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# Laser ablation–accelerator mass spectrometry reveals complete bomb $^{14}\text{C}$ signal in an otolith with confirmation of 60-year longevity for red snapper (*Lutjanus campechanus*)

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**Abstract.** Bomb-produced  $^{14}\text{C}$  has been used to make valid estimates of age for various marine organisms for 25 years, but fish ages that lead to birth years earlier than the period of increase in  $^{14}\text{C}$  lose their time specificity. As a result, bomb  $^{14}\text{C}$  dating is limited to a minimum age from the last year of prebomb levels because the temporal variation in  $^{14}\text{C}$  in the marine surface layer is negligible for decades before c. 1958. The longevity of red snapper (*Lutjanus campechanus*) in the Gulf of Mexico remains unresolved despite various forms of support for ages near 50–60 years. Although the age and growth of red snapper have been verified or validated to a limited extent, some scepticism remains about longevity estimates that exceed 30 years. In this study, red snapper otoliths were analysed for  $^{14}\text{C}$  using a novel laser ablation–accelerator mass spectrometry technique to provide a continuous record of  $^{14}\text{C}$  uptake. This approach provided a basis for age validation that extends beyond the normal limits of bomb  $^{14}\text{C}$  dating with confirmation of a 60-year longevity for red snapper in the Gulf of Mexico.

**Additional keywords:** age validation, carbon-14, Gulf of Mexico, Lutjanidae, radiocarbon.

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## Introduction

Bomb-produced  $^{14}\text{C}$  has been used to make valid estimates of age, growth and longevity for various marine organisms for 25 years (Kalish 1993). A long history of success with this approach began with fish otoliths and the validation of purported annual growth zones (Kalish 1995; Campana 1999). Early studies were crude and involved instrumentation that is currently outdated. Not only have the precision and accuracy of the instrumentation and analytical approaches used to measure  $^{14}\text{C}$  increased, but the amount of material required for an otolith measurement has also decreased considerably and continues to improve (Andrews *et al.* 2015; Grammer *et al.* 2015). In concert with these improvements, extraction techniques are more precise – most studies now use a micromilling machine – and there is a more thorough understanding of bomb-produced  $^{14}\text{C}$  in the marine environment (e.g. Grottoli and Eakin 2007; Andrews *et al.* 2016a, 2016c; Druffel *et al.* 2016). As a result, questions of age and longevity have been answered for fishes throughout the world (e.g. Kalish *et al.* 2001; Andrews *et al.* 2011, 2012, 2019; Kestelle *et al.* 2016; Campana *et al.* 2016) and, in some cases, the limits were pushed for species with very small otoliths and geographical origins that were not well constrained (Ishihara *et al.* 2017; Andrews *et al.* 2018a).

Bomb  $^{14}\text{C}$  dating is a modern form of radiocarbon dating (Libby 1955) that relies on the rise of  $^{14}\text{C}$  because of atmospheric testing of thermonuclear devices in the 1950s and 1960s as a temporal reference (Reimer *et al.* 2004; Druffel *et al.* 2016). For marine fishes, the approach typically relies on the extraction and  $^{14}\text{C}$  analysis of calcium carbonate from the earliest otolith growth (core material), for which an alignment is made with a bomb  $^{14}\text{C}$  reference from the marine environment (e.g. Campana 1997; Andrews *et al.* 2011, 2016b). Sampling beyond the core is rarely done because the temporal specificity of the extraction rapidly becomes more difficult to establish as otolith growth layers become thinner with increasing age (e.g. Cook *et al.* 2009). In most fish age validation studies, it was the period of the increase in  $^{14}\text{C}$  (from c. 1958 to c. 1970) that functioned as a diagnostic reference for determining a birth year from otolith  $^{14}\text{C}$  measurements. Recent advances include more extensive coral  $^{14}\text{C}$  records that have extended the utility of the technique in some tropical regions to more recent periods (c. 1980 to present) using the post-peak  $^{14}\text{C}$  decline to age younger fish (Andrews *et al.* 2013, 2016b, 2018a; Ishihara *et al.* 2017; Barnett *et al.* 2018; DeMartini *et al.* 2018). However, fish ages that lead to birth years earlier than the  $^{14}\text{C}$  rise period (prebomb) lose the ability to determine a specific date of formation because

**Table 1.** Data for the red snapper (*Lutjanus campechanus*) specimens and otoliths that were selected for  $^{14}\text{C}$  assays

Fish length, otolith mass and estimated age indicated these fish were good candidates for capturing the full bomb  $^{14}\text{C}$  signal because birth years may predate the rise of bomb-produced  $^{14}\text{C}$  in c. 1958 for the Gulf of Mexico. Estimated ages covered a range of interpretations from various age readers (historical records from Panama City Laboratory, estimates from Barnett *et al.* (2018) and the present study). The potential birth years led to a range of hypothetical bomb  $^{14}\text{C}$  scenarios that can be explored with laser ablation–accelerator mass spectrometry. TL, total length

Laboratory number (Specimen number)	Length (cm TL)	Otolith mass (g)	Estimated age (years)	Collection date	Possible birth years	$^{14}\text{C}$ Scenario
RS07 (2004RS348–2)	83.0	3.78 <sup>A</sup>	40–55 <sup>B</sup>	12 Sept. 2004	1949–64	Prebomb to rise
RS04 (2002RS532–125)	85.3	3.67 <sup>A</sup>	33–48 <sup>B</sup>	6 Aug. 2002	1954–69	Prebomb to rise
RS17 (2010RS286–1293)	88.0	3.88	36–56 <sup>B</sup>	23 May 2010	1954–74	Prebomb to peak

<sup>A</sup>Mean value for both whole otoliths

<sup>B</sup>A. H. Andrews aged sections from RS07, RS04 and RS17 to 55, 48 and 43 years respectively.

$^{14}\text{C}$  levels in the marine surface layer plateau in the decades before the  $^{14}\text{C}$  rise. Hence, bomb  $^{14}\text{C}$  dating is limited by prebomb birth years in terms of establishing longevity (Baker and Wilson 2001; Cook *et al.* 2009; Andrews *et al.* 2013) – only a minimum age can be validated from the last year of prebomb levels, leaving longevity in question.

Although the age and growth of red snapper was verified or validated to a limited extent using otolith margin analyses, tag–recapture data, lead–radium dating and bomb  $^{14}\text{C}$ , some scepticism remains about longevity estimates that exceed 30 years (e.g. Szedlmayer and Beyer 2011). Otolith margin or tagging studies have led to some observations of red snapper age but are limited to early growth and short time spans (Patterson *et al.* 2001; Wilson and Nieland 2001). Lead–radium dating can be useful for age estimates on the order of a decade to ~100 years (Andrews *et al.* 2002, 2009; Andrews 2016; Tracey *et al.* 2017), but the method is typically limited to providing approximate ages for groups of fish otoliths (studies are subject to minimum mass and lead–radium activity requirements; Andrews *et al.* 1999a, 1999b). This approach has led to support for red snapper longevity exceeding 30 years (Baker *et al.* 1999). In contrast, bomb  $^{14}\text{C}$  dating has provided valid estimates of age for individual red snapper up to ~38 years (Baker and Wilson 2001). This method has evolved considerably since its inception, with recent indications that red snapper can live to at least 44 years (Barnett *et al.* 2018); the upper limit uncertainty for this age is based on a lack of time specificity for prebomb  $^{14}\text{C}$  measurements from modern otoliths.

