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Catch-at-age data for southern bluefin tuna in the New Zealand long line fishery, 2015–2018 seasons

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A. Sutrovic,
K. Krusic-Golub

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This report describes the collection of age data that adds to the long-term ageing data of southern bluefin tuna (*Thunnus maccoyii*) caught in the New Zealand longline fishery. Southern Bluefin Tuna (STN) are managed by the Commission for the Conservation for Southern Bluefin Tuna (CCSBT) and form a valuable fishery in New Zealand waters, taken by domestic and chartered longline vessels as well as recreational fishermen. Cohort strength had previously been determined using cohort slicing to convert catch-at-length to catch-at-age. However, the CCSBT has recognised that it is preferable to use direct age estimation from otoliths. Sub-samples of otoliths collected during each fishing season since 2000/01 have been aged under various Ministry for Primary Industries projects and used to determine catch-at-age for each fishing year.

The STN otoliths selected for ageing were sub-samples that came from the routine collection of otoliths by observers aboard New Zealand domestic and foreign charter vessels, as well as otoliths collected from reproductive and recreational samples between 10/05/2015 and 06/07/2018. Ageing protocols developed at the “Direct Age Estimation Workshop of the CCSBT” held 11–14 June 2002, in Queenscliff, Australia were followed to provide estimates of age. Due to operational reasons, there was a shortfall in otolith numbers in the 2017/18 year. For this reason, otoliths collected from reproductive samples from previous trips were used to compensate. This resulted in a total of 361 age estimates being provided to the Ministry for Primary Industries. Due to some samples received being from the 2015 fishing year, these data have been combined with previously aged data from the 2014/15 report, previously submitted to Ministry for Primary Industries (project SEA2015/19). Age estimates ranged from 2 to 27 years. Repeat readings were performed and results were well within the predetermined acceptable precision limits, indicating a low level of error within the readings.

Proportions-at-age in the catch were estimated by applying the standard age length-key method to the age and size frequency distributions obtained from sampling the catch. The results of the projects showed that the age composition for the 2015 (2014/15) and 2016 (2015/16) seasons were dominated by age classes 6 to 10, whereas the 2017 (2016/17) season was dominated by age classes 4 to 8. The 2018 (2017/18) season had a small sample size, and no data were supplied for this analysis. As in previous years (2005 to present) there were very few samples less than 4 years of age in the catch.

1. INTRODUCTION

Southern Bluefin Tuna (STN) are managed by the Commission for the Conservation for Southern Bluefin Tuna (CCSBT) and form a valuable fishery in New Zealand waters targeted by domestic and chartered longline vessels.

The assessments and indicators presented at the 2004 Stock Assessment Group (SAG) agreed that there was at least one year of markedly low recruitment amongst the 1999–2001 year-classes. These support the recruitment concerns outlined in the 2003 SAG report. Moreover, the lack of small fish in the longline fisheries and other indicators raised concern that there may have been several years of markedly lower recruitment among those year classes. Concerns were also raised regarding possible reductions in spawning stock size. Therefore, accurately estimating the cohort strength from year to year is critical for assessing the status of the stocks and future abundance levels.

Given the low current biomass levels of the stock, information on recent year class strength is particularly important. Previously, cohort strength has been determined using cohort slicing to convert catch-at-length to catch-at-age, but it has been increasingly realised that it is more reliable to use direct age estimation.

Sub-samples of otoliths collected from 2001 (2000/01) through to 2014 (2013/14) have previously been aged under MPI (formally MFish) projects IFA2004/03, STN2006/01, STN2007/01, STN2009/01, STN2011/01, and SEA2015–19 to determine catch-at-age for these years. The results of this work were found to be useful for providing indicators of recruitment strength.

The sample size for age estimation for each fishing year is based on estimates described by Morton & Bravington (2003). They concluded that 100–200 otolith age readings per year would be sufficient for the Australian surface fishery, 200 for the Japanese longline fishery, and 500 for the Indonesian fishery. At the beginning of this work (IFA2004/03), 200 fish per year were aged and, although the patterns in the data were generally consistent across years, it was apparent that an increased number of otoliths would likely reduce the uncertainty in the proportions of younger ages taken in the catch.

Based on this, a minimum of 250 otoliths were required to be aged from otolith collections from the New Zealand fishery each year. The otoliths sampled for this project were collected during the 2014/15, 2015/16, 2016/17, and 2017/18 fishing seasons, herein referred to as the 2015, 2016, 2017, and 2018 seasons, respectively. The report of the "Direct Age Estimation Workshop of the CCSBT" held 11–14 June 2002, in Queenscliff, Australia forms the basis for the protocols employed in this project. These protocols include double blind readings of each otolith and the determination of an agreed final age.

The main output from this project was to estimate the catch-at-age of STN sampled from vessels fishing in New Zealand waters from 2015 to 2018 and generate individual age data for the CCSBT age database. As stipulated by the CCSBT, it is a requirement that each otolith be assigned an individual identification number in which the data—year, month, latitude and longitude of capture, fish length, otolith age estimate, and any other relevant comments—can be linked to.

This report describes the collection and analysis of age data provided from project STN2016-01 and is a continuation of work started under IFA2004/03 to monitor the ages of STN taken in the New Zealand fishery.

1.1 Objectives

- Age 250 STN otoliths collected by scientific observers aboard vessels fishing in New Zealand waters during the 2015 (2014/15), 2016 (2015/16) and 2018 (2017/18) fishing seasons.
- Provide marginal increment measurements and an image of each otolith section.
- Prepare a report describing collection and analysis of age data from project STN2016-01.

2. METHODS

Appendix 1 provides a full description of the methods used in this project. Full methods with detailed diagrams have also been presented previously in document CCSBT-ESC/0509/12, "Catch at age of Southern Bluefin Tuna in the New Zealand longline fishery, 2001–2004", prepared for the CCSBT SAG/SC meetings in Taipei, Taiwan 28 August to 8 September 2005.

