

## Twenty-five-year longevity of European hake (*Merluccius merluccius*) from novel use of bomb radiocarbon dating in the Mediterranean Sea

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**Abstract.** The high variability of growth and longevity estimates for European hake (*Merluccius merluccius*) reflects the existence of two opposing hypotheses on growth rates that differ by a factor of 2: (1) a fast-growing hypothesis (FGH) with a maximum age near 15 years; and (2) a slow-growing hypothesis (SGH) with a maximum age near 30 years. A recently established regional radiocarbon (<sup>14</sup>C) reference led to a first-time application of bomb <sup>14</sup>C dating in the Mediterranean Sea to three of the largest-sized and potentially oldest-catch female European hake. Because age reading of otoliths is very subjective and poorly defined, these fish were aged blind with bomb radiocarbon (<sup>14</sup>C) dating as an independent estimate of validated age. The validated ages were compared with the theoretical maximum ages from the most reliable FGH and SGH von Bertalanffy growth functions. Among the three bomb <sup>14</sup>C ages, the most diagnostic length-at-age was an alignment with the bomb <sup>14</sup>C rise period for two of the three fish, providing validated ages of 22 years (74.5-cm total length) and 25 years (88-cm total length). The results provide estimates of length-at-age that are in agreement with the SGH and cannot be accounted for by the FGH.

**Additional keywords:** age validation, carbon-14, growth, lifespan, Merlucciidae.

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### Introduction

The potential longevity of commercial species is relevant to fisheries management because of its relationship to key parameters in stock assessment models, such as growth rates and estimates of mortality (Tyler *et al.* 1989). This is particularly true in the case of the European hake (*Merluccius merluccius* L., 1758) because it is the dominant commercial fish species in demersal fisheries, both in the north-eastern Atlantic and Mediterranean Sea (Alheit and Pitcher 2012). Age investigations for this species have focussed primarily on interpretation of larval, juvenile and adult fish otoliths, with little concern for the accuracy of longevity estimates. Uncertainties on the validity of current age estimation methods, which vary considerably in age reading protocol, are widely reported and controversial (e.g. Orsi-Relini *et al.* 1992; Morales-Nin *et al.* 1998; García-Rodríguez and Esteban 2002; de Pontual *et al.* 2006; Mellon-Duval *et al.* 2010).

Based on the use of contradictory age-reading approaches in estimating European hake growth (e.g. Ragonese *et al.* 2012), two opposing hypotheses are under consideration in the present study: (1) a fast-growing hypothesis (FGH), with a maximum

age near 15 years (e.g. García-Rodríguez and Esteban 2002; Mellon-Duval *et al.* 2010); and (2) a slow-growing hypothesis (SGH), with a maximum age near 30 years (e.g. Aldebert and Carries 1989; Levi *et al.* 1994). Thus, European hake longevity differs by a factor of 2 and is still a challenge for stock assessments that are regularly conducted by the General Fisheries Commission for the Mediterranean (GFCM).

A recently established regional radiocarbon (<sup>14</sup>C) reference in the Ligurian Sea from a 50-year-old coral led to an opportunity to apply bomb <sup>14</sup>C dating in the Mediterranean Sea. The aims of the present study were to: (1) independently estimate the age of three of the largest female European hake using bomb <sup>14</sup>C dating (e.g. Andrews *et al.* 2012); and (2) compare these validated age estimates with the maximum theoretical ages derived from contrasting von Bertalanffy growth functions for European hake of the Mediterranean Sea.

### Material and methods

The available otoliths of large female European hake caught off the South of Sicily (Geographical Sub-Area (GSA) 16) in the

**Table 1. List of specimen and radiocarbon data for the three European hake used in the present study**

Otolith core extraction mass with resultant  $\Delta^{14}\text{C}$  value provided a basis for validated birth years, ages and a determination of fast-growing hypothesis (FGH) v. the slow-growing hypothesis (SGH)  
TL, total length; TW, total weight

Specimen	EH-1	EH-2	EH-3
Fish length (TL; cm)	88.0	74.5	60.3
Fish weight (TW; g)	4805	3578	1376
Mean whole otolith weight (g)	1.042	0.705	0.482
Year of collection	1991	1985	1985
Radiocarbon			
Otolith mass extracted (mg)	8.9	8.7	8.7
Mean ( $\pm 2$ s.d.) $\Delta^{14}\text{C}$ (‰)	$59.9 \pm 4.0$	$45.0 \pm 4.0$	$66.3 \pm 4.0$
Validated birth year (range)	1966 (1968–1964)	1963 (1967–1965)	>1967 (>1967)
Age range from reference time series (years)	$25 \pm 2$	$22 \pm 2$	<18
Validated hypothesis	SGH	SGH	FGH/SGH

**Table 2. Von Bertalanffy growth function parameters that lead to expected longevity estimates that differ by a factor of 2**

$L_\infty$ , asymptotic length;  $t_0$ , the age at which the fish has zero length;  $k$ , growth coefficient;  $t_{\max}$ , longevity

