.*, 2009; Butler *et al.*, 2013). By applying statistical methods adopted from the dendrochronological research, individual shell growth series data are used for constructing robust master chronologies (e.g., Wanamaker *et al.*, 2012; Butler *et al.*, 2013; Reynolds *et al.*, 2022). These chronologies can be related to modern environmental conditions and used for the reconstruction of past climate changes (e.g. Black *et al.*, 2009; Butler *et al.* 2010; Edge *et al.*, 2021). Further, efforts have been devoted to the construction of shell chronology networks, based on data from multiple sites within a certain geographic region, in order to obtain more comprehensive insight into past environmental variability on a larger scale (e.g. Butler *et al.*, 2009; Reynolds *et al.*, 2017; Peharda *et al.*, 2019).

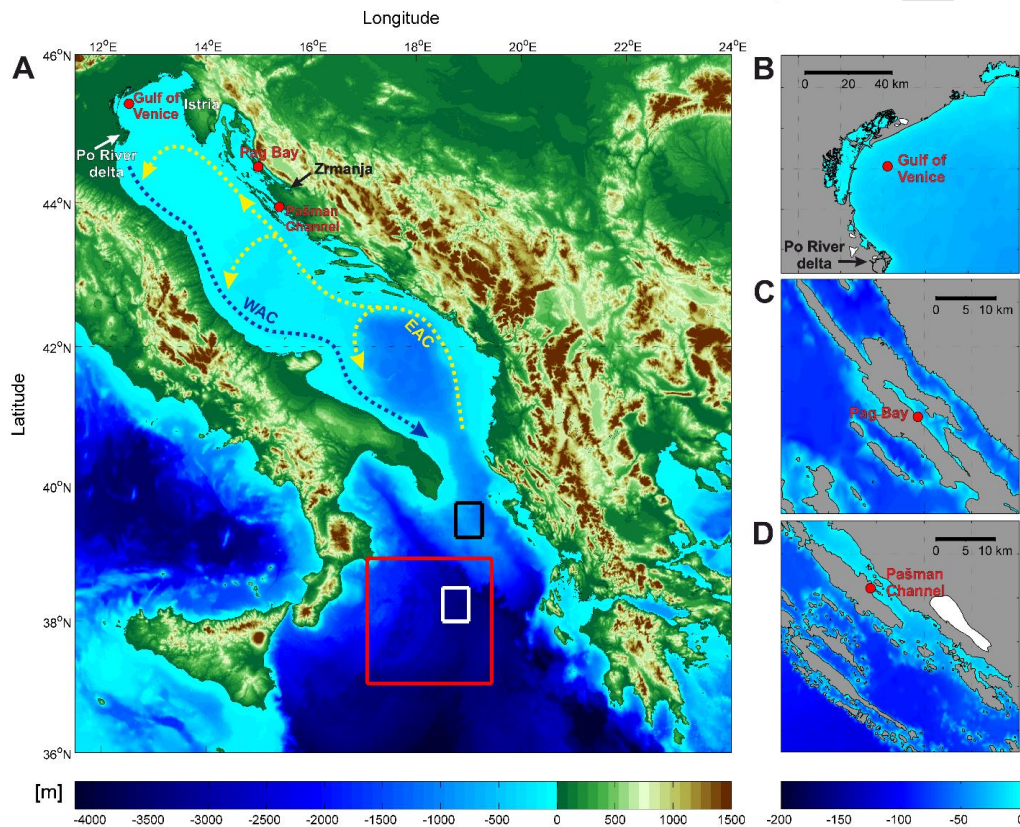
73 While the majority of bivalve chronologies have been developed at relatively high latitudes  
74 and in major ocean basins, such studies are still very rare in the Mediterranean and limited to  
75 the Adriatic Sea and to species from the genus *Glycymeris*, including *G. bimaculata* (Bušelić  
76 *et al.*, 2015) and *G. pilosa* (Peharda *et al.*, 2016; Peharda *et al.*, 2018; Peharda *et al.*, 2019).  
77 Previous studies on age and growth of the commercially important venerid bivalve *Callista*  
78 *chione* have shown that the longevity of this species can extend up to four decades (Forster,  
79 1981; Ezgeta-Balić *et al.*, 2011), making it an interesting target species for sclerochronological  
80 studies (Purroy *et al.*, 2018a). *Callista chione* is a relatively large (up to 10 cm), shallow-  
81 burrowing suspension-feeding bivalve that inhabits sandy sediments in coastal waters at depths  
82 up to 180 m and is widely distributed in the eastern Atlantic Ocean and the Mediterranean Sea,  
83 including the Adriatic Sea (Pope and Goto, 2000). It is commercially exploited in several  
84 countries from the eastern Mediterranean to the Atlantic, including Croatia, France, Greece,  
85 Italy, Morocco, Portugal, and Spain (e.g., Gaspar *et al.*, 2001, 2002; Tirado *et al.*, 2002;  
86 Metaxatos, 2004; Moura *et al.*, 2009; Ezgeta-Balić *et al.*, 2011; Baeta *et al.*, 2014; Bouzaidi *et al.*,  
87 2020). Previous studies conducted on *C. chione* include analyses of the reproductive cycle  
88 (Valli *et al.*, 1984; Tirado *et al.*, 2002; Moura *et al.*, 2008; Purroy *et al.*, 2019), population  
89 dynamics (Metaxatos, 2004), age and shell growth rate (Hall *et al.*, 1974; Forster, 1981; Keller  
90 *et al.*, 2002; Leontarakis and Richardson 2005; Moura *et al.*, 2009; Ezgeta-Balić *et al.*, 2011,  
91 Purroy *et al.*, 2018b). Understanding growth synchrony within and among populations in the  
92 species *C. chione* is needed in order to obtain an insight into its growth dynamics in relation to  
93 environmental conditions.