2.1 Otolith collection

Observers aboard New Zealand domestic vessels and foreign charter vessels routinely obtained length estimates and collected otoliths from STN. Otoliths were stored within marked envelopes by NIWA. Data detailing the trip number, fish number, fish length, date of capture, area of capture, and sex for each sample were obtained from NIWA. These data were supplied in electronic form, with reference to trip and fish number as the unique identifiers. Fish Ageing Services (FAS) has an internal numbering system which groups samples into batches based usually on the location, date, or area of capture. For the STN sample registration, a separate batch was assigned to otoliths from each trip number. Table 1 details the FAS batch numbers, the corresponding trip numbers, the number of otolith available from that trip, and the various capture details relevant to this project.

Table 1: Batch detail of otoliths registered over the duration of this project. Fisheries Management Area (FMA).

Batch	Trip	N	Fishing year	Origin	FMA
098	4 656	40	2015/16	SOP	CHA
099	4 683	61	2015/16	SOP	CHA
105	4 989	19	2016/17	SOP	CHA
106	5 043	48	2016/17	SOP	CHA
107	5 050	29	2016/17	SOP	AKE
108	5 008	22	2016/17	SOP	AKE
109	5 017	1	2016/17	SOP	CEE
114	57 001–57 008	20	2017/18	REC	BPLE
115	Non-Survey	12	2017/18	BWM	BPLE
116	5 310	5	2017/18	SOP	CHA
118*	4 380	47	2014/15	SOP	CHA
119*	4 379	84	2014/15	SOP	CHA

*131 additional samples were available from 2014/15 as part of a reproductive study.
SOP = Scientific Observer Programme – Catch Sampling
REC = Recreational
BWM = Non-survey collected at weigh station

2.2 Sample selection

The targeted number for otolith processing each year is 270. More samples are selected for preparation than are required for age estimation to account for the small proportion that are rejected for ageing, either because the otolith was too damaged for preparation or the otolith section was too difficult to interpret. The number of "unreadable" otoliths is typically less than 5% for STN. In previous projects the number of otoliths collected each year has exceeded the number required for processing. To obtain an adequate sample for determination of catch-at-age, it was necessary to select a random sub-sample from the total otolith sample available.

Due to operational reasons, for each of the three fishing years analysed in this study, the number of otoliths was less than targeted 250. Consequently, no sub-sampling was necessary, and all available otoliths were selected for preparation. To increase the overall number of otoliths examined, a further 32 otoliths sampled from recreationally caught STN along with a set of 131 otoliths from samples collected from 2015 as part of a larger reproductive study. This increased the total number of samples for the project from 225 to 388. Because additional samples from 2015 were provided for ageing during this project, to be sure that the total 2015 otolith sample selected for ageing was still representative, the original 2015 comparison was updated. Results are shown in Appendix 2 and indicated that the length frequency of the otolith samples aged was still similar to that of both the length frequency of the total otolith samples collected and the length frequency of the catch.

One otolith from each pair was weighed to the nearest milligram on an electric balance. Only undamaged otoliths were considered for weighing. Otolith weight is a useful diagnostic tool in assessing potential errors in age estimates and for examining patterns of otolith growth. Morison et al. (1998) found that otolith weight has a strong relationship with fish size and age and in medium to long-lived species, the relationship of otolith weight against estimated age may show an increased slope if the ages have been underestimated. Large variation about the relationship may indicate a lack of precision in the estimates. Also, any outlying data points in the otolith weight/age and the fish length/otolith weight relationship may indicate an incorrect assignment of age or an issue with the length measurements.

2.3 Preparation and ageing

Otoliths were prepared and aged following protocols outlined in the report of the “Direct Age Estimation Workshop of the CCSBT” held 11–14 June 2002, in Queenscliff, Australia. Opaque zones were counted along a transect starting at the primordium and running out through the ventral arm to the otolith edge where possible; otherwise, opaque zones were counted along a transect starting at the primordium and running out through the dorsal arm to the otolith edge (Figure 1). For each otolith section aged, a single image was taken, the marginal increment was measured, and the otolith edge was classified as wide translucent (WT), narrow translucent (NT), or opaque (O).

Figure 1: Southern Bluefin Tuna otolith section indicating the count path (black line) and the opaque zones counted (white arrows) for determining age.

2.4 Birthdate

Assigning age to STN from zone counts is problematic due to the theoretical birthdate being January 1, because opaque zones are thought to be formed during winter (April to August). The sampling of the New Zealand fishing seasons occurs through the middle of the year (May to August), and otoliths sampled from this period may exhibit both opaque and translucent margins. Using the number of opaque zones as an estimate of age can be misleading. For example, STN that are biologically the same age can differ by 1 year depending if the opaque increment has formed on the otolith edge. To adjust for this, zone counts can be converted to age estimates by a theoretical birthdate adjustment. This can, however, lead to difficulties when comparing age estimates and biological parameters from samples caught in the middle of the year with those caught at the end of the year.

To be consistent with previous approaches (Krusic-Golub 2005, Krusic-Golub 2012) the last opaque zone formed prior to the edge of the otolith was only counted when the reader could see translucent otolith material between this opaque zone and the edge. The edge type was then recorded as wide or narrow depending on the amount of translucent material on the marginal edge relative to the thickness of the previously completed translucent zone. If all otoliths are aged consistently to this protocol, then zone counts can be post-adjusted according to requirements of the data users. In addition, it was suggested that the marginal increment of each otolith should be measured.

2.5 Marginal increment measurement

The percentage completion of the marginal increment formation was examined by calculating the mean index of completion (*C*). Indices of completion were calculated using the equation:

$$C = Wn/Wn\text{-}1$$

Where *Wn* is the width of the marginal increment (distance from the start of the last opaque zone to the marginal edge) and *Wn-1* is the width of the previously completed annulus (the distance from the start of the second most outer opaque zone to the last opaque zone).