In this study, red snapper otoliths were analysed for  $^{14}\text{C}$  using a novel laser ablation–accelerator mass spectrometry (LA-AMS) technique to provide a continuous record of  $^{14}\text{C}$  uptake (Welte *et al.* 2016) and a basis for age validation that may extend beyond the normal limits of bomb  $^{14}\text{C}$  dating. This approach may reveal the location in the otolith cross-section where bomb-produced  $^{14}\text{C}$  rises in a single continuous measurement (youngest to oldest material and covering the lifespan of the fish). The LA-AMS  $^{14}\text{C}$  time series from an otolith cross-section can be aligned with estimated years of formation from age reading of growth zones. The goal would be to assess the alignment or misalignment of the initial  $^{14}\text{C}$  rise (c. 1958) with the regional  $^{14}\text{C}$  reference to potentially refine age estimates that may not be accurate. In some scenarios, validated ages within the prebomb period are possible by locating the initial  $^{14}\text{C}$  rise at advanced ages away from the earliest otolith growth. The approach may also differentiate ages

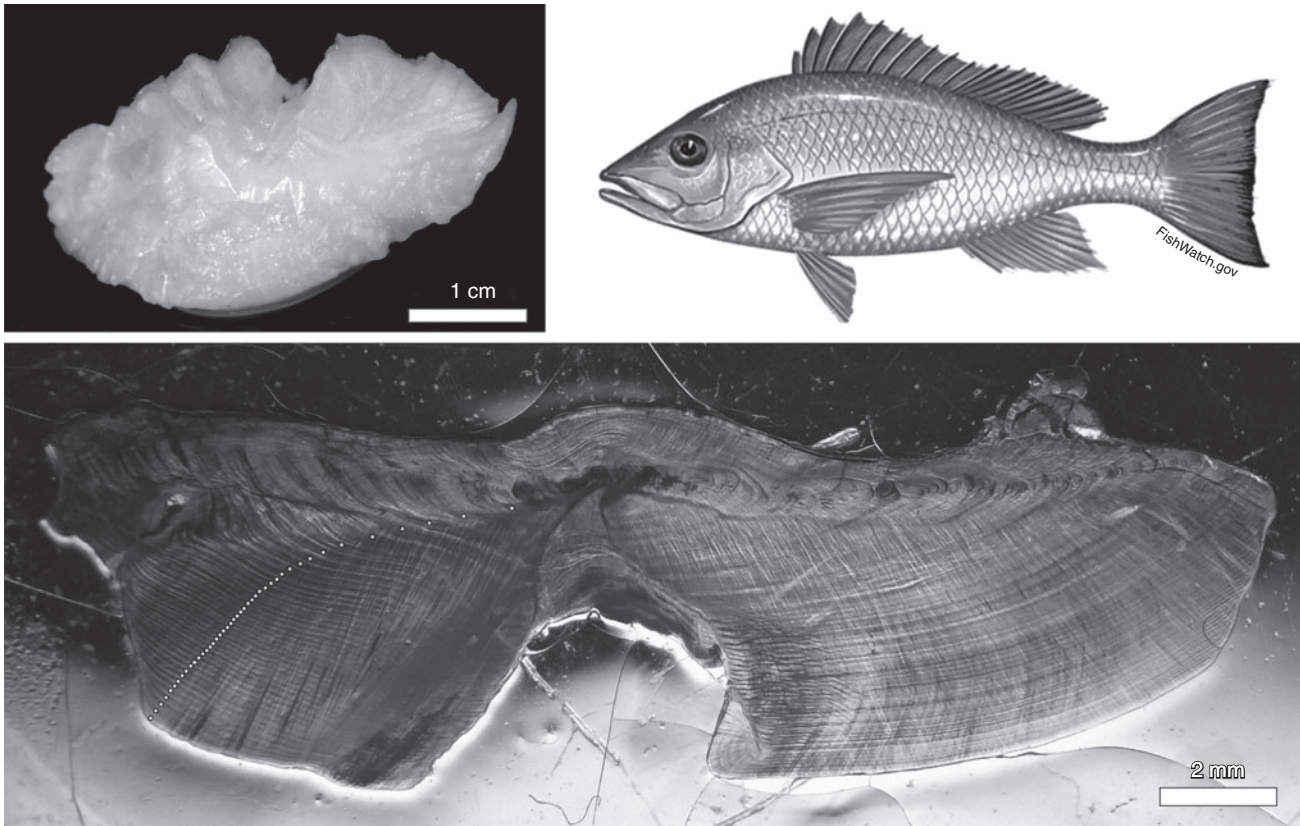
from  $^{14}\text{C}$  values that lead to ambiguous birth years (potentially attributed to either the  $^{14}\text{C}$  rise or decline period), a discrepancy that can be on the order of several decades.

## Materials and methods

The red snapper otoliths selected for LA-AMS were from fish specimens with age estimates that provided lifespans covering the bomb  $^{14}\text{C}$  signal. Great otolith mass (approaching or exceeding 4 g) was also considered to increase the chances of using the oldest fish. Based on these observations, three specimens were selected for the study (Table 1). The study began with a series of preliminary assays on the first otolith (RS07), followed by refined assays on two additional otoliths (RS04, RS17). These fish were among the largest for this species, exceeding 80-cm total length (TL), with whole otolith masses approaching 4 g (Fig. 1). Each fish was collected from the Gulf of Mexico by port samplers associated with the Panama City Laboratory (PCL) of the Southeast Fisheries Science Center, National Oceanic and Atmospheric Administration (NOAA) Fisheries. Estimated ages for the three selected specimens ranged from 33 to >50 years, with some level of variability in estimated age for each specimen from various age reader interpretations. These age estimates were either historical (anonymous) from age readers at PCL or from age reading during the present study (by A. H. Andrews). Similar estimates were made for two of the specimens in a parallel study (Barnett *et al.* 2018) and were within the range provided for the present study. Collection dates with age estimates revealed potential birth years in the 1950s for each specimen, but two fish may have been younger with birth years as early as 1964 and 1974, given age reading is providing an accurate age within the proposed age ranges. Two fish probably extend into the prebomb period, whereas one fish may have started life during the bomb  $^{14}\text{C}$  rise or peak period (Table 1).

## Conventional AMS

Two of the three otoliths were sampled for the earliest otolith growth (core material) using a New Wave Research (ESI-NWR Division; Fremont, CA, USA) micromill and were analysed in the standard graphitisation AMS manner (Table 2; for details, see Andrews *et al.* 2015). These  $^{14}\text{C}$  values were used to generally confirm the predicted bomb  $^{14}\text{C}$  scenario and exemplify the limitations of single-sample bomb  $^{14}\text{C}$  dating (Barnett *et al.* 2018).



**Fig. 1.** Illustration of red snapper (*Lutjanus campechanus*) and of one of the largest otoliths at more than 4 g. The bottom image is a transverse thin section of the otolith from specimen RS17. The series of small white dots on the left side of the section mark the growth zones counted for an age estimate of 43 years (dorsal).

**Table 2.** Radiocarbon data for the red snapper (*Lutjanus campechanus*) otoliths where the core (birth year) material of the adult otolith was extracted with a micromilling machine

The single measurements allowed refinement of the age from the original range of estimates to a minimum age. No initial core measurement was made for RS17. Fraction modern ( $F^{14}C$ ) data are given as the mean  $\pm$  2 s.d.

Laboratory number (Specimen number)	$F^{14}C$ (core)	$\delta^{13}C$	$F^{14}C$ age scenario	Birth years	Minimum age (years)
RS07 (2004RS348–2)	$0.9464 \pm 0.0020^A$	–2.69	Prebomb	<1958	>46
RS04 (2002RS532–125)	$0.9502 \pm 0.0022^A$	–3.59	Prebomb	<1958	>44

<sup>A</sup>Sample processed with standard graphitisation accelerator mass spectrometry at National Ocean Sciences Accelerator Mass Spectrometry facility at Woods Hole Oceanographic Institution NOSAMS (WHOI).