94 The main objectives of this study were to: (1) determine if well-replicated, annually- resolved,  
95 shell growth chronologies can be developed from *C. chione* shells collected at three localities  
96 in the Adriatic Sea, (2) determine the potential of *C. chione* for developing a network of bivalve  
97 chronologies in the Adriatic Sea, and (3) test possible environmental influences on the shell  
98 growth of this species.

## 99 2. Materials and methods

100 Individuals of *Callista chione* were collected live at three localities in the Adriatic Sea:  
101 (1) the Gulf of Venice, Italy (45.344° N, 12.525° E; 17-18 m depth); (2) the Pag Bay, Croatia  
102 (44.495° N, 14.987° E; 4-6 m depth); and (3) the Pašman Channel, Croatia (43.948° N, 15.388°  
103 E; 1.5-3 m depth) (Fig. 1). Specimens were obtained from the catch of the commercial fishing  
104 vessels at the locality in the Gulf of Venice in 2018, and by SCUBA and skin-diving at the  
105

106 other two localities on several occasions from 2013 to 2020 (Supplementary Material; Table  
 107 S1). For the laboratory analysis, we selected the largest-sized shells (>60 mm).



108

109 **Figure 1.** The study area: **A.** The bathymetry of the Northern Ionian Sea and the Adriatic Sea.  
 110 The localities of *Callista chione* sampling in the Adriatic are denoted by red dots. Schematic  
 111 representation of the Adriatic circulation is also depicted (the yellow line represents the Eastern  
 112 Adriatic Current – EAC, while the blue line represents the Western Adriatic Current – WAC).  
 113 The river mouth of the Zrmanja River (Croatia) is indicated by black arrow, while the delta of  
 114 the Po River (Italy) is denoted by white arrow. Black and white rectangles in the Northern  
 115 Ionian denote the areas between which the height difference of the absolute dynamic  
 116 topography (ADT) was calculated. The red rectangle in the Northern Ionian represents the area  
 117 used for computing vorticity. **B.** Gulf of Venice (the Po River delta is indicated by black  
 118 arrow), **C.** Pag Bay and **D.** Pašman Channel.

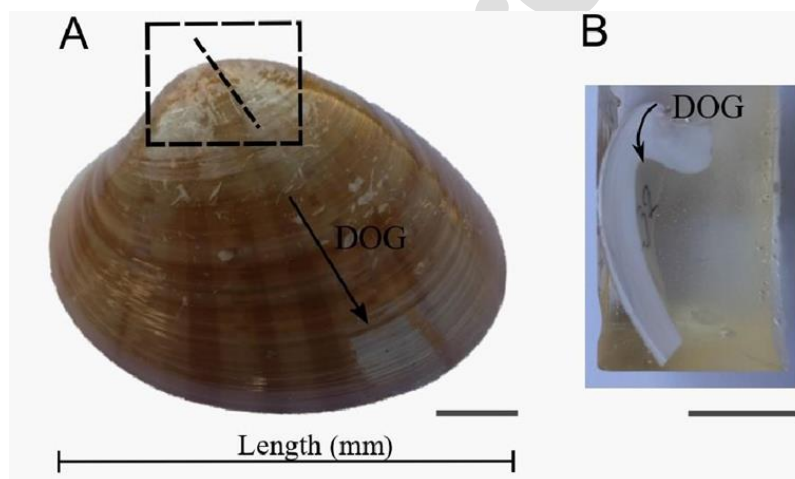
119 Immediately after collection, the specimens were frozen, and in the laboratory the  
 120 shells were thawed and the tissue removed. The shell length (anterior-posterior axis) of each

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121 specimen was measured with a digital caliper to the nearest 0.1 mm, and the dry weight was  
122 determined using a scientific scale with a precision of 0.01 g.

123 The hinge area of each shell was cut and embedded in epoxy resin. The resin blocks  
124 containing the hinge were cut along the axis of maximum growth, ground, and polished. The  
125 polished surfaces were etched in 0.1 M HCl for 2 min. After etching, samples were rinsed in  
126 tap water and left to air dry. Acetate peels were prepared by pouring ethyl acetate with a pipette  
127 on the polished and etched shell section and covering the surface with a piece of acetate sheet.  
128 Acetate peels were placed between two microscope glass slides and photographed using an  
129 Axio Lab A1 microscope equipped with a Zeiss AxioCam ERc 5s camera. Multiple  
130 photographs were taken for each sample and then stitched together into a single composite  
131 photograph using Image-Pro Plus 10 software. Due to the better visibility of annual growth  
132 lines, all measurements were done in the inner shell layer and were conducted from the  
133 ontogenetically youngest part of the shell towards the oldest part (Fig. S1).



134  
135 **Figure 2.** A. *Callista chione* shell, black rectangle represents hinge area that was cut from each  
136 sample, while dashed black line presents axis of maximum growth along which the shells were  
137 sectioned. B. Shell of *C. chione* embedded in epoxy resin. DOG – direction of growth. Scale  
138 bar 1 cm.