2.6 Data analysis

The relationship between biological attributes can be used to determine whether any inconsistencies or outliers are present within the data. Age estimates were combined with fish length and otolith mass data and checked for outliers. The additional 2014/15 samples were combined with those ages produced during the last project (SEA2015–19) and the resulting tables and figures for that year were updated and included in this report for completeness.

Length frequency distributions were produced for each of the fishing seasons. A von Bertalanffy growth curve was fitted to the combined length-at-age data using the non-linear least squares method. The growth equation was determined using the equation:

$$L(t) = L_\infty\,(1\text{-}e^{-k(t-to)})$$

where L_∞ indicates the mean asymptotic fork length (FL, cm), k represents the growth constant, and t_o is the theoretical age at length zero.

Summaries were produced for the number of fish at each age and length, and number of fish at each length for each age (age-length key), derived from Excel spreadsheets. The age composition of the sampled catch was estimated using the age-length key (ALK), and then applied to the length-frequency data for each corresponding fishing year as follows:

$A_t=\sum_x(L_x p_{tx})$ where

A_t = the estimated number of fish of age t in the length-frequency sample, L_x = the number of fish of length x in the length-frequency sample, and P_{tx} = the proportion of aged fish of length x which were age t.

2.7 Precision estimates

All otoliths were read twice without reference to anything other than the otolith ID. Re-reading of the selected otoliths by the same reader/s provides a measure of precision within the readings. The purpose of the re-reading is to provide an indication of error associated with the estimates, not an agreed age. The readings were compared, and the Index of Average Percent Error (IAPE) was calculated. The distribution of the differences between repeat readings was also inspected as another indicator of ageing errors, and of any potential bias between readings. To provide a measure of intra-reader variability, for each fishing season a 10% sub-sample of otolith sections were read by a secondary reader experienced in reading STN otoliths.

After reading each otolith twice, a final agreed age was recorded. If the two initial readings differed the otolith sample was re-read a third time and a final age assigned. The final age reading was completed with the knowledge of the two previous readings and date of capture.

3. RESULTS

3.1 Otolith data

Numbers of otoliths per year available for age estimation year are shown in Table 2. The length distributions of the sampled catch and the length distributions of the otolith sub-sample for each fishing season are presented in Figure 2.

Table2: Numbers of Southern Bluefin Tuna otoliths collected, prepared, and aged from each fishing season for the duration of this project.

Fishing year	Samples	Prepared	Aged
2015	131	131	126*
2016	101	101	89
2017	119	119	108
2018	37	37	37

*Some figures and tables may show a number greater than 126 for aged samples because they have been updated with previously aged 2014/15 samples from the previous SEA2015-19 report (Krusic-Golub 2017).