Growth scenario	$L_\infty$	$k$	$t_0$	$t_{\max}$	Area	Reference
Fast	100.7	0.24	0	13	Gulf of Lyons	Mellon-Duval et al. (2010)
Fast	108.0	0.21	0.11	14	Spanish coast	García-Rodríguez and Esteban (2002)
Slow	80.2	0.11	-0.52	27	Gulf of Lyons	Aldebert and Carries (1989)
Slow	80.8	0.09	-1.74	32	Strait of Sicily	Levi et al. (1994)

late 1980s–90s were chosen from the archive of the Institute for Marine and Coastal Environment unit of Mazara del Vallo, Italy (IAMC-CNR; Table 1). Because age reading of otoliths is very subjective and poorly defined, bomb  $^{14}\text{C}$  dating was used as an independent estimate of age. The effectiveness of the bomb  $^{14}\text{C}$  assays was based on possible birth years of either 1958–1966 (SGH) or 1967–1983 (FGH). This determination would be diagnostic in terms of validated age because of the well-differentiated  $^{14}\text{C}$  levels defined by a regional coral reference and would be independent of age-reading protocol.

Radiocarbon analyses were performed by Beta Analytic Radiocarbon Dating ([www.radiocarbon.com](http://www.radiocarbon.com), accessed 21 April 2016) on ~9 mg samples of otolith core material. The extractions were made by hand grinding the external layers away from the core of the otolith and likely represented a few months of growth based on 1-year-old otoliths that weighed ~35 mg. Core extraction was guided by observations of a well-defined translucent band that is formed at settlement from the pelagic phase (Morales-Nin et al. 1998). In addition, the concentric nature of the earliest layers provided a well-centred extraction. Otolith calcium carbonate was processed for  $^{14}\text{C}$  content using standard accelerator mass spectrometry procedures. The measured  $^{14}\text{C}$  values, reported as  $\Delta^{14}\text{C}$  based on a deviation from a pre-nuclear standard (Stuiver and Polach 1977), were used to calculate an approximate year-of-formation (birth year) from an alignment with a regional  $\Delta^{14}\text{C}$  reference series. Calculated fish age was based on the difference from collection year to the  $\Delta^{14}\text{C}$  reference alignment. The  $\Delta^{14}\text{C}$  reference was a recently validated time series from a 50-year-old coral (*Cladocora caespitosa*) from the Ligurian Sea (Tisnérat-Laborde et al.

2013). This  $\Delta^{14}\text{C}$  record is appropriate for direct comparison with the juvenile portion of otoliths (core material) because each formed in the mixed layer of the central Mediterranean where ocean circulation patterns and mixed layer stratification is well documented (Tisnérat-Laborde et al. 2013).

Two von Bertalanffy growth functions (VBGF) were chosen as a representation of each growth scenario (FGH and SGH; Table 2). In particular, the VBGF obtained by Mellon-Duval et al. (2010) and García-Rodríguez and Esteban (2002) were considered to be the best representation of the FGH. Those produced by Aldebert and Carries (1989) and Levi et al. (1994) were chosen as the best representation of the SGH. The theoretical lifespan of European hake for each growth scenario, assuming longevity ( $t_{\max}$ ) is the time required to attain 95% of asymptotic length ( $L_\infty$ ) (Froese and Binohlan 2000), was determined by replacing length at age  $t$  ( $l_t$ ) with  $0.95 L_\infty$  and solving for  $t_{\max}$  as follows:

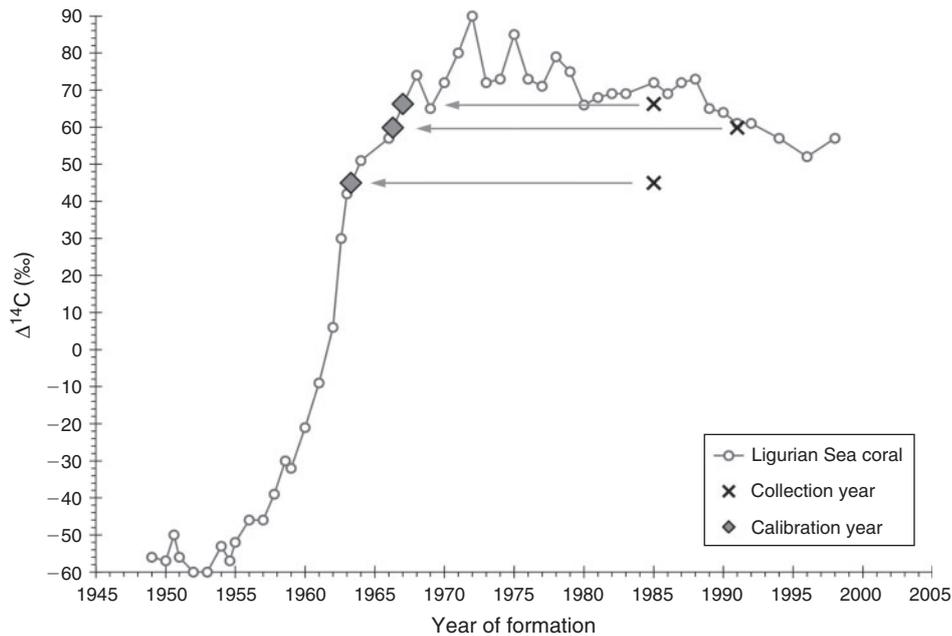
$$t_{\max} = \frac{3}{k} + t_0$$

where  $k$  is the growth coefficient defined in Table 2.