139 At each locality the age was estimated for all specimens that had clearly visible growth  
140 lines, but only shells that had clearly delineated growth increments boundaries were used to  
141 construct master chronologies. Counting of growth lines was performed independently by two  
142 experienced researchers and age estimates are reported only for shells for which estimates

143 matched. Acetate peel images were visually cross-dated using the list-year method  
144 (Yamaguchi, 1991) to ensure that each increment was assigned to the correct year of formation.  
145 This technique is based on the assumption that some aspects of the environment limit growth,  
146 and as **the environmental conditions** vary over time, they induce a synchronous growth pattern  
147 in contemporaneous individuals sampled from the same area (Fritts, 1976; Yamaguchi, 1991).  
148 The correct calendar year of each increment was assigned by cross-dating backward from the  
149 known year of sampling. Cross-dating was also checked using the program COFECHA  
150 (Holmes, 1983). It is important to note that analysis of stable oxygen isotope composition  
151 ( $\delta^{18}O$ ) in shells of *C. chione* specimens collected from Adriatic Sea showed that this species  
152 deposits an annual growth line during late summer/early autumn (Purroy *et al.*, 2018b;  
153 Uvanović, 2022) while the growth stop/cessation occurs during January and February. This  
154 means that a given annual growth increment can contain environmental data from two calendar  
155 years. For example, the annual growth increment **marked as “2015”** in our time series contains  
156 shell material deposited over two time periods: from September to December 2014, and from  
157 March to August 2015. **This was taken into account when analysing the potential correlations**  
158 **with environmental data.**

159 After visual cross-dating, individual growth increment widths were measured from the  
160 internal shell surface **towards the** external surface (Figure 2B) along the axis of maximum  
161 growth and perpendicular to **the** growth increment boundaries using **the** Image-Pro Plus 10  
162 software (Supplementary Material; Figure S1). All detrending and chronology construction  
163 were performed in R (R Core Team, 2022) using dplR package (Bunn, 2008). Detrending was  
164 performed with a cubic spline with a rigidity of 10 years and a cut-off frequency of 50%.  
165 Population-level signal strength in the chronology was assessed using the Expressed  
166 Population Signal (EPS). Although arbitrary, an EPS  $\sim 0.85$  is considered the threshold at which  
167 the sample set adequately reflects the theoretical population from which it was drawn (Wigley  
168 *et al.*, 1984). EPS was calculated over a window of 5 years. For each locality, **two versions of**  
169 **the** chronologies were built: **a standard and a residual version**. To build **the** residual chronology,  
170 each individual growth time series was first prewhitened by fitting an autoregressive model, in  
171 order to remove autocorrelation. Then all prewhitened time series were averaged using the  
172 Tukey’s biweight robust mean, in order to minimize the effect of outliers.

173 In order to test possible environmental influences on the growth of *C. chione*, available  
174 data of surface **seawater** temperature (**SST**) and chlorophyll-a concentrations were **correlated**  
175 with the shell growth chronologies. Seawater temperature can affect growth directly, but also



176 indirectly - through its influence on bivalve reproductive cycle and food supply (Schöne *et al.*,  
177 2005). Bivalve shell growth is known to be strongly influenced by food quantity and quality  
178 (Ballesta - Artero *et al.*, 2018). In order to highlight possible relationships between *C.chione*  
179 master chronologies and environmental data, correlation maps (Pearson's  $r$ ) were computed  
180 over a large area (36°-46°N 12°-22°E) around our three localities. The sea surface temperature  
181 data used to compute correlation maps were retrieved from the NOAA Optimum Interpolation  
182 Sea Surface Temperature (OI SST V2) dataset, provided by the NOAA PSL, Boulder,  
183 Colorado, USA (<https://psl.noaa.gov>). An estimation of the monthly concentration of  
184 chlorophyll-a (Chl- a) in the Adriatic, Ionian and Tyrrhenian Seas was obtained for the period  
185 1998-2019 from GlobColour (<http://globcolour.info>). High-resolution (1/24°, i.e. approx. 4 km  
186 × 4 km) ocean colour data retrieved from different sensors (SeaWiFS, MERIS, MODIS Aqua,  
187 and VIIRS) were averaged using the Garver-Siegel-Maritorena (GSM) model (Maritorena and  
188 Siegel, 2005). All files were downloaded in NetCDF4 format. For each cell of the gridded  
189 environmental dataset, pairwise Pearson's correlations were computed between the annual  
190 values of the master chronology and environmental data for the months of March, April, May,  
191 June, July and August. Only correlations at a 90% significance level (or higher) are displayed  
192 on these maps. NetCDF4 files were processed using R 4.2.0 "Vigorous Calisthenics" (R Core  
193 Team, 2022), after loading of packages 'rgdal', 'raster', and 'ncdf4'. **Correlations between the  
194 chronologies and environmental time series developed for the sampled locations were  
195 performed in order to detect the environmental force at each site. All additional information  
196 can be find in Supplementary material.**

197 The regime of the Adriatic - Ionian Bimodal Oscillating System (BiOS, Gačić *et al.*,  
198 2010) indicates the origin of waters entering the Adriatic Sea. In order to estimate the state of  
199 the BiOS, the monthly values of the height difference of the absolute dynamic topography  
200 (ADT), here termed ADT\_difference, were computed for a 0.5°x0.5° area centered on the  
201 northern edge of the eddy in the northern Ionian Sea (black rectangle in Fig. 1A) in relation to  
202 an area of the same dimensions at the center of the eddy (white rectangle in Fig. 1A). Another  
203 parameter used to describe the state of the BiOS was vorticity, calculated in for the northern  
204 part of the Ionian Sea (37.0°N - 39.0°N; 17.0°E - 19.5°E; red rectangle in Fig. 1A), using the  
205 procedure described by Shabrang *et al.* (2016). When the value of the parameter  
206 ADT\_difference is positive, the level in the center of the vortex is lower than the level at the  
207 edge of the vortex, which indicates cyclonic (counter-clockwise) rotation of the vortex. In the  
208 mentioned periods, the vorticity mostly has a positive value. The positive value of the