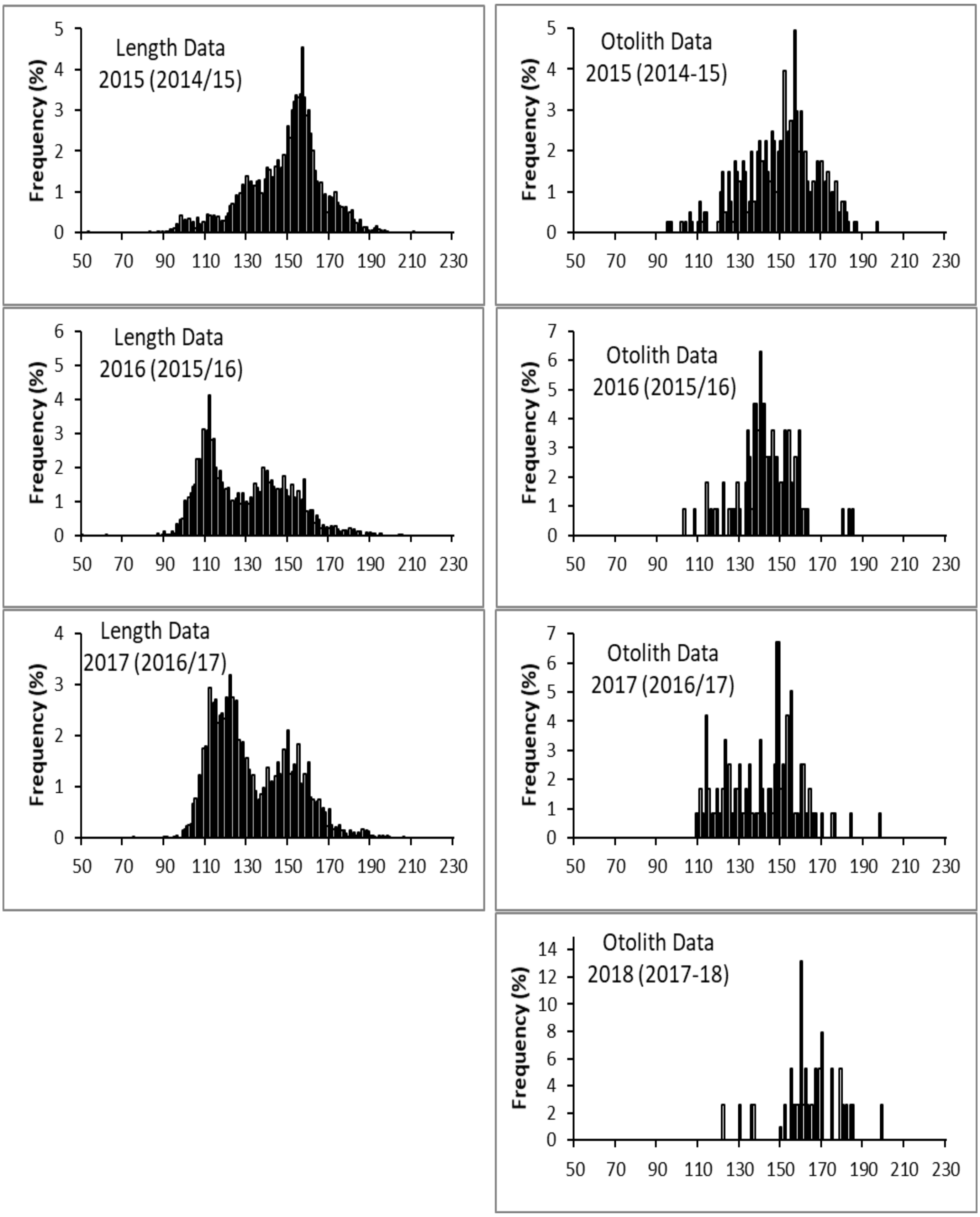

Figure 2: Length frequency sampled for the 2015, 2016, and 2017 fishing seasons from the length at catch. The 2018 length data were not available at the time of reporting. Note: the 2015 season otolith data include previously aged data from SEA2015/19 (Krusic-Golub 2017).

3.2 Age estimation and precision

Age was estimated for 360 of the 388 STN otoliths available. Seven percent (28 samples) could not be read because the otoliths were either too broken to prepare or the otolith increment structure within the sectioned otolith was too difficult to interpret. For the samples aged during this project, age ranged from 2 to 27 years. The age-otolith weight relationships for each sampling year along with the age-fish length relationship of the combined samples (including previously aged 2014/15 samples) are shown in Appendix 3 and show reasonably good fits for all years. Comparison of the first and second readings showed that 48% of the age estimates agreed and 90% were within one year (Appendix 4). The index of average percentage error (IAPE) for the primary reader was 4.7 %. The IAPE between the primary and secondary reader for a selected subset of otoliths (n=49) was 3.4 %.

Growth parameters estimated for the three fishing seasons (2015, 2016, and 2017) are presented collectively in Table 3 as STN2016-01. The growth parameters for 2018 were not estimated due to the small sample size; no data were supplied for analysis. The growth curve for all years combined is presented and overlaid with the age-at-length data in Figure 3.

Table 3: Von Bertalanaffy growth parameters estimated from each STN catch-at-age project (STN2016-01, SEA2015/19, STN2011/01, STN2009/01, STN2007/01, STN2006/01, and IFA2003/04).

Project	L∞	K	to
STN2016-01	182.64	0.16	-2.32
SEA2015-19	180.35	0.18	-2.00
STN2011/01	178.19	0.20	-1.30
STN2009/01	185.26	0.17	-.2.48
STN2007/01	179.27	0.24	-0.34
STN2006/01	187.96	0.12	-5.65
IFA2003/04	183.50	0.16	-2.52

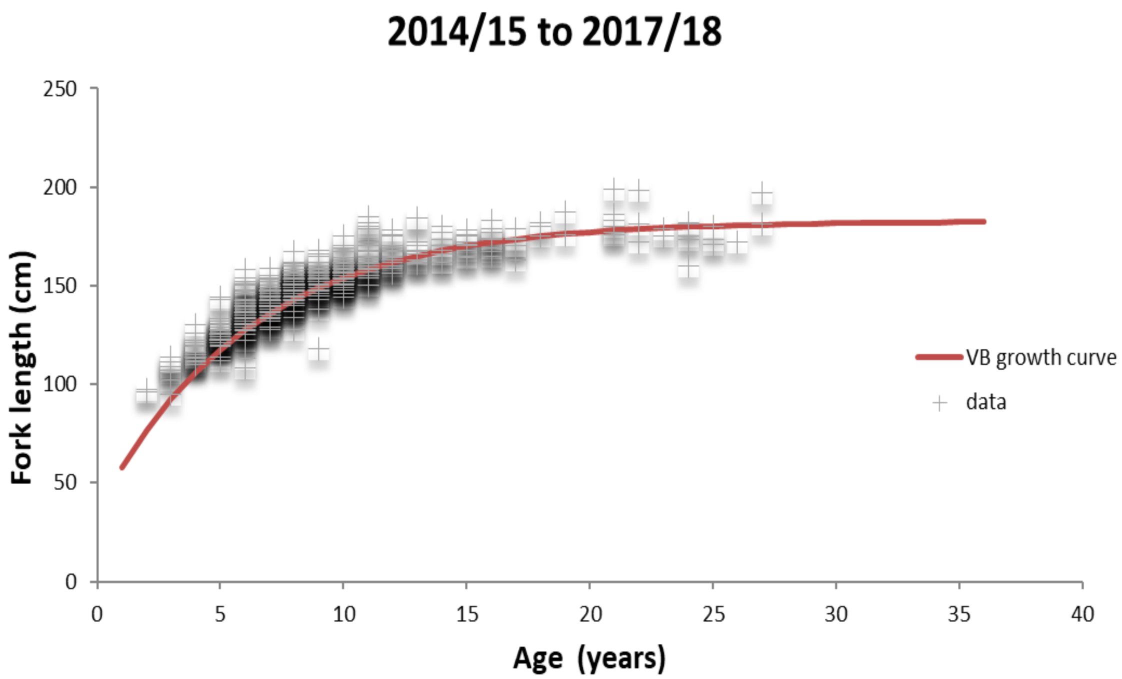

Figure 3: Length-at-age for STN for project STN2016-01 – years 2015 to 2018 (n=613 – includes previously 14/15 aged samples from SEA2015–19).

3.3 Proportions-at-age

The ALK for the direct age estimates for 2015, 2016, 2017, and 2018 are given in Table 4 to Table 7, and the proportions-at-age estimated by the ALK methods for 2015, 2016, and 2017 are given in Table 8 and illustrated in Figure 4. The proportions-at-age from the past 3 fishing seasons have also been included for comparison. Proportions-at-age were not estimated by the ALK methods for 2018 because no data were supplied.