### Gas AMS

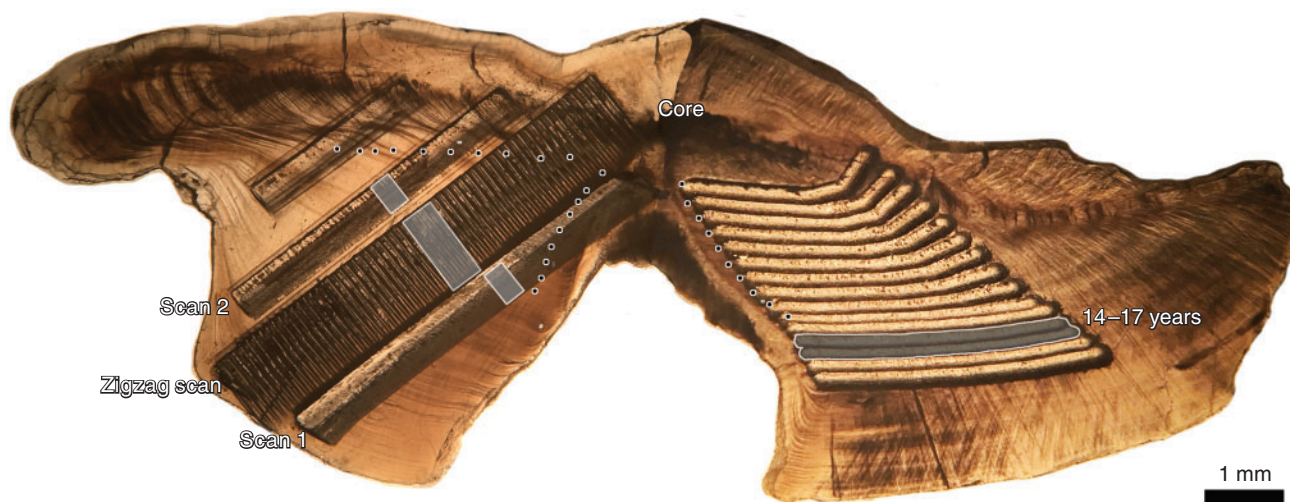
The first otolith analysed (RS07) was sampled beyond the initial milled core  $^{14}C$  measurement and into the growth zone sequence using the micromill. This series of samples was analysed by gas AMS (Rosenheim *et al.* 2008; Wacker *et al.* 2013). In this time-efficient online approach, the carbonate samples are placed in septum-sealed vials under a helium atmosphere.  $CO_2$  is released from the carbonates by the addition of phosphoric acid. In contrast with conventional graphite AMS analysis where the liberated  $CO_2$  is reduced to graphite and measurements are performed on solid targets, here the  $CO_2$  gas is concentrated by means of a zeolite trap. In a final step, the  $CO_2$  is transferred with He carrier gas into the syringe of the gas interface system, where it is further diluted with helium and fed into the gas ion source of

a Mini Carbon Dating System (MICADAS) AMS system (Ionplus, Dietikon, Switzerland) for gas  $^{14}C$  analysis. The goal was to locate the position in the otolith section where the bomb  $^{14}C$  rise occurred using the radial sampling and to corroborate the findings from exploratory LA-AMS assays (Table 3; Fig. 2).

### Laser ablation accelerator mass spectrometry

The initial LA-AMS test scans were performed on an otolith section (RS07) originally prepared for making estimates of age from growth zone counting. The section was very thin ( $\sim 0.3$  mm) and previously mounted to a glass microscope slide with an unknown mounting medium. This sample was used opportunistically in a first attempt at locating the bomb  $^{14}C$  rise





**Fig. 2.** Transverse section of the red snapper otolith from specimen RS07 with the series of samples taken using both conventional micromilling extraction (right side) and laser ablation–accelerator mass spectrometry (LA-AMS; left side). Counts of well-defined growth zones were made before the sampling and are denoted as black dots radiating out from the core (earliest growth) to 10 years of age. Extractions with the micromill were estimated to be annual out to 10 years and then composed of 2–3 years of material out to an estimated age of 23 years. Analysis of the milled samples for  $^{14}\text{C}$  revealed the bomb  $^{14}\text{C}$  rise at 14–17 years (1958–61). Measurements of  $^{14}\text{C}$  from LA-AMS revealed the rise in bomb  $^{14}\text{C}$  in a similar structural location to the micromilling series for all LA-AMS scans (grey boxes). Scans 1 and 2 were early feasibility and instrument optimisation runs that provided a first look at the region of  $^{14}\text{C}$  rise (see Fig. S1, S2). The zigzag scan was focused on providing a stronger and longer-lasting signal, before development of the parallelogram zigzag or precision scan (Fig. 3). The earliest core extraction with the micromill was performed on the other otolith of the pair and is not shown here (Table 2).

**Table 3.** Radiocarbon data for the otolith core and radial sample series taken from sample RS07

Prebomb  $^{14}\text{C}$  levels covered a time span greater than expected (dating back to 1944 and an age of 60 years). Growth year was the year of formation for the respective portion of the otolith (see Fig. 2). Ages and years were rounded to the nearest whole number. Post-corrected  $\Delta^{14}\text{C}$  values are provided for comparison with other records. Fraction modern ( $F^{14}\text{C}$ ) and  $\Delta^{14}\text{C}$  data are given as the mean  $\pm$  2 s.d.

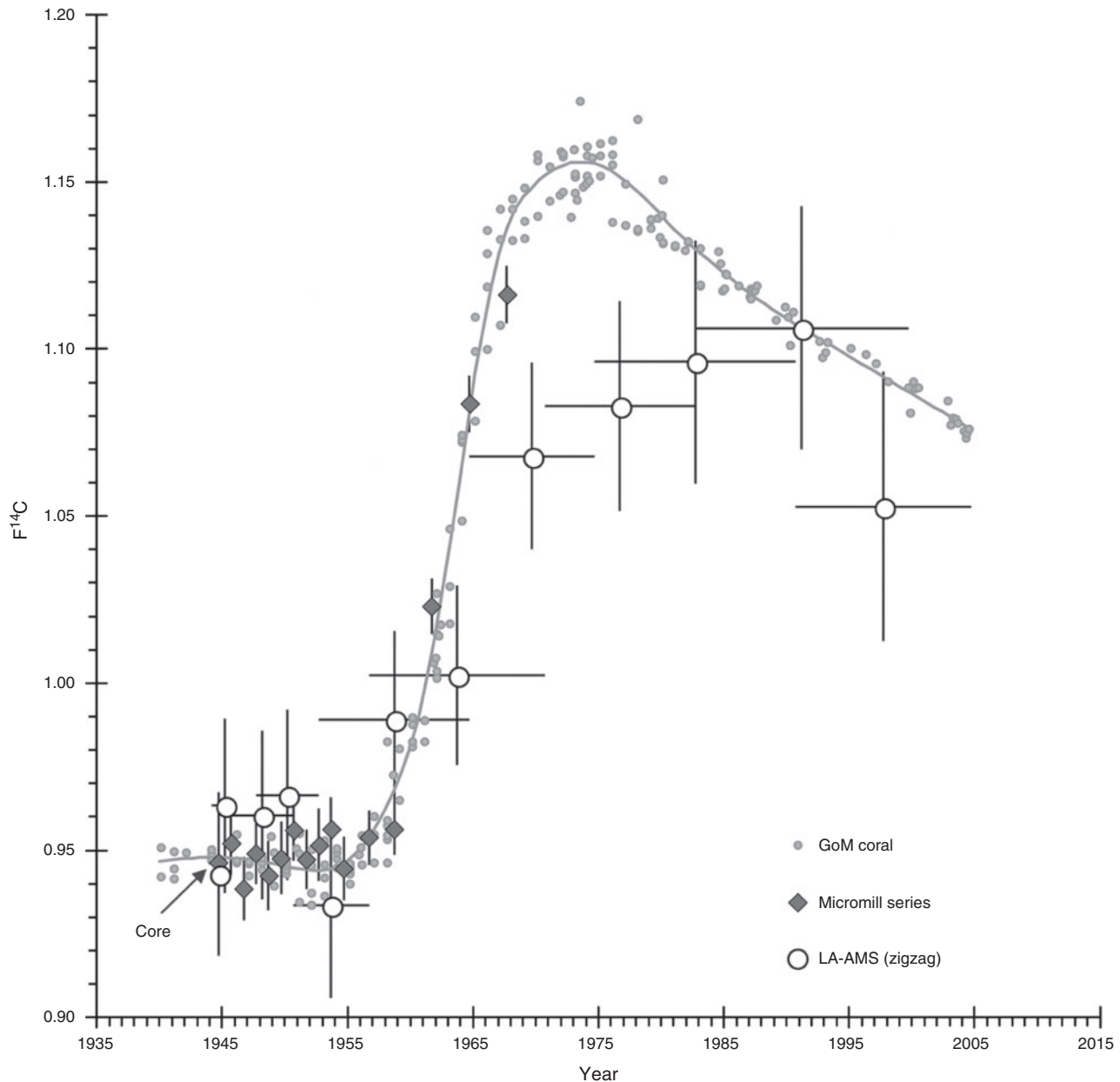
Laboratory number (Specimen number)	$F^{14}\text{C}$	$F^{14}\text{C}$ age scenario	Growth year	Sample age (years)	$\Delta^{14}\text{C}$ (‰)
RS07 core	$0.9464 \pm 0.0020^{\text{A}}$	Prebomb	0 (1944)	60	$-53.0 \pm 2.0$
RS07-S01	$0.9523 \pm 0.0097^{\text{B}}$	Prebomb	1 (1945)	59	$-47.2 \pm 9.7$
RS07-S02	$0.9384 \pm 0.0095^{\text{B}}$	Prebomb	2 (1946)	58	$-61.2 \pm 9.5$
RS07-S03	$0.9490 \pm 0.0090^{\text{B}}$	Prebomb	3 (1947)	57	$-50.8 \pm 9.0$
RS07-S04	$0.9425 \pm 0.0105^{\text{B}}$	Prebomb	4 (1948)	56	$-57.4 \pm 10.5$
RS07-S05	$0.9477 \pm 0.0109^{\text{B}}$	Prebomb	5 (1949)	55	$-52.3 \pm 10.9$
RS07-S06	$0.9560 \pm 0.0092^{\text{B}}$	Prebomb	6 (1950)	54	$-44.1 \pm 9.2$
RS07-S07	$0.9473 \pm 0.0090^{\text{B}}$	Prebomb	7 (1951)	53	$-52.9 \pm 9.0$
RS07-S08	$0.9517 \pm 0.0110^{\text{B}}$	Prebomb	8 (1952)	52	$-48.7 \pm 11.0$
RS07-S09	$0.9563 \pm 0.0094^{\text{B}}$	Prebomb	9 (1953)	51	$-44.1 \pm 9.4$
RS07-S10	$0.9446 \pm 0.0094^{\text{B}}$	Prebomb	10 (1954)	50	$-56.0 \pm 9.4$
RS07-S11	$0.9538 \pm 0.0082^{\text{B}}$	Prebomb	12 (1956)	48	$-47.0 \pm 8.2$
RS07-S12	$0.9564 \pm 0.0079^{\text{B}}$	Prebomb	14 (1958)	46	$-44.6 \pm 7.9$
RS07-S13	$1.0230 \pm 0.0083^{\text{B}}$	Rise	17 (1961)	43	$21.6 \pm 8.3$
RS07-S14	$1.0837 \pm 0.0085^{\text{B}}$	Rise	20 (1964)	40	$81.7 \pm 8.5$
RS07-S15	$1.1164 \pm 0.0086^{\text{B}}$	Rise	23 (1967)	37	$114.0 \pm 8.6$