The bomb  $^{14}\text{C}$  ages were ultimately compared with the theoretical  $t_{\max}$  to evaluate its consistency with the longevity estimates from each VBGF scenario.

## Results and discussion

Each otolith sample provided a robust  $^{14}\text{C}$  measurement that could be compared with the recently defined  $\Delta^{14}\text{C}$  reference series from the Ligurian Sea in the first application of bomb



**Fig. 1.** Plot of the  $\Delta^{14}\text{C}$  reference series from a coral in the Ligurian Sea (Tisnérat-Laborde *et al.* 2013) with the measured  $\Delta^{14}\text{C}$  values from otolith cores of three of the largest adult European hake from the Mediterranean Sea. Each measured value is plotted initially at the date of collection and is then projected back in time to the  $\Delta^{14}\text{C}$  reference to determine the year of formation or validated birth year.

$^{14}\text{C}$  dating of a fish species in the Mediterranean Sea (Table 1). The most diagnostic  $\Delta^{14}\text{C}$  measurement was from specimen EH-2 at 45.0‰ because of its well-defined relationship to the  $\Delta^{14}\text{C}$  rise period (before 1967). This  $\Delta^{14}\text{C}$  value was  $\sim 20\%$  lower than the peak and post-peak plateau of the bomb  $^{14}\text{C}$  signal defined by the coral for the region (Fig. 1). Hence, the calibrated birth year of 1963 is well defined with a corresponding validated age of 22 years. The oldest fish was specimen EH-1, where the  $\Delta^{14}\text{C}$  value of 59.9‰ was calibrated to a birth year of 1966. This value is just below the beginning of the peak period, which approaches a low value of 65‰ in 1969, but is below the peak plateau (Fig. 1). Assuming there are no major fluctuations in the reference record plateau, the birth year of this fish was most likely 1966 for an age of 25 years. This finding is further supported by the more massive whole otolith of EH-1 relative to EH-2 (1.042 v. 0.705 g); given the mass–growth of European hake otoliths is a reasonable proxy for age. As expected, the least diagnostic  $\Delta^{14}\text{C}$  value was from specimen EH-3, the smallest fish used in the present study, at 66.3‰. In this case, the  $\Delta^{14}\text{C}$  value was within the range of  $\Delta^{14}\text{C}$  values for the peak plateau of the coral  $\Delta^{14}\text{C}$  record (Fig. 1). Hence, this fish could have had a birth year any time more recently than 1967; however, it can be clearly stated that this fish was not older than 18 years of age because of the temporal limitations of the known bomb  $^{14}\text{C}$  rise period throughout the marine environment (Grottoli and Eakin 2007). Similarly, specimens EH-1 and EH-2 could not have been older than 27 and 24 years of age respectively. This is assuming a 2-year margin of uncertainty in the coral reference record, similar to what has been observed elsewhere with other coral records (Andrews *et al.* 2012, 2013, 2015).

Otoliths have been the hard part of choice for European hake age reading in the Mediterranean Sea since the mid-1960s, but the feasibility debate continues (e.g. Piñeiro and Sainza 2003; Courbin *et al.* 2007). The findings of the present study provide a baseline from which this controversy can begin to be resolved. Use of the VBGF parameters from the FGH and SGH scenarios provided a theoretical range for maximum age ( $t_{\text{max}}$ ) of 13–14 and 27–32 years respectively (Table 2). Because of the diagnostic alignment of the  $\Delta^{14}\text{C}$  values measured in the otolith cores of the two largest fish, it can be concluded that the FGH underestimates maximum age and is not a valid growth scenario for the entire lifespan of European hake (Tables 1, 2). Furthermore, it must be considered that Mellon-Duval *et al.* (2010) have validated by tag–recapture a high growth rate for 96 juvenile European hake females (<24-cm total length). At this time, the validated longevity of large adult European hake obtained in the present study and the tag–recapture validated early growth represent the only reliable length-at-age information for the Mediterranean. This indicates the full life history of European hake could be a combination of rapid early growth followed by a prolonged lifespan. In this regard, it is worth noting that the longevity reported herein refers to fish born in the 1960s and the increase of fishing effort over the past 50 years could have affected biological traits (Hidalgo *et al.* 2009). Moreover, throughout the Mediterranean, this species inhabits heterogeneous habitats characterised by a wide range of abiotic and biotic factors that might induce adaptive changes in growth. Despite the low sample size because of limited availability of European hake at the largest sizes and potentially oldest catch, the goal of estimating longevity (independent of age reading protocol) was achieved with bomb  $^{14}\text{C}$  dating.

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