ADT\_difference parameter indicates the inflow of ultra-oligotrophic and highly saline water from the eastern Mediterranean into the Adriatic, which is characterized by low nutrients and lower primary production (Civitarese *et al.*, 2010). When the value of the parameter ADT\_difference is negative, the level at the center of the vortex is higher than the level at the edge of the vortex, indicating an anticyclonic (clockwise) rotation. Then, the vorticity mostly has negative values. In anticyclonic periods, the inflow of less oligotrophic water with lower salinity goes from the western Mediterranean into the Adriatic prevails

### 3. Results

In the Gulf of Venice, it was possible to estimate the age of 34 out of 52 processed shells, 19 of which were used in chronology construction. In Pag Bay, a total of 85 shells were processed and it was possible to estimate age for 64 shells, 47 of which were used to construct the chronology. In the Pašman Channel it was possible to estimate the age of 33 out of 37 processed shells, 26 of which were used in chronology construction (Table 1).

Table 1. Overview of the *Callista chione* specimens analysed in this study.

		Gulf of Venice	Pag Bay	Pašman Channel
				224
Shells used for chronology construction	N	21	48	27
	L (mm)	66.5 - 85.5	62.2 - 81.3	69.3 - 87.3
	min - max ( $\bar{x} \pm sd$ )	(73.4 $\pm$ 3.3)	(73.4 $\pm$ 3.7)	(76.2 $\pm$ 4.7)
	Age (years)	18 - 46	18 - 41	21 - 41
	min - max ( $\bar{x} \pm sd$ )	(29.9 $\pm$ 8.8)	(26.3 $\pm$ 4.9)	(26.8 $\pm$ 4.8)
Other shells	N	13	16	6
	L (mm)	70.1 - 79.5	70.0 - 76.9	56.0 - 88.6
	min - max ( $\bar{x} \pm sd$ )	(72.9 $\pm$ 2.4)	(73.4 $\pm$ 2.1)	(76.2 $\pm$ 10.9)
	Age (years)	15 - 39	14 - 32	21 - 33
	min - max ( $\bar{x} \pm sd$ )	(27.4 $\pm$ 6.9)	(25.4 $\pm$ 5.3)	(22.2 $\pm$ 7.8)

Shells used in chronology were of similar size and age compared with those for which only age was estimated. An exception was the case of several younger shells (n=5; <16 years) that were omitted from chronology development. All shells older than 40 were included in our analysis. Pronounced variation was noted in the estimated age of similar sized shells; for example, shells with length from 70 to 80 mm had an estimated age from 20 to more than 40

238 years (Supplementary Material; Figure S2). The mean correlation between each detrended time  
 239 series and the average of others (series intercorrelation) for shells from Gulf of Venice was  
 240 0.542. The average mean sensitivity which expresses the year-to-year variability of the values  
 241 in a time series was 0.256. For shells from Pag Bay, the series intercorrelation was 0.579 and  
 242 mean sensitivity was 0.279, while for the Pašman Channel samples, the series intercorrelation  
 243 was 0.543 and mean sensitivity was 0.263.

244 During the first few years of life, shell growth is very rapid and often the annual growth  
 245 lines are not sufficiently clear, so it is not possible to accurately measure the distances between  
 246 them. Therefore, the widths of annual growth increments during the earlier ontogeny (~first 5  
 247 years) were not measured. Similarly, in samples collected in periods before late summer/early  
 248 autumn when the annual growth line is formed, last year at the very edge was not measured.  
 249 Also, due to very narrow growth increments, the positions of annual growth lines at the very  
 250 edge (late ontogeny) were not sufficiently clear in some specimens and in such cases were not  
 251 measured.

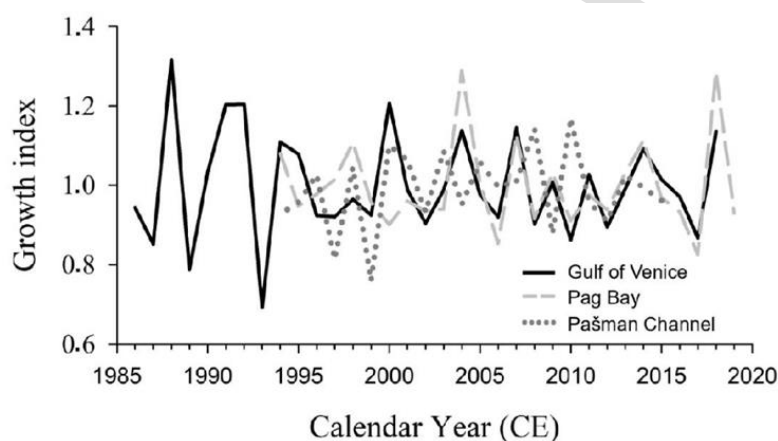
252 Data on the width of annual growth increments for *C. chione* specimens collected at the  
 253 three localities were obtained for the periods showed in Table 2.

254 Table 2. Overview of the periods of measured annual growth increments for *Callista chione*  
 255 specimens and constructed master chronologies per localities.