<sup>A</sup>Sample processed with standard graphitisation accelerator mass spectrometry (AMS) at National Ocean Sciences Accelerator Mass Spectrometry (NOSAMS) facility.

<sup>B</sup>Sample processed with gas AMS at ETH Zurich.

by LA-AMS within an otolith. The set-up used for these initial measurements is described in Welte *et al.* (2016); the sample was ablated by a 193-nm laser (ArF excimer laser: Ex5, GAM LASER, Orlando, FL, USA) with a rectangular spot

measuring  $110 \times 680 \mu\text{m}$  in its focus. A helium carrier gas flow rate of  $\sim 1.5 \text{ mL min}^{-1}$  was used to transport the gases formed during laser ablation (including CO and CO<sub>2</sub>) into the gas ion source of the MICADAS for  $^{14}\text{C}$  analysis. An



**Fig. 3.** Comparison of  $^{14}\text{C}$  measurements from both micromilling and the laser ablation–accelerator mass spectrometry (LA-AMS) zigzag scan (Fig. 2) with the regional coral bomb  $^{14}\text{C}$  reference series for the Gulf of Mexico (GoM; Andrews *et al.* 2013). The initial core measurement indicated the age was  $>46$  years but could have been up to 55 years given the high count scenario (Table 2). The micromilled sample series pushed the age to 60 years with a well-matched time series to the coral  $^{14}\text{C}$  record ( $\pm 1$ – $2$  years). The zigzag LA-AMS data were in approximate agreement with the coral  $^{14}\text{C}$  record considering the cross-growth zone pattern of this initial assessment. Regardless of the low resolution, the goal of detecting a complete bomb  $^{14}\text{C}$  signal within an otolith was achieved. The attenuated peak may be attributed to an influence of deep water with a slightly depleted  $^{14}\text{C}$  record later in adult life. Horizontal error bars for LA-AMS were derived from the age range of the scan path width for each sample block and vertical error bars were 2 s.d. of the block mean.

exploratory scan (Fig. 2, Scan 1) revealed a  $^{14}\text{C}$  inflection that could be attributed to the rise of bomb-produced  $^{14}\text{C}$ , and the difference was measurable across the otolith section. This finding led to a more refined linear scan (Fig. 2, Scan 2) that further revealed the  $^{14}\text{C}$  inflection in greater detail. This exploratory series led ultimately to a ‘rectangular’ zigzag scan (precision scan; Welte *et al.* 2016) where the zigzag-shaped

moving pattern of the sample under the laser was performed perpendicular to its continuous axial moving direction, ultimately resulting in a rectangular sampling area (Fig. 2). A more thorough sampling of the otolith material could be achieved with the zigzag scanning and allowed documentation of the precise location of the  $^{14}\text{C}$  rise in the otolith section (for these exploratory scan lines, see Fig. 2).

Based on the initial results, a refined LA-AMS analysis procedure was used for the next two otoliths (RS04, RS17) including modifications of: (1) the sample preparation procedure; (2) the LA-AMS set-up; and (3) the LA-based sampling strategy (see below).

#### *Modifications to sample preparation*

The small otoliths required low-loss preparation to allow for precise positioning of the laser ablation scan lines relative to the growth zone structure in the otolith. Consequently, thicker (~1 mm) transverse sections were prepared and mounted in an epoxy (Epo-Tek 301-2/1LB/A with hardener 301-2/1LB/B; Epoxy Technology, Billerica, MA, USA) using disposable plastic embedding moulds (Peel-A-Way, R-40 22 × 40-mm rectangular, 20 mm deep; Catalogue number 18646C; Poly-science Inc., Warrington, PA, USA). Sections were placed directly on the bottom of the embedding moulds after minor polishing to make sure the cut surface was flat. Epoxy was added to the top of the otolith section to a depth of a few millimetres to create a wafer that allowed easy handling. The mounted sections were polished with 1200-grit silicon carbide wet-dry sandpaper on a lapidary wheel to remove epoxy and other surface contamination from the analysis surface. The embedded sample was cut for fitting and proper orientation within the LA-AMS sample carriage.

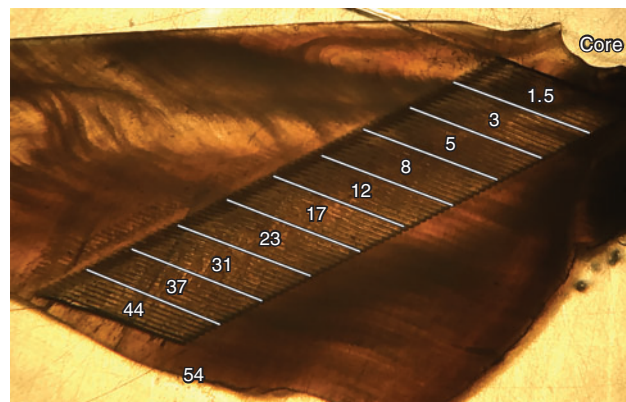
#### *Modifications to the LA-AMS set-up*

LA-AMS analysis of the two otoliths (RS04, RS17) was performed using a refined set-up with a smaller laser spot of  $75 \times 140 \mu\text{m}^2$  on the sample allowing for improved spatial resolution.

#### *Modifications to the LA-based sampling strategy*

The rectangular zigzag scanning pattern used for the LA-AMS test scan on RS07 proved to be insufficient for revealing the imprinted  $^{14}\text{C}$ -bomb signature within this sample (compare Fig. 3 and the Supplementary material) because it crossed multiple growth zones and consequently different years of growth during one zigzag step. In order to provide greater spatiotemporal specificity, the zigzag scans performed on RS04 and RS17 were improved by positioning the zigzag movement and the continuous axial displacement at an optimised angle. This procedure ensured sampling within the same growth zone structure during single zigzag steps and yielded an overall sampling area of a parallelogram (compare Fig. 4 and 5).

On each of the two otoliths (RS04, RS17), two LA-AMS sampling areas across the dorsal and ventral portions were selected. Because for the refined LA-AMS analysis thicker otolith sections were prepared, four stacked scans were performed at each sampling location, yielding eight scans for each otolith. On the RS04 otolith (Fig. 4), one LA-AMS sampling area may have been compromised by inadvertent inclusion of  $^{14}\text{C}$ -depleted epoxy (Welte *et al.* 2016) and was thus not considered. The overall analysis time per otolith sample was on the order of 2 h, whereas ~6–8 mg of material was consumed. The resulting precision using the refined setup and sampling method is in the order of 1% for a single LA-AMS data point on a modern sample.