The length-adjusted numbers at age for 2015, 2016, and 2017 are shown in Figure 5 and the results for all years are summarised as follows:

- Ages ranged from 2 to 27 years.
- The age composition was dominated by age classes 4 to 10 years.
- Low number of age estimates less than 4 years, and more than 11 years were recorded.

Table 4: Age-length-key for direct age estimates – fishing season 2015.

Year (2015)	2014/15

	Age																				
Fork length (cm)	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20+	Total
80																						0
85																						0
90																						0
95			2	1																		3
100				1																		1
105				2																		2
110				2	4	1	1															8
115																						0
120					5	8	2															15
125					1	11	7	2														21
130					1	7	13	2														23
135							15	10														25
140							7	13	13		1											34
145							2	16	10	4	1											33
150							2	5	15	13	11	2										48
155								1	12	17	21	7	1									59
160									3	2	8	9	6	1	2	1		1			1	34
165										3	1	3	3	3	3	2	4					22
170												2	1	1	3	3	4	4			5	23
175													2		1	3	2	1	1	1	5	16
180															1				1		7	9
185																				1	1	2
190																						0
195																					1	1
200																						0
205																						0
210																						0
Total	0	0	2	6	11	27	49	49	53	39	43	23	13	5	10	9	10	6	2	2	20	379

Note: This table includes 254 age estimates from SEA2015-19.

Table 1: Age-length-key for direct age estimates – fishing season 2016.

Year (2016)	2015/16

	Age																				
Fork length (cm)	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20+	Total
80																						
85																						
90																						
95																						
100																						
105							1															1
110						1																1
115					1	1																2
120						1	1															2
125						2	1	1	1													5
130							2	2	1													5
135							4	6	2	1												13
140						2	4	10	5	1												22
145								4	3	3	2											12
150							2	1	2	3	2											10
155										1	6	3										10
160											3			1								4
165																						
170																						
175																						
180																	1		1			2
185																						
190																						
195																						
200																						
205																						
210																						
Total	0	0	0	0	1	7	15	24	14	9	13	3	0	1	0	0	1	0	1	0	0	89

Table 2: Age-length-key for direct age estimates – fishing season 2017.

Year (2017)	2016/17

	Age																				
Fork length (cm)	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20+	Total
80																						0
85																						0
90																						0
95																						0
100																						0
105				1																		1
110				2	4																	6
115					3					1												4
120					1	7																8
125						7	1															8
130						2	4	1														7
135							4	2														6
140							4	4	1													9
145							2	7	9	4												22
150							2		4	4	3	1										14
155								2	2	2	2											8
160									5	1		1	1	1								9
165									1			1										2
170																						0
175													1			1						2
180														1								1
185																						0
190																						0
195																					1	1
200																						0
205																						0
210																						0
Total	0	0	0	3	8	16	17	16	22	12	5	3	2	2	0	1	0	0	0	0	1	108

Table 3: Age-length-key for direct age estimates – fishing season 2018.

Year (2018)	2017/18

	Age																					
Fork length (cm)	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20+	Total
80																						0
85																						0
90																						0
95																						0
100																						0
105																						0
110																						0
115																						0
120						1																1
125																						0
130						1																1
135								2														2
140																						0
145																						0
150									1		1											2
155							1		2	1		1	1									6
160									1	3	3	1	1									9
165											2		2	1								5
170											1		1	1								3
175											2	1					1					4
180												2		1								3
185												1										1
190																						0
195																					1	1
200																						0
205																						0
210																						0
Total	0	0	0	0	0	2	1	2	4	4	9	6	5	3	0	0	1	0	0	0	1	38

Table 4: Proportions-at-age for the three fishing seasons (2015 –2017) using the age-length-key method. No data were supplied for the 2018 (2017/18) season because of low sample numbers.

	Age										
Season	0	1	2	3	4	5	6	7	8	9	10
2015 (2014/15)			0.0039	0.0149	0.0158	0.0553	0.1129	0.1181	0.1604	0.1206	0.1345
2016 (2015/16)					0.0168	0.0693	0.1035	0.1026	0.0653	0.0629	0.0795
2017 (2016/17)				0.0349	0.1337	0.2007	0.0923	0.0682	0.1107	0.0679	0.0258

	Age									
	11	12	13	14	15	16	17	18	19	20+
2015 (2014/15)	0.0722	0.0301	0.0085	0.0196	0.0203	0.0054	0.0204		0.0015	0.0190
2016 (2015/16)	0.0070		0.0035			0.0008		0.0020		
2017 (2016/17)	0.0177	0.0081	0.0036		.0015					.0005