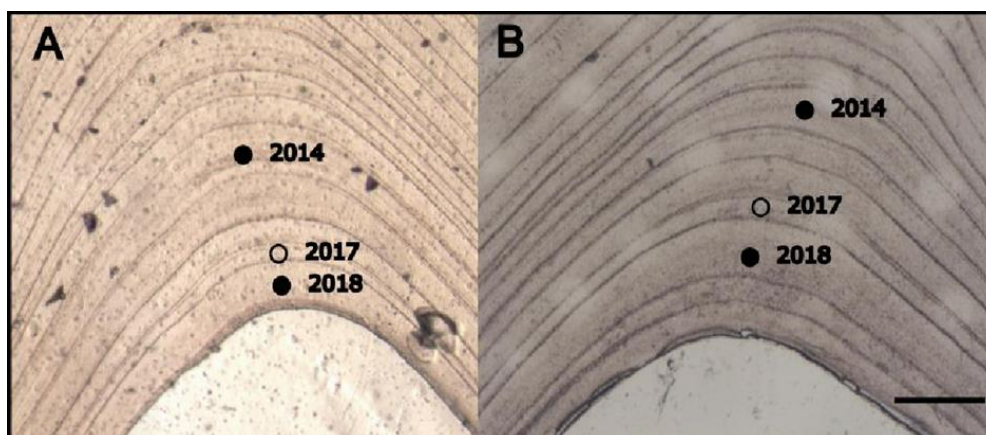
Locality	Measured annual growth increments	Master chronology period
Gulf of Venice	1997 - 2018	1986 - 2018
Pag Bay	1984 - 2019	1994 - 2019
Pašman Channel	1982 - 2015	1994 - 2015

257 **Figure 3.** Individual detrended growth time series of *Callista chione* from; **A.** The Gulf of  
 258 Venice (1979-2018), **B.** Pag Bay (1984-2019), and **C.** The Pašman Channel (1982-2015). Solid  
 259 black line represents standard chronology. The growth index varies around 1, with higher  
 260 values indicating positive wider and lower values indicating negative narrower growth than  
 261 expected by the model. Sample depth (denoting number of samples, grey shading area), 0.85  
 262 EPS threshold (straight dashed line) and calculated EPS (black line) are shown for each  
 263 sampling locality below corresponding chronology data.

264 Comparison of the three residual master chronologies obtained for *C. chione* individuals  
 1 265 collected from the three localities in the Adriatic Sea was made for the time periods over which  
 2 266 each chronology was determined to be suitable for environmental comparison based on  
 3 267 EPS  $\sim$  0.85. A statistically significant correlation was obtained only between the Gulf of  
 4 268 Venice and Pag Bay chronologies from 1994 to 2018 ( $r = 0.607$ ;  $p < 0.001$ ). The correlation  
 5 269 between these two localities is supported by the synchrony found in years 2004, 2007, 2014,  
 6 270 and 2018 that were characterized by higher growth indexes (wider increments). Likewise,  
 7 271 coincidences were also found in the years 2006, 2008, 2012, and 2017 that showed lower  
 8 272 growth indexes (narrower increments; Fig. 5). Interestingly, the years 2002, 2006, and 2012  
 9 273 were characterized by low growth for all three localities (Fig. 4).



274  
 275 **Figure 4.** Comparison of *Callista chione* residual shell master chronologies from the three  
 276 sampling localities. The growth index varies around 1, with higher values indicating wider  
 277 growth and lower values indicating narrower growth than expected by the model.