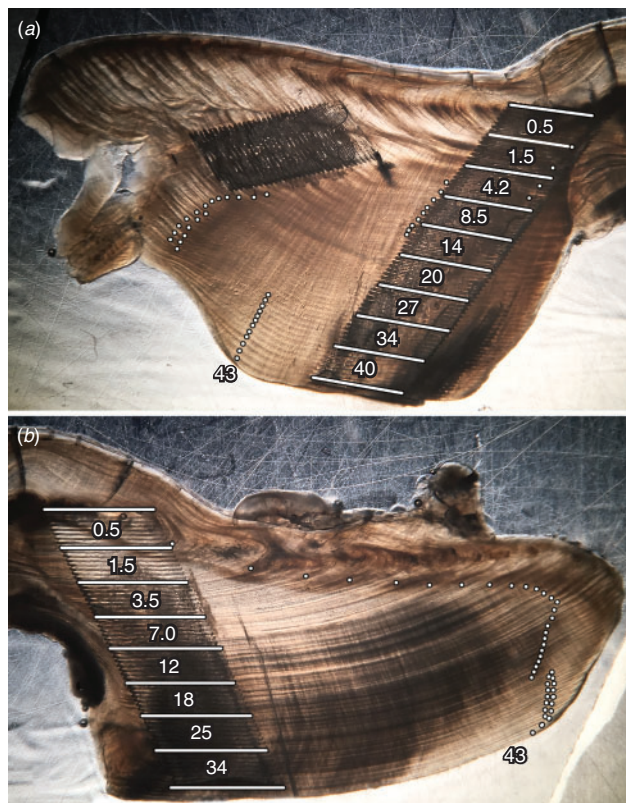


**Fig. 4.** Cross-sectioned otolith of specimen RS04 with the micromilled core extraction visible as a notch in the upper right and the laser ablation–accelerator mass spectrometry precision scan (zigzag parallelogram) progressing diagonally from near the core to near the outer otolith edge. Delineated are the scan analysis blocks (600  $\mu\text{m}$ ) that provided mean  $^{14}\text{C}$  values for the respective regions of the otolith. Mean age estimates from growth zone counting are enumerated along the centre of the scan. The original zone counting age estimate was shifted by 6 years from the maximum counted age of 48 years for an alignment with the  $^{14}\text{C}$  rise and an age of 54 years (Fig. 6). Zone counts for this section could not be clearly imaged and are not specifically marked.

Because the  $^{14}\text{C}$  data from LA-AMS is provided quasi-continuously (in 10-s intervals), it must be processed and recombined offline to reach the anticipated spatial resolution. For each scan, the number of  $^{14}\text{C}$  data points per unit time, and thus scan distance, were chosen in a way that provided enough spatial resolution while minimising instrument measurement variability (e.g. a wider range of years leads to lower instrument measurement error but can compromise the resolution needed to resolve the initial  $^{14}\text{C}$  rise). Therefore, a dedicated data reduction strategy was applied (Yeman *et al.* 2017). Briefly, first, single-cycle (10 s) AMS data were blank subtracted, isotopic fractionation corrected using concurrently acquired  $\delta^{13}\text{C}$  data and normalised using standard data processing routines (Wacker *et al.* 2010). Because the focus was on relative changes in  $^{14}\text{C}$  within the scans, the systematic error from standard normalisation (0.5%) was not considered. Second, single-cycle data points were subdivided and subsequently recombined (using a weighted average) so that for each resulting data point a spatial resolution (along the otolith growth axis) of 500–600  $\mu\text{m}$  was reached. The final data are reported as  $F^{14}\text{C}$  (fraction modern), which corresponds to the activity ratio of the sample relative to the primary  $^{14}\text{C}$  standard (Oxalic acid I, SRM 4990B) and consequently does not depend on the year of sample formation (Reimer *et al.* 2004). Because the main goal of this study was to determine the age of formation of the otolith layers, this is the most appropriate  $^{14}\text{C}$  unit. Calculated  $\Delta$  (often also used as  $\Delta^{14}\text{C}$ ) values are provided for comparison with previous studies, but are not primarily used because of the circularity in correcting to the time of formation (Stuiver and Polach 1977).

By comparing the section images used for growth layer counting with the corresponding laser tracks, ages were assigned to the LA-AMS data and then correlated with a regional coral  $^{14}\text{C}$  reference record (Andrews *et al.* 2013). Final adjustments to





**Fig. 5.** Laser ablation accelerator mass spectrometry (LA-AMS) precision scans for both sides of the otolith section from specimen RS17 where growth zone structure was well defined: (a) long scan (dorsal), (b) short scan (ventral). Age was estimated as 43 years with original estimates that ranged from 36 to 56 years using different interpretations. Zone counts from the 43-year scenario were used to estimate mean age for the LA-AMS block means enumerated within the scans. The resulting  $^{14}\text{C}$  time series from each scan was well aligned with the coral  $^{14}\text{C}$  reference (Fig. 6) and proved useful in validating an age from near peak values; normally, a single measurement near the  $^{14}\text{C}$  peak is not diagnostic for age. No initial core  $^{14}\text{C}$  measurement was made for this specimen.

the age estimates and consequent year of formation because of small initial misalignments observed for the  $^{14}\text{C}$  rise in the otolith data were made to achieve congruence with the regional coral  $^{14}\text{C}$  reference. The refined ages were compared to growth zone counts and, in some cases, to the images of well-defined otolith sections to cross-check the age model.

## Results

### Conventional AMS and LA-AMS feasibility

The first transverse otolith section used in exploring the feasibility of LA-AMS to measure the bomb  $^{14}\text{C}$  signal through the ontogeny of an adult red snapper (RS07) revealed a consistent location in the cross-section and year of formation for the rise of bomb  $^{14}\text{C}$  between the methods (Fig. 2, 3). An original low estimate of 40 years was invalid based on a single core extraction that revealed the fish must be older than 46 years from prebomb levels (Table 2; Fig. 3). As a result, age estimates were reassessed in the sectioned otolith by counting finer growth zone

structure that led to ages up to 55 years and were consistently quantified. Hence, it was hypothesised that the bomb  $^{14}\text{C}$  rise would occur between the core and the 10th year of growth in the otolith section. The subsequent extraction and conventional gas AMS  $^{14}\text{C}$  analysis of 15 micromilled radial samples revealed the bomb  $^{14}\text{C}$  rise was in a more recently formed part of the otolith section, corresponding to mean estimated fish ages of 14–17 years (Table 3; Fig. 2).

### LA-AMS application: exploratory scans

Measurements of  $^{14}\text{C}$  in specimen RS07 using LA-AMS revealed that the bomb  $^{14}\text{C}$  rise could be detected and was in a similar location and year of formation relative to a conventional AMS analysis of the micromilled series (Fig. 2, 3, S1, S2). The proof-of-concept for use of LA-AMS to detect the bomb  $^{14}\text{C}$  rise in an otolith section was addressed initially with two exploratory scans: Scan 1 revealed an estimate of the bomb  $^{14}\text{C}$  signal and Scan 2 was used to further investigate the  $^{14}\text{C}$  signal and to optimise the instrument, which was followed by a first trial of the zigzag scan on an otolith (Fig. 2). This approach was used to increase the sampling time within single growth layers and to provide a stronger measure of the bomb  $^{14}\text{C}$  rise, especially considering this otolith section was very thin (0.3 mm) and could not be analysed for an extended period in one location without reaching the mounting medium. Although the zigzag approach led to a longer signal collection time along the sample, the results suffered from a mix of  $^{14}\text{C}$  levels from various formation years through the scanning analysis timeline (offset from growth direction and layering; Table 4, Fig. 2). Specifically, as the scan entered the bomb  $^{14}\text{C}$  rise region of the otolith, the zigzag pattern of the laser criss-crossed otolith growth zones that were formed in different years, some of which would have had markedly different  $^{14}\text{C}$  levels. The result was a muted bomb  $^{14}\text{C}$  record across the otolith; however, the goal of this specimen analysis was a proof-of-concept for moving forward with analysis of additional fish specimens. LA-AMS on otolith RS07 achieved this goal by tracing a complete bomb  $^{14}\text{C}$  signal covering the prebomb, peak and decline periods in an otolith section on three separate occasions (Fig. 2, 3, S1, S2). These findings led to the development of a parallelogram zigzag pattern (precision scan) to maintain greater time specificity on the two following red snapper otolith specimens.