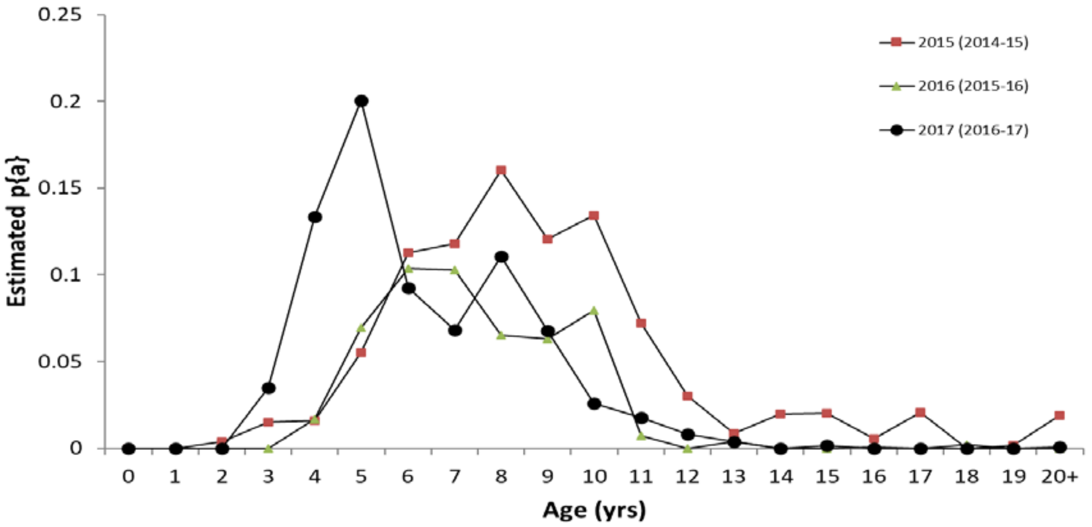

Figure 4: Proportions-at-age for the 2015, 2016, and 2017 fishing seasons, estimated using the Age-length-key method. No data were supplied for the 2018 season because of low sample numbers. The 2015 data include samples aged under project SEA2015-19.

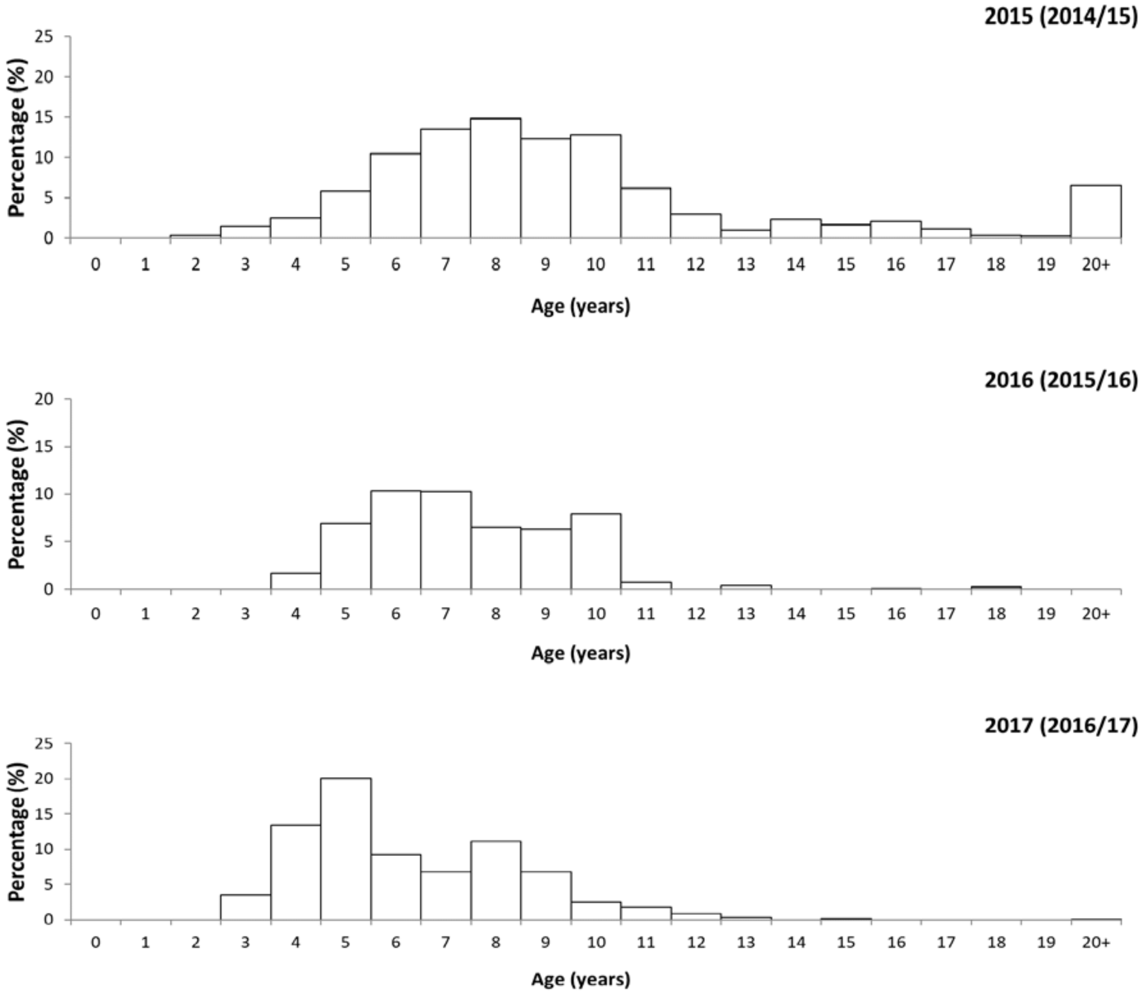

Figure 5: Adjusted age composition weighted by length frequency of the catch for fishing seasons 2015 (2014/15), 2016 (2015/16), and 2017 (2016/17). Samples from the 2018 season were not included due to small sample size and no data were supplied for the analysis. The 2015 season data include SEA2015/19 data.

4. DISCUSSION

The estimates of precision between first and second readings from the primary reader and readings between the primary and secondary reader were within the acceptable limits suggested by Morison et al. (1998); these authors suggest that levels of APE less than 5% are considered acceptable for age estimation studies. The low levels of error estimated in this study suggest consistent interpretation between readings and readers.

The estimated proportions-at-age for years 2015 (revised), 2016, and 2017 were similar to previous fishing seasons in that there were few samples estimated to be less than 4 years of age (CCSBT-ESC/0509/12) and majority of the age composition from 2009 to 2014 was dominated by four or five year age classes (Appendix 5). The proportions-at-age were not estimated for 2018 due to the low sample size.