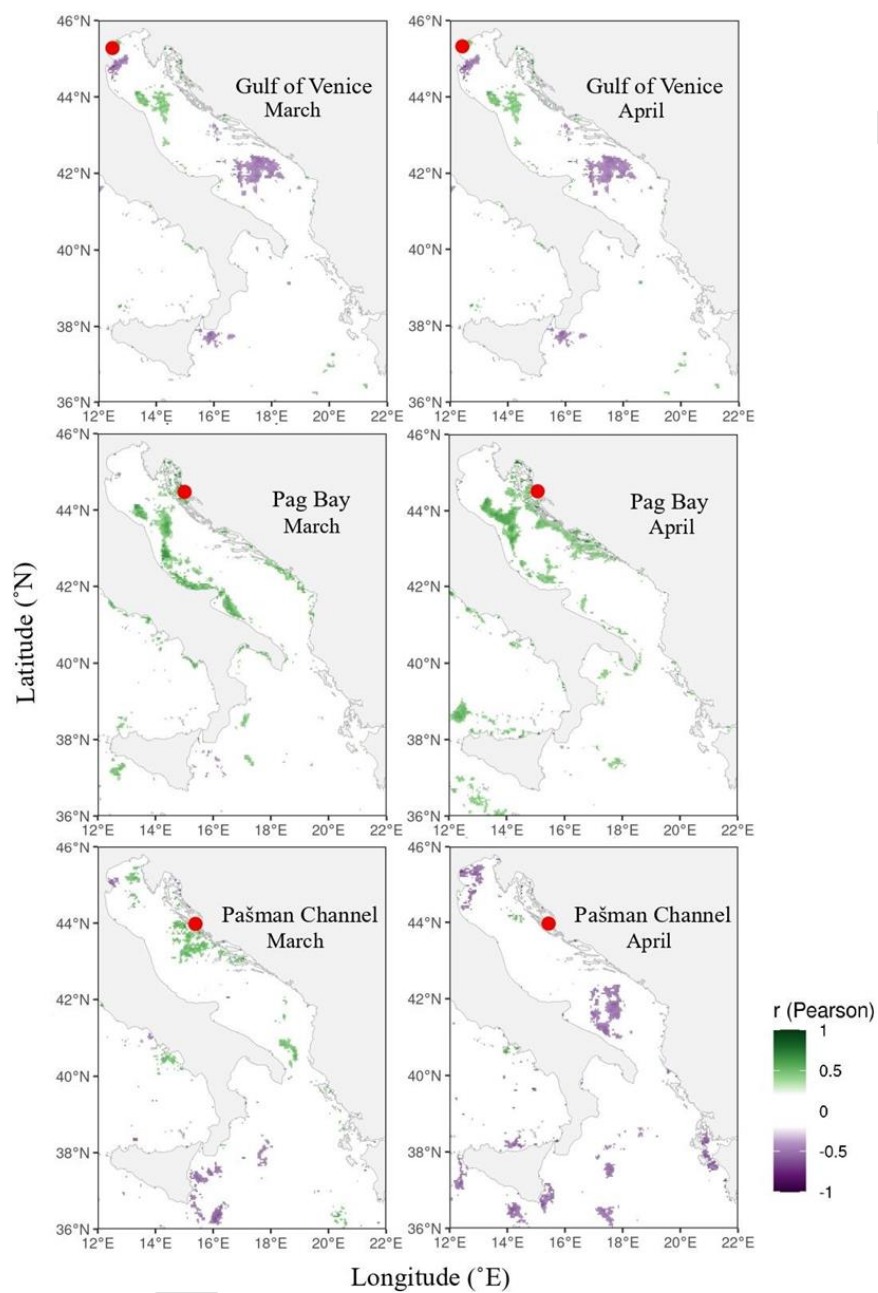


278

279 **Figure 5.** Acetate peels of shells collected in **A.** Gulf of Venice, (ITA2), and **B.** Pag Bay  
280 (PAG39). Full circles indicate wide growth increments, open circles indicate narrow growth  
281 increments. Scale bar 100  $\mu$ m.

282 No significant correlations were found between the Gulf of Venice shell growth  
283 chronology and chlorophyll *a* concentrations in the nearby area of the Northern Adriatic,  
284 however, some areas of positive and negative correlation were observed further south **of this**  
285 **locality**. Pearson's *r* correlations at a 90% significance level (or higher) were observed between  
286 the shell master chronologies and chlorophyll concentration for March and April. Positive  
287 correlations were observed between shell data for Pag Bay and chlorophyll values at nearby  
288 **sites** for the same months, while for shells from the Pašman Channel significant positive  
289 correlation **was** only **found** for March (Fig. 6). Comparisons between shell growth chronologies  
290 and sea surface temperatures (**not shown**) showed no significant correlations in any area of the  
291 Adriatic **for any of the three** master chronologies.

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293 **Figure 6.** Correlation maps between shell master chronology and chlorophyll concentration for

294 the Gulf of Venice (1999-2018), Pag Bay (2002-2019) and the Pašman Channel (1996-2015)

295 in March and April. Red circle represents locality of shell collection.

296



## 4. Discussion

### 4.1. *Callista chione* age estimation

Growth rate and age are **widely used** biological parameters for the management and conservation of living marine resources. The age of a bivalve can be estimated using surface growth rings or checks that can be clearly seen on the external shell surface (Richardson 2001). They are particularly clear in *C.chione* during early ontogeny, when the rings are widely spaced and easy to identify (Forster, 1981). However, later in ontogeny (>20 years) they are deposited more closely together at the shell margin and are not always easily discernible. Results of this study show that age of individuals of in this species can pronouncedly vary with respect to length, as estimated age of specimens measuring ~ 75 mm shell length ranged from 22 to 45 years (Supplementary Material; Figure S2). Slow growth during late ontogeny and high **intraspecies** variation in shell growth rates were previously noted for *C.chione* (Ezgeta-Balić *et al.*, 2011), as well as for a number of other bivalves species including for example *Arctica islandica* (e.g. Witbaard *et al.*, 1999, Schöne, 2013), and *Lithophaga lithophaga* (Peharda *et al.*, 2015).- Therefore, it is **more appropriate** to use acetate peel replicas examined under magnification **to estimate the age of larger sized *C.chione* individuals**, as opposed to size-age relationships, suggested by Ezgeta-Balić *et al.* (2011).

*Callista chione* is a moderately long-lived bivalve that can attain an age of >40 years (Forster, 1981; Ezgeta-Balić *et al.*, 2011). **However, according to previous studies it is more common to find populations with individuals showing a maximum age** of 16 years (Leontarakis and Richardson, 2005) or 17 years (Hall *et al.*, 1974; Metaxatos, 2004; Moura *et al.*, 2009) have been recorded in most of the studied populations. The population structure of *C.chione* in the Adriatic Sea has been documented by Ezgeta-Balić *et al.* (2011) at four localities along the eastern coast (Rab, Pag Bay, Kaštela Bay and the estuary of the Cetina River); individual ages ranged between 3 and 44 years, and contribution of older specimens in a given population varied pronouncedly with respect to locality. In this study, estimated age ranged from 14 to 46 years and 90% of analysed specimens were over 20 years old. It is important to note that for the purpose of chronology construction, larger individuals with >60 mm in length were chosen. Maximal longevity of 46 years estimated in **this study** is two years longer than previous estimates for specimens from Rab (Ezgeta-Balić *et al.* 2011). Due to **the** commercial importance of this species, it is highly likely that larger sized and older specimens have already been selectively removed from the Adriatic populations. Age data is a prerequisite to generate

330 the information on population structure, longevity, mortality, recruitment and fluctuations in  
331 fishery, which could contribute to the improvement of the national management plans of the  
332 species stock (Pauly *et al.*, 2002).