The findings from both conventional gas AMS of micromilled samples and the LA-AMS scans supported an estimated age for RS07 that was slightly older than the maximum estimated age of 55 years from growth zone counting (observed by A. H. Andrews). Given the rise of bomb  $^{14}\text{C}$  within the section was located at counts near the mid-teens and the minimum age of 46 years was established with the single core  $^{14}\text{C}$  measurement, the birth year would be 1946 for an age of 60 years and a potential uncertainty of just a 1–2 years from the milled extraction series width (Table 3; Fig. 3).

### LA-AMS application: precision scans

The otolith sections from RS04 and RS17 were used to refine the application of LA-AMS to measure bomb  $^{14}\text{C}$  with greater precision through the ontogeny of adult red snapper. The original lowest estimates of age for RS04 were eliminated below 44 years by the single core extraction  $^{14}\text{C}$  measurement



**Table 4. Radiocarbon and age data as block means from the laser ablation–accelerator mass spectrometry zigzag scan and the growth zone counting for specimen RS07**

Ages and years were rounded to the nearest whole number. Post-corrected  $\Delta^{14}\text{C}$  values are provided for comparison with other records. Fraction modern ( $F^{14}\text{C}$ ) and  $\Delta^{14}\text{C}$  data are given as the mean  $\pm$  2 s.d.

Laboratory number (Specimen number)	$F^{14}\text{C}$	$F^{14}\text{C}$ age scenario	Growth year	Sample age (years)	$\Delta^{14}\text{C}$ (‰)
RS07-Z01	$0.943 \pm 0.024$	Prebomb	0.5 (1945)	60	$-56.6 \pm 2.4$
RS07-Z02	$0.963 \pm 0.026$	Prebomb	1.0 (1945)	59	$-36.4 \pm 2.6$
RS07-Z03	$0.961 \pm 0.025$	Prebomb	3.5 (1948)	56	$-39.5 \pm 2.5$
RS07-Z04	$0.967 \pm 0.025$	Prebomb	5.5 (1950)	54	$-33.8 \pm 2.5$
RS07-Z05	$0.934 \pm 0.028$	Prebomb	9.0 (1954)	51	$-66.9 \pm 2.8$
RS07-Z06	$0.989 \pm 0.026$	Rise	14 (1959)	46	$-11.7 \pm 2.6$
RS07-Z07	$1.002 \pm 0.027$	Rise	19 (1964)	41	$0.8 \pm 2.7$
RS07-Z08	$1.068 \pm 0.028$	Rise	25 (1970)	35	$65.8 \pm 2.8$
RS07-Z09	$1.083 \pm 0.031$	Peak	32 (1977)	28	$79.7 \pm 3.1$
RS07-Z10	$1.096 \pm 0.036$	Peak	38 (1983)	22	$91.8 \pm 3.6$
RS07-Z11	$1.106 \pm 0.036$	Peak	46 (1991)	13	$100.9 \pm 6.6$
RS07-Z12	$1.053 \pm 0.040$	Decline	53 (1998)	7	$46.4 \pm 4.0$

**Table 5. Radiocarbon data for the laser ablation–accelerator mass spectrometry precision scan taken from specimen RS04**

The timeline determined from growth zone counting to 54 years provided a well-matched  $^{14}\text{C}$  rise time, extending the minimum age from the single core  $^{14}\text{C}$  measurement by 10 years and older than what growth zone counting covered (Table 1). Ages and years were rounded to the nearest whole number. Post-corrected  $\Delta^{14}\text{C}$  values are provided for comparison with other records. Fraction modern ( $F^{14}\text{C}$ ) and  $\Delta^{14}\text{C}$  data are given as the mean  $\pm$  2 s.d.

Laboratory number (Specimen number)	$F^{14}\text{C}$	$F^{14}\text{C}$ age scenario	Growth year	Sample age (years)	$\Delta^{14}\text{C}$ (‰)
RS04-L01	$0.937 \pm 0.009$	Prebomb	1.5 (1950)	53	$-59.3 \pm 9.0$
RS04-L02	$0.944 \pm 0.009$	Prebomb	3.0 (1952)	51	$-50.2 \pm 9.0$
RS04-L03	$0.946 \pm 0.009$	Prebomb	5.0 (1954)	49	$-60.7 \pm 9.0$
RS04-L04	$0.953 \pm 0.009$	Prebomb	8.0 (1957)	46	$-21.9 \pm 9.0$
RS04-L05	$1.000 \pm 0.009$	Rise	12 (1961)	42	$28.1 \pm 9.0$
RS04-L06	$1.063 \pm 0.009$	Rise	17 (1966)	37	$72.8 \pm 9.0$
RS04-L07	$1.097 \pm 0.009$	Peak	23 (1972)	31	$107.7 \pm 9.0$
RS04-L08	$1.104 \pm 0.009$	Peak	31 (1979)	23	$107.1 \pm 9.0$
RS04-L09	$1.111 \pm 0.010$	Peak	37 (1986)	17	$92.9 \pm 10.0$
RS04-L10	$1.081 \pm 0.013$	Decline	44 (1993)	10	$75.4 \pm 13.0$

(Table 1); a prebomb birth year earlier than 1958 was the only possible scenario (Table 2). No core measurement was made for RS17 and age could have been 36–56 years with prebomb to peak birth years (Table 1). LA-AMS revealed two different bomb  $^{14}\text{C}$  patterns from which the prebomb scenario for RS04 was confirmed and a younger age scenario was discovered for RS17. Although both had the potential to be fish with prebomb birth years (Table 1), RS17 was restricted to near peak  $^{14}\text{C}$  levels followed by a  $^{14}\text{C}$  decline and RS04 was similar to RS07 with a full bomb  $^{14}\text{C}$  signal and greater precision (Tables 5, 6).

The time-specific parallelogram zigzag design (precision scan) led to circumstances that were easier to align (relative to RS07) with the otolith age estimates through ontogeny (Fig. 4, 5). Age estimates were reassessed for RS04 and RS17 in the sectioned otoliths by counting fine growth zone structure (observed by A. H. Andrews), which is similar to the growth zone structure that was used for RS07 and is exemplified by the RS17 section (Fig. 1). These counts led to consistent ages

of 48 and 43 years for RS07 and RS17 respectively (Fig. 4, 5). Although the ages and dates of formation from the LA-AMS data series for RS17 were in alignment with the coral  $^{14}\text{C}$  reference record, RS04 required an additional 6 years to the maximum growth zone-derived age of 48 years to align properly (Fig. 6). Furthermore, although the single core  $^{14}\text{C}$  value led to a minimum age of 44 years (Table 2), the LA-AMS data series indicated RS04 was 54 years old because of the extent of prebomb levels in the earliest growth. This is similar to what was found for RS07, where the age was extended 5 years beyond the growth zone counting maximum (55 years). The minor misalignment for the more recent adult samples within the series, attenuated and phase lagged relative to the coral  $^{14}\text{C}$  record (Fig. 6), may be attributed to either uptake from deeper  $^{14}\text{C}$ -depleted waters or the laser sampling more years of growth than expected, leading to a dampened signal. Either way, the results were consistent with the findings for RS07 (Fig. 3). The replicated sample series from RS17 provided a consistent alignment with the bomb  $^{14}\text{C}$  peak

**Table 6. Radiocarbon data for the long and short laser ablation–accelerator mass spectrometry precision scans taken from both sides of the otolith section of specimen RS17**

The  $^{14}\text{C}$  timelines agreed with the estimated ages relative to the coral  $^{14}\text{C}$  reference. Each RS17 series precluded alignment with a younger age associated with a strictly  $^{14}\text{C}$  decline scenario. Post-corrected  $\Delta^{14}\text{C}$  values are provided for comparison with other records. Fraction modern ( $F^{14}\text{C}$ ) and  $\Delta^{14}\text{C}$  data are given as the mean  $\pm$  2 s.d.