Results were presented using unadjusted age (zone count=age) and no adjustment for birthdate and edge type was considered. The issue of edge adjustment and comparison with data from end of year catches (Indonesian, Japanese, and Australian surface fishery samples) is a complex one. The estimated proportions of catch-at-age were calculated from unadjusted zone counts. For 79% of samples aged from 2014/15, 70% of samples aged in 2015/16, 94% of samples aged in 2016/17, and 92% of samples aged in 2017/18, the marginal edge of the otolith section was classified as wide. Thought needs to be given to the effect that this classification may have on comparisons between the age composition of mid-year and end-year catches if they are to be compared.

Although it was suggested that no adjustment to zone count was required when estimating proportions for catch-at-age, adjustments will be necessary if growth estimates from this study are to be compared with growth estimates from other sources (i.e., end of year samples). However, because STN do not grow at a consistent rate throughout the year, if zone counts are universally adjusted to either wide or narrow (-1 or +1), there will also be a bias. Growth in juvenile STN has been shown to peak during January to May (J. Farley, CSIRO, pers. comm.). Therefore, if all zone counts are adjusted for a narrow edge (as reported in IFA2004/03), the mean length for a given age class for mid-year samples will be higher than for the end of year samples. This difference may incorrectly infer that STN from New Zealand grow more quickly than from other areas. To compare growth, a method such as randomly allocating a wide or narrow edge to each sample may be necessary. Because this was not part of the project scope, this is only a recommendation and accordingly no results have been provided using this method.

The project was able to meet all the objectives of STN2016-01. A total of 360 age estimates were produced from STN otoliths collected by observers working aboard longline vessels fishing in New Zealand waters and otoliths sampled from the recreational survey programme. By applying these data to the size frequency distributions obtained from sampling the catch, proportions-at-age for the 2015, 2016, and 2017 seasons were estimated. Proportions were not estimated for the 2018 season due to the low number of otoliths available for that year. The continued direct ageing data provides important information for future stock management.

5. ACKNOWLEDGMENTS

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APPENDIX 1: Detailed methods

Otolith mass

Otolith weight is a useful diagnostic tool in assessing potential errors in age estimates and for examining patterns of otolith growth. Otolith weight has a strong relationship with fish size and age. In long-lived species, the relationship of otolith weight against estimated age may show an increased slope if the ages have been underestimated. Large variation about the relationship may indicate a lack of precision in the estimates. Also, any single outlier in the relationship may indicate incorrect assignment of age and/or incorrect length and otolith weight measurements.

All undamaged otoliths were weighed to the nearest 0.001 g on an electronic balance.

Preparation of otoliths

Otoliths were prepared and aged according to standard FAS procedures and protocols. The FAS procedures are modified from Morison et al. (1998).

Otoliths were embedded, in rows of five, within blocks of clear casting resin ensuring that the primordium of each otolith is inline. Four sections, approximately 300 µm thick, were cut through their centres with a modified high-speed gem-cutting saw with a 250 µm thick diamond impregnated blade. Sections from each sample were cleaned, dried, and mounted on glass microscope slides (50 x 76 mm) with resin. Sections were then covered with further resin and two glass coverslips (22 x 60 mm) were placed over the top of the resin. Prepared slides were placed in an oven at 40 °C and allowed to dry overnight.

Counts and measurements

Age estimation assumes:

- Increments counted were formed on an annual basis.
- One translucent zone and subsequent opaque zone represents one annual growth increment.

Sections were examined using transmitted light under a Leica M80 Zoom Stereomicroscope at 25x magnification. Higher magnification was sometimes required for the examination of the fine growth increments near the otolith edge from larger, presumably older fish. Each section of the otolith was inspected, and the section with the clearest incremental structure was chosen for ageing. This was usually, but not necessarily the section closest to the primordium.

A customised image analysis system is used to age sectioned and whole otoliths. This system counts and measures manually marked increments and collects an image from each sample aged.

A CCD digital camera is mounted onto the stereomicroscope (Leica M80) and a live image is displayed on the monitor. Using the image analysis system, a transect is drawn on the otolith image from the primordium to the edge of the sample. The positions of the opaque zones along this transect and the otolith edge are marked with a screen cursor. Additional structures such as sub-annual zones or transition points can also be marked. The numbers of zones marked and the measurements from the primordium to each subsequent mark along the transect are exported to a Microsoft database.

In addition, the otolith image is automatically captured and exported, along with the x and y coordinates of the marked zones, into an image database. Images are captured in the Joint Photographic Experts Group (JPG) format.

To avoid the potential for biasing age estimates, all counts were initially made without knowledge of fish size, sex, location, or date of capture. Once ageing of all otoliths were completed, the ageing data were combined with biological data (fish length, date of capture) and otolith weight for subsequent analyses.

Precision of age estimates

Repeated readings of the same otoliths provide measures of intra-reader and inter-reader variability. Repeat readings do not validate the assigned ages but provide an indication of magnitude of the error to be expected with a set of age estimates, due to variation in interpretation of an otolith. Beamish & Fournier (1981) have developed an index of average percent error (IAPE), which has become a common method for quantifying this variation. The IAPE is calculated as:

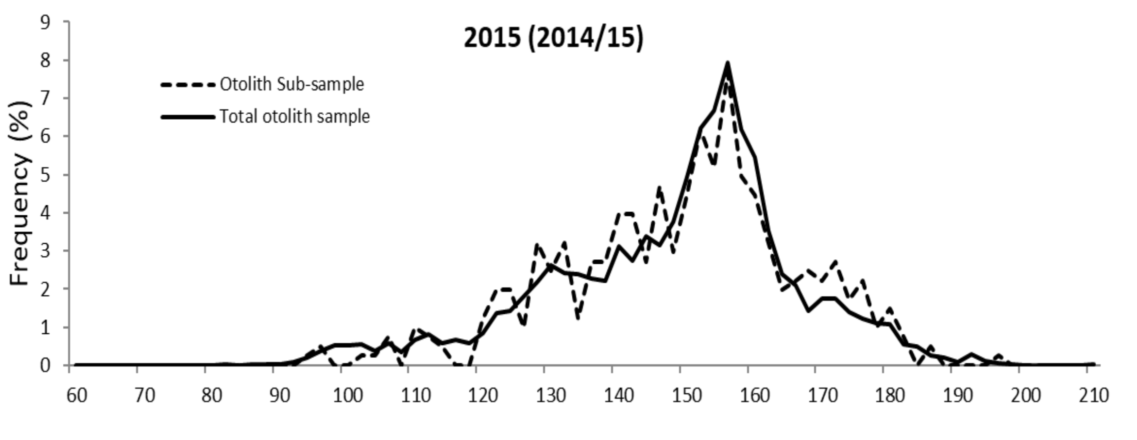
$$IAPE = \frac{100}{N} \sum_{j=1}^{N} \left[\frac{1}{R} \sum_{i=1}^{R} \frac{|X_{ij} - X_j|}{X_j} \right]$$

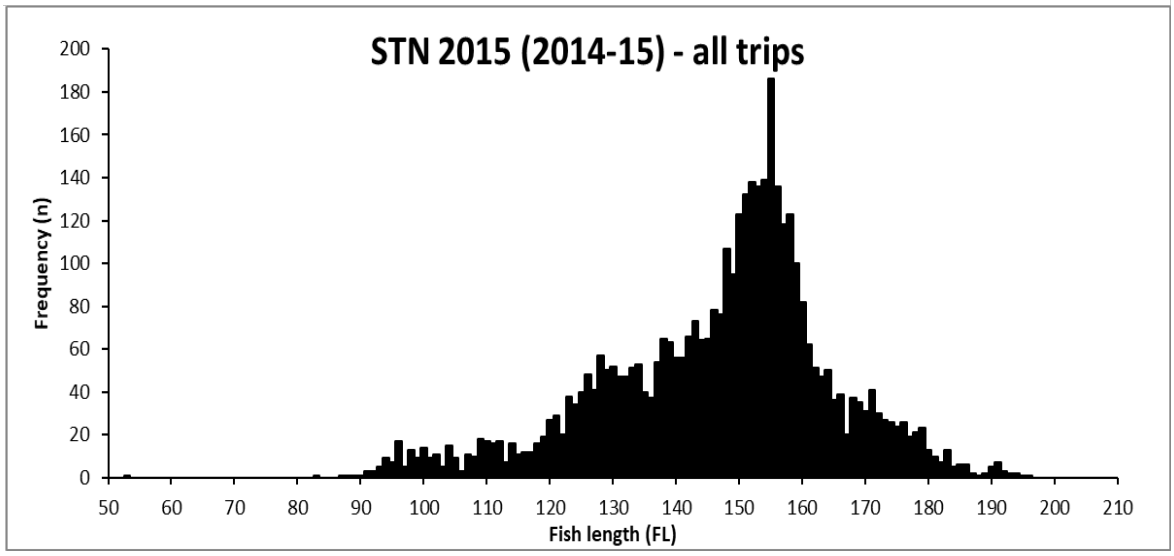
where N is the number of fish aged, R is the number of times the fish are aged, X_{ij} is the *i*th determination for the *j*th fish, and X_j is the average estimated age of the *j*th fish. The index has the property that differences in age estimates for younger fish will contribute more to the final value than will the same absolute error for older fish (Anderson et al. 1992).

All otoliths were read twice without reference to anything other than the otolith ID and an IAPE was calculated. The distribution of the differences between repeat readings was also inspected as another indicator of ageing errors, and of any bias between readings. Re-reading of the selected otoliths by the same reader/s provides a measure of precision within the readings. The purpose of the re-reading is to provide an indication of error associated with the estimates, not an agreed age.

After reading each otolith twice, a final agreed age was given. If the two readings differed the otolith sample was re-read a third time and a final age assigned.

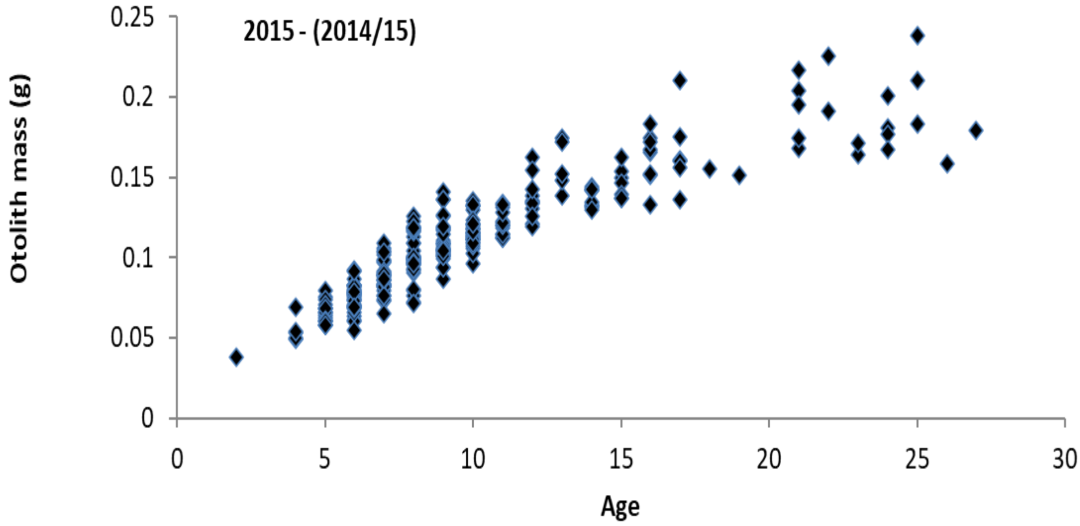
APPENDIX 2: Comparison of length for sub-sampled otoliths and total – 2014/15 samples.