#### 333 4.2. *Callista chione* master chronologies

334 In the last decade, at least 25 species of the Veneridae family have been the subject of  
335 sclerochronological studies, with most species analysed in only one or two studies, indicating  
336 the ongoing quest for suitable target species as well as the expanding interest for  
337 sclerochronological studies (Peharda *et al.*, 2021). Nevertheless, there is a relatively small  
338 number of studies on venerid species that focus on the construction of master chronologies.  
339 One of the first such studies was conducted by Schöne (2003) on three short-lived species (<10  
340 years) collected in the Gulf of California, Mexico. Multidecadal master-chronologies of long-  
341 lived venerids *Mercenaria stimpsoni* (~100 years) were built by Tanabe *et al.* (2017) and Shirai  
342 *et al.* (2018), thanks to its long lifespan, clearly visible growth lines, and synchronized growth  
343 between individuals as well as clearly visible growth lines. It should be noted that the  
344 chronology published in the work of Shirai *et al.* (2018) was made on data for only three  
345 specimens. To the best of our knowledge, there are no other published studies related to the  
346 construction of master chronology for Veneridae species, and data from our study contribute  
347 to the understanding of growth in this bivalve family.

348 Previous research aimed at developing master chronologies of bivalves in the Adriatic  
349 Sea, but also in the Mediterranean Sea, has been limited to species of the genus *Glycymeris*  
350 that were targeted for their longevity. The genus *Glycymeris* has been interesting for  
351 sclerochronology research in other parts of the world, including research conducted on *G.*  
352 *glycymeris* from the Northeast Atlantic (Brocas *et al.*, 2013; Featherstone *et al.*, 2017; Reynolds  
353 *et al.*, 2017; Alexandrof *et al.*, 2021); *G. longior* from the Southwest Atlantic (Gimenez *et al.*,  
354 2020a; 2020b) and *G. vanhengstumi* from North Atlantic (Nemeth and Kern, 2018). In the  
355 Adriatic Sea, a 16 years long master chronology of the species *G. bimaculata* was constructed  
356 based on growth increment data obtained from shells collected in Pag Bay (Bušelić *et al.*,  
357 2015). Maximal estimated longevity of *G. bimaculata* in that study was 57 years. Given the  
358 relatively small number of *G. bimaculata* individuals that were older than 20 years, research  
359 focused on master chronology construction later on targeted the species *G. pilosa*. The length  
360 of the chronologies for this species ranged from 17 years in the Pašman Channel (Peharda *et al.*  
361 *et al.*, 2016) to 42 years at the locality Drače (Peharda *et al.*, 2019) and they were constructed  
362 using a larger number of specimens. Maximal estimated longevity of *G. pilosa* in the Adriatic

363 Sea was estimated at impressive 97 years (Peharda *et al.*, 2019). In our study, the lengths of  
1 364 the constructed master chronologies of *C.chione* were 32 years for the Gulf of Venice, 25 years  
2 365 for Pag Bay, and 21 years for the Pašman Channel.  
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6 366 Collecting growth data at multiple localities allows the development of networks of  
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8 367 chronologies and the analysis of spatial similarities and differences (Black, 2009; Butler *et al.*,  
9  
10 368 2009; Brocas *et al.*, 2013; Reynolds *et al.* 2017; Peharda *et al.*, 2019). So far, such research has  
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12 369 been conducted at several localities in the Pacific, Atlantic, and Mediterranean, and has  
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14 370 included species such as *Panopea generosa* (Edge *et al.*, 2021), *Arctica islandica* (Butler *et al.*,  
15  
16 371 2009), *G. glycymeris* (Brocas *et al.*, 2013) and *G. pilosa* (Peharda *et al.*, 2019). Research  
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18 372 conducted in open marine systems in the Pacific and Atlantic has shown that there are some  
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20 373 statistically significant correlations between master chronologies from different localities, as  
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22 374 well as correlations with environmental factors (Black *et al.*, 2009; Butler *et al.*, 2009; Brocas  
23  
24 375 *et al.*, 2013; Reynolds *et al.*, 2017). Most of these studies have analysed the spatial coherence  
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26 376 among populations from a single species, while a limited number of studies also focus on  
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28 377 networks that include multiple species. Such approach was conducted by Reynolds *et al.* (2017)  
29  
30 378 and included the species *A. islandica* and *G. glycymeris* collected from several localities along  
31  
32 379 the western British continental shelf, and by Reynolds *et al.* (2022) which analysed the species  
33  
34 380 *Astarte borealis* and *Liocyma fluctuosa* collected from the eastern Chukchi Sea in the Arctic  
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36 381 Ocean. In the eastern Adriatic, a comparison of chronologies of *G. pilosa* growth at several  
37  
38 382 localities revealed similarities between chronologies for samples collected along the western  
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40 383 coast of Istria and the Pašman Channel, and between chronologies at Živogošće and Drače  
41  
42 384 (Peharda *et al.*, 2019). In this study, similarities were found between the master chronologies  
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44 385 of *C.chione* at the Gulf of Venice and Pag Bay, while the chronology obtained for the Pašman  
45  
46 386 Channel did not correlate with the two northern chronologies. The sampled localities are in the  
47  
48 387 shallow coastal area of the Adriatic Sea and according to Peharda *et al.* (2019) the heterogeneity  
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50 388 in bivalve growth can potentially reflect heterogeneous climate regimes or the influence of  
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52 389 local limiting environmental factors on their growth.