Laboratory number (Specimen number)	$F^{14}\text{C}$	$F^{14}\text{C}$ age scenario	Growth year	Sample age (years)	$\Delta^{14}\text{C}$ (‰)
Long precision scan					
RS17-L01	$1.127 \pm 0.012$	Upper rise	0.5 (1967)	44	$126.1 \pm 12.0$
RS17-L02	$1.136 \pm 0.010$	Peak	1.5 (1969)	42	$133.8 \pm 10.0$
RS17-L03	$1.137 \pm 0.010$	Peak	4.3 (1972)	40	$130.0 \pm 10.0$
RS17-L04	$1.150 \pm 0.010$	Peak	8.5 (1976)	36	$149.0 \pm 10.0$
RS17-L05	$1.123 \pm 0.010$	Decline	14 (1981)	30	$123.3 \pm 10.0$
RS17-L06	$1.124 \pm 0.009$	Decline	20 (1987)	24	$122.5 \pm 9.0$
RS17-L07	$1.115 \pm 0.009$	Decline	27 (1994)	17	$105.0 \pm 9.0$
RS17-L08	$1.088 \pm 0.009$	Decline	34 (2001)	10	$84.8 \pm 9.0$
RS17-L09	$1.083 \pm 0.009$	Decline	40 (2007)	5	$70.2 \pm 9.0$
Short precision scan					
RS17-S01	$1.142 \pm 0.010$	Upper rise	0.5 (1967)	44	$134.1 \pm 10.0$
RS17-S02	$1.142 \pm 0.009$	Peak	1.5 (1968)	43	$146.2 \pm 9.0$
RS17-S03	$1.155 \pm 0.009$	Peak	3.5 (1970)	41	$149.3 \pm 9.0$
RS17-S04	$1.146 \pm 0.009$	Peak	7.0 (1973)	37	$143.8 \pm 9.0$
RS17-S05	$1.128 \pm 0.009$	Decline	12 (1978)	32	$131.6 \pm 9.0$
RS17-S06	$1.146 \pm 0.009$	Peak <sup>A</sup>	18 (1984)	26	$143.9 \pm 9.0$
RS17-S07	$1.129 \pm 0.009$	Decline	25 (1991)	19	$111.1 \pm 9.0$
RS17-S08	$1.087 \pm 0.010$	Decline	34 (2001)	10	$80.7 \pm 10.0$

<sup>A</sup>Prebomb based on measured levels from scans that cross numerous dates of formation.

and decline periods (Fig. 6) and indicated the age of this fish was 43 years; younger age estimates would not provide a proper alignment with the coral  $^{14}\text{C}$  peak and decline periods.

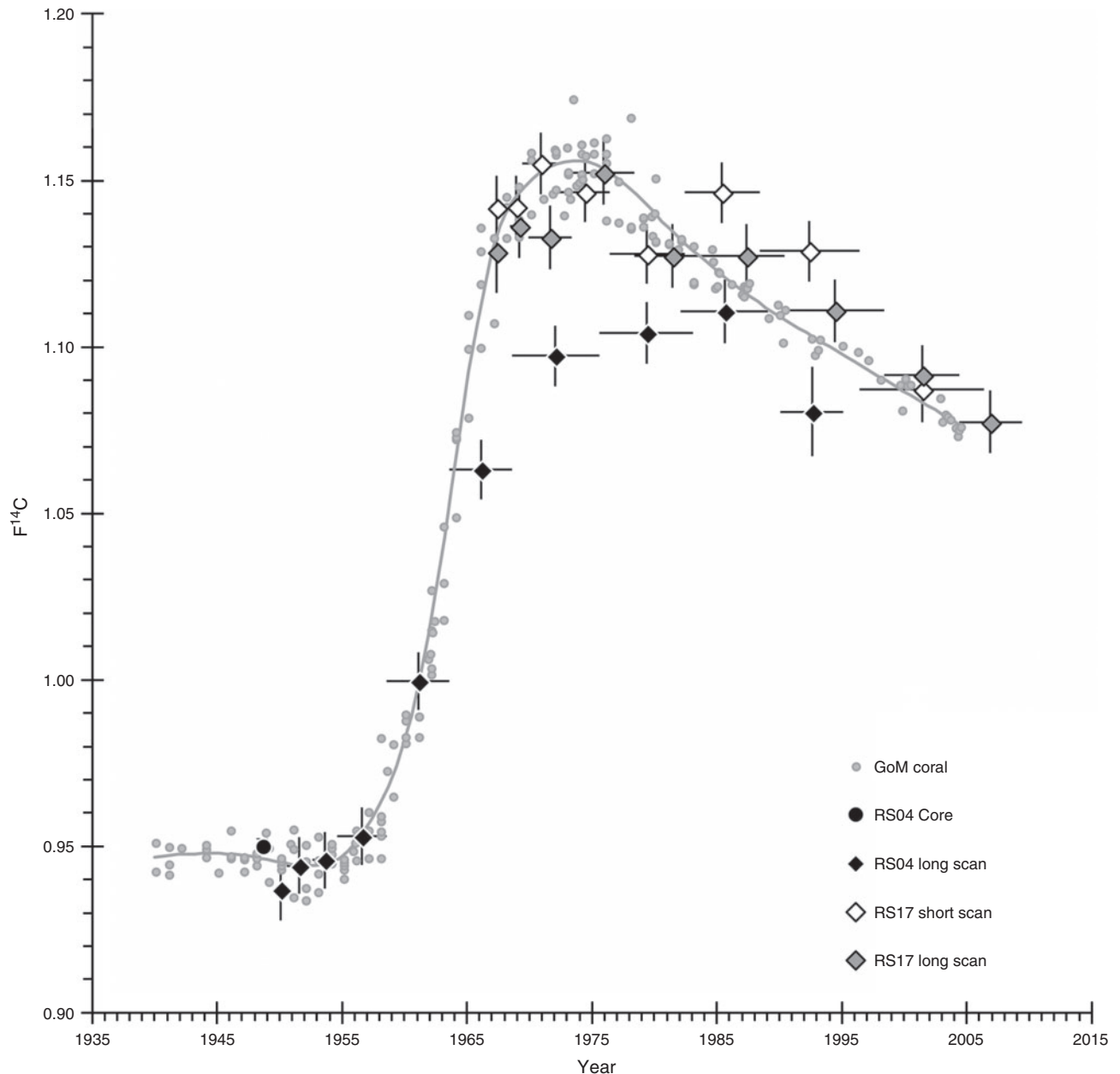
## Discussion

A 60-year longevity was strongly supported for red snapper in the Gulf of Mexico from both the conventional micromilled and LA-AMS  $^{14}\text{C}$  analyses. The radial micromilled samples placed the bomb  $^{14}\text{C}$  rise time at an estimated age of 14–17 years and well away from the earliest otolith growth for the opportunistic otolith section (RS07). In this case, the minimum age determined from a single  $^{14}\text{C}$  measurement in the otolith core was 46 years based on the last year of prebomb levels. Thus, it was necessary to rely on other studies that have validated early growth for the addition of 14 years to attain the 60-year estimate. Because biannual growth zone deposition during the early life history of red snapper (Szedlmayer and Beyer 2011) was not supported in a recent study using the post-peak decline period (Barnett *et al.* 2018), an age of 60 years for this 83-cm-TL red snapper was most probable and greater than previous estimates from growth zone counting by at least 5 years.

The utility of LA-AMS in revealing valid estimates of age by locating the rise of bomb-produced  $^{14}\text{C}$  was confirmed as feasible with two exploratory linear scans and a zigzag scan in the first otolith section (RS07). Even though the otolith section thickness was not optimal for LA-AMS, each scan revealed a complete bomb  $^{14}\text{C}$  signal and provided a time-specific location for the bomb  $^{14}\text{C}$  rise in the otolith that correlated with the

results from micromilling. Although the radial micromilling provided what may have been greater precision in determining the age at which the bomb  $^{14}\text{C}$  rise occurred, these preliminary LA-AMS results indicated a more concerted effort on thicker samples prepared for LA-AMS analyses would yield a complete bomb  $^{14}\text{C}$  signal for fish with prebomb birth years and valid estimates of age.

Valid ages for two additional adult red snapper (RS04, RS17) were determined with the development and application of the LA-AMS precision scan, a zigzag pattern within a parallelogram that had greater time specificity for the laser track over the otolith surface. The thicker otolith section preparation allowed longer scan times and replicate runs across the same transect. In each case, the full length of the scan path (core to edge) was replicated four times and, as a result, precision increased. The rectangular laser spot sample size of  $75 \times 140 \mu\text{m}$  is estimated to have sampled close to 1 year of accreted growth based on otolith increment widths (narrowest zones) and may have sampled as little as 2 years of growth simultaneously as the zigzag pattern crossed the most compressed growth zones – these narrow zone widths appeared to begin after the first 10 years of growth and progress to the otolith edge. The finalised data points correspond to the average of numerous 10-s intervals (i.e. 36 measurements across 0.6 mm of the scan) to improve counting statistics and reduce noise. For the measurements closer to the core, where growth zones are thickest, the integration time corresponded to 1–2 years of growth. Hence, the greatest precision for calculated  $^{14}\text{C}$  mean values from the precision scans was within the region of earliest and most rapid otolith growth. Translating these measurements to a date and fish age is consequently related to



**Fig. 6.** Comparison of  $^{14}\text{C}$  measurements from laser ablation–accelerator mass spectrometry (LA-AMS) precision scanning of two red snapper otoliths (Fig. 4, 5) with the regional coral bomb  $^{14}\text{C}$  reference series for the Gulf of Mexico (GoM; Andrews *et al.* 2013). The initial core measurement of specimen RS04 indicated the minimum age was >44 years, but an alignment of the LA-AMS series indicated the fish was 54 years old. The precision scan for RS04 agreed with the coral  $^{14}\text{C}$  record with regard to the time of  $^{14}\text{C}$  rise and was slightly attenuated relative to the  $^{14}\text{C}$  reference, similar to the findings for specimen RS07. Both precision scans on specimen RS017 agreed with the coral  $^{14}\text{C}$  record from the upper rise through the peak period into the decline. This alignment illustrates the potential utility of LA-AMS over single core  $^{14}\text{C}$  analyses because of the potentially ambiguous alignment with either the rise or decline portion of the reference record. Horizontal error bars for LA-AMS were derived from the age range of the scan path width for each sample block and vertical error bars were 2 s.d. of the block mean.  $F^{14}\text{C}$ , fraction modern.

the position on the coral bomb  $^{14}\text{C}$  reference curve, and temporal resolution is necessarily dependent upon the position within the otolith.

The second oldest fish of this study (RS04) was validated to be at least 44 years with the single core  $^{14}\text{C}$  measurement and was assumed to be 48 years based strictly on growth zone

counting. This estimate was increased further by the LA-AMS precision scans to 54 years. Similar to the results from RS07, the timeline established for RS04 relied on support from early otolith age reading because of prebomb  $^{14}\text{C}$  levels in the first few years of growth. In this case, the  $^{14}\text{C}$  rise time was closer to the core and between estimated ages of 8 and 12 years, which



led to an age of 54 years, greater than the highest estimate for this red snapper from various age readers (33–48 years) and similar in outcome to the underestimated age for RS07. It is most probable that this 85.3-cm-TL red snapper was 54 years old.

The third and youngest red snapper studied (RS17) provided an example of another kind of result that can be obtained from LA-AMS analyses on otoliths. This fish was estimated to be between 36 and 56 years from anonymous historical and recent age readers, and there was no initial core  $^{14}\text{C}$  value to narrow down birth year assignment. However, the age reading performed in this study provided an age estimate that was well defined at 43 years (Fig. 1). The series of LA-AMS precision scans on this specimen resulted in the best-case scenario for obtaining otolith  $^{14}\text{C}$  measurements in that both dorsal and ventral sides of the otolith section were scanned successfully. The results were similar for each side and provided two well-matched time series for both the most recent age reading (43 years) and agreement with the contour of the coral  $^{14}\text{C}$  reference. In this case, if a single core measurement had been made, the alignment of this value to the bomb  $^{14}\text{C}$  reference would have been ambiguous with potential birth years that span the peak (1967–82) and ages of 28–43 years. The advantage with the continuous  $^{14}\text{C}$  profile from LA-AMS was the elimination of the younger age scenario because of an obvious offset of the entire data series from the coral record when using younger age scenarios. Hence, the LA-AMS works well to resolve issues associated with core  $^{14}\text{C}$  values that can be ambiguous and may only be resolved with well-defined growth zone counting or the use of other otolith proxies for age, such as otolith mass (e.g. Andrews *et al.* 2013, 2016b, 2019). The age of this 88.0-cm-TL red snapper was well constrained with a valid estimate of 43 years.

A potential complication arises from the use of a continuous bomb  $^{14}\text{C}$  record within an otolith because of possible ontogenetic changes in habitat. The LA-AMS scans for the two oldest red snapper in this study provided information that is consistent with a deeper dwelling part of its life history – each fish exhibited an attenuated bomb  $^{14}\text{C}$  peak. This would be expected to some extent if the fish was living near or within the thermocline, a depth at which the well-mixed surface layer begins to give way to or mixes with deeper  $^{14}\text{C}$ -depleted waters (e.g. Grammer *et al.* 2015; Campana *et al.* 2016; Andrews *et al.* 2018b). A similar scenario was exhibited in the Gulf of Mexico from a study of yellowedge grouper (*Epinephelus flavolimbatus*), where a series of extractions from younger to older otolith material indicated there may have been a deep-water effect on otolith  $^{14}\text{C}$  because of ontogenetic habitat changes (Cook *et al.* 2009). However, even if the bomb  $^{14}\text{C}$  signal is attenuated to some extent from the mixing of surface waters with deeper waters near the thermocline, the timing of the initial  $^{14}\text{C}$  rise would not be expected to be phase lagged to a significant extent in tropical waters (likely at most 1–2 years and more related to the attenuation of the signal because of dilution, as opposed to a late arrival). This factor would be a concern for organisms that live well below the thermocline or in waters that are strongly affected by upwelling. In general, success with bomb  $^{14}\text{C}$  dating of organisms with this kind of habitat or life history is best addressed with a series of otolith core

measurements in concert with known-age reference material, such as juvenile otoliths, to provide some ground truthing. Examples of a series of successes in this regard can be illustrated with a long series of studies on rockfishes (Family Sebastidae) of the north-eastern Pacific Ocean (e.g. Kerr *et al.* 2004; Piner *et al.* 2005; Andrews *et al.* 2007; Kestelle *et al.* 2008).

Otoliths of red snapper were selected for this pioneering study because the age estimates led to birth years in the prebomb or bomb  $^{14}\text{C}$  rise periods and the Gulf of Mexico provides the broadest, regionally consistent coral  $^{14}\text{C}$  reference records (Andrews *et al.* 2013; Barnett *et al.* 2018). Because the selected red snapper otoliths could span all or most of the bomb-produced  $^{14}\text{C}$  signal of this marine environment, it was hypothesised that an entire bomb  $^{14}\text{C}$  signal could be traced with LA-AMS based on previous results from other carbonates (Welte *et al.* 2016). A factor in selecting red snapper for this study was that the otoliths are massive and can reach or exceed 4 g. Hence, the high mass was a best-case scenario for a feasibility study using the new LA-AMS technology because it provided the greatest amount of carbonate through this 50- to 60-year period of formation that exhibits  $\Delta^{14}\text{C}$  changes in the marine environment on the order of 100–200‰ (Grottoli and Eakin 2007). The first specimen (RS07) was opportunistic because it had been prepared as a thin section for age reading and was only 0.3 mm thick. From previous analyses of another marine carbonate (black-lip pearl oyster *Pinctada margaritifera*; Welte *et al.* 2016), it was known that the laser could easily pass through this thin section and may be influenced by contamination from  $^{14}\text{C}$ -depleted mounting medium. In some circumstances with the black-lip pearl oyster, it was clear from the LA-AMS signal that the carbonate  $^{14}\text{C}$  levels were being diluted with sources that were well below what was expected from the marine environment. This was likely the reason for the low levels observed on one side of the RS04 otolith that led to it being dismissed from further analysis.

This research addresses a frontier in bomb  $^{14}\text{C}$  dating by providing avenues that may lead to the determination of age for marine organisms that have lived through all or part of the bomb  $^{14}\text{C}$  period when few other options exist. An example of a circumstance where this approach could be useful is with the otoliths of blue marlin (*Makaira nigricans*). A recent study revealed that the age of a large adult blue marlin (3.7-m fork length and 565 kg) was 20 years using a series of deductions from different otolith sample types (Andrews *et al.* 2018a). The initial measurement of the otolith core revealed the fish could have been born during either the bomb  $^{14}\text{C}$  rise or decline period, a discrepancy of up to several decades. It was the use of the whole lapillus and other otolith samples where a series of deductions was made to refine the age of this fish to 20 years. Assuming LA-AMS technology continues to improve well enough to deal with the very small otoliths from this species (<10 mg for full-sized adults), the whole otolith could be scanned from core to edge in a single continuous measurement within 30 min, thereby alleviating the need to make other  $^{14}\text{C}$  assays.

### Conflicts of interest

The authors declare that they have no conflicts of interest.

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