53 390 One possible explanation as to why the Pašman Channel chronology differs from other  
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55 391 two sites could be related to different environmental conditions affecting the Gulf of Venice  
56  
57 392 and Pag Bay sites. Namely, the general Adriatic surface circulation is cyclonic (Orlić *et al.*,  
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59 393 1992), with a north-westward flow along the eastern coast (Eastern Adriatic Current – EAC,  
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61 394 Fig. 1A)  
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395 and a south-eastward current along the western coast (Western Adriatic Current – WAC, Fig.  
1A). The Pašman Channel sampling site is therefore more affected by the inflow of warmer  
396 and saline waters coming from the south than the two other sites, as Pag Bay and the Gulf of  
397 Venice sampling sites are located in somewhat geographically and hydrographically isolated  
398 Adriatic areas. Pag Bay is a part of the deeply indented north-eastern Adriatic coastline,  
399 strongly influenced by the severe wintertime outbreaks of cold and dry Bora wind (Grisogono  
400 and Belušić, 2009) and local fresh water sources (Zrmanja River and submarine fresh water  
401 springs, e.g., Novosel *et al.*, 2002; Šupraha *et al.*, 2011). The oceanographic properties of the  
402 northernmost part of the Adriatic are strongly dependent on the Po River discharges and  
403 characteristics of the air-sea fluxes, particularly those related to the strong Bora outbreaks,  
404 which excite the cyclonic gyres encompassing the area of the sampling site (Orlić *et al.*, 1994;  
405 Kuzmić *et al.*, 2006). Additionally, a persistent thermohaline front positioned to the south and  
406 southwest of the Istrian peninsula (Istrian Front), separating colder and less saline waters of  
407 the northernmost part of the northern Adriatic from the warmer and more saline southern  
408 waters, was reported during oceanographic surveys and in model simulations (e.g., Kokkini *et al.*,  
409 2017 and references therein).

#### 4.3. Environmental influence on the growth of *Callista chione*

Growth synchrony among *C. chione* specimens collected from the same locality suggests  
412 that a common environmental parameter influences individual growth. However, relationships  
413 between shell growth and external factors can be complex, especially in shallow coastal  
414 environments in semi-enclosed seas such as the Adriatic Sea (Epple *et al.*, 2006). In these areas,  
415 local processes, including precipitation, riverine input and terrestrial runoff, may be highly  
416 localised (Gillanders, 2005). Some previous sclerochronological studies have identified  
417 seawater temperature as one of the main drivers of shell growth. Positive correlations between  
418 seawater temperature and bivalve growth have been observed for several bivalve species  
419 including *Arctica islandica* (e.g., Marali and Schöne, 2015), *Glycymeris glycymeris* (Reynolds  
420 *et al.*, 2013; Royer *et al.*, 2013), and *Glycymeris bimaculata* (Bušelić *et al.*, 2015). Negative  
421 correlation between seawater temperature and bivalve growth have also been observed in the  
422 case of master chronologies of species *Ciliatocardium ciliatum* in the northwest Barents Sea  
423 (Carroll *et al.*, 2014) and species *A. islandica* in the southern Barents Sea (Mette *et al.*, 2016).  
424 Some of the studies found very low (e.g., Butler *et al.*, 2013) or time-unstable (e.g., Marali and  
425 Schöne, 2015) correlation between seawater temperature and shell growth thus further  
426 highlighting the complexity in growth-temperature relationships among and even within

428 **species**. In the case of *C.chione* master chronologies, no correlations were observed with  
1 429 seawater temperature in the Adriatic Sea.

430 Previous studies of *G. pilosa* in the Adriatic Sea have identified the impact of decadal (5  
431 - 10 years) oscillation of water masses referred to as the Adriatic - Ionian Bimodal Oscillating  
432 System (BiOS), on populations at the Barbariga and Pašman Channel localities (Peharda *et al.*,  
433 2019). BiOS is known to be the dominant driver of the thermohaline and biogeochemical  
434 oscillations across most of the basin, including southern and middle Adriatic and the shallow  
435 northern areas (Civitarese *et al.*, 2010; Vilibić *et al.*, 2012; Batistić *et al.*, 2014; Dautović *et*  
436 *al.*, 2017). **However, in the same study, at two southern localities Živogošće and Drače, growth**  
437 **of *G. pilosa* was influenced more by the local factors**. In this study, no correlations of *C. chione*  
438 master chronologies and parameters describing BiOS were observed.

439 **In this study, positive and negative correlations between shell growth chronologies and**  
440 **nearby chl a values were observed for the three different locations**. Although satellite  
441 chlorophyll a data provide insights into state of phytoplankton communities in an area, this  
442 does not monitor sudden changes of conditions in enclosed bays or straight channels between  
443 islands (Kirk, 2011). A previous study of trophic ecology of the *C.chione* conducted in Pag  
444 Bay and the Cetina River estuary showed that the diet differs between localities (Purroy *et al.*,  
445 2018a). According to Purroy *et al.* (2018a), for the bivalves in Pag Bay the most important  
446 food source are particles deposited on sediment, while in the estuary of the Cetina River the  
447 most important food source is suspended particulate matter. Correlation maps obtained in our  
448 study indicate different relationships between the shell master chronology and chlorophyll a  
449 concentration in the region. Significant correlations were mostly limited to March and April  
450 and showed spatial variations. **However, the environmental variables that influence bivalve**  
451 **growth can vary over fine spatial scales, especially in coastal environments in semi enclosed**  
452 **seas where local processes can be highly localized (Gillanders, 2005). While no direct influence**  
453 **of environmental data on the growth of bivalve *C. chione* were observed**, results in this study  
454 highlight the need to combine sclerochronological analyses with ecological studies **and in situ**  
455 **measurements of parameters such as food availability and quality** to understand life history  
456 traits of bivalves as archives of environmental variables.

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**Declaration of interests**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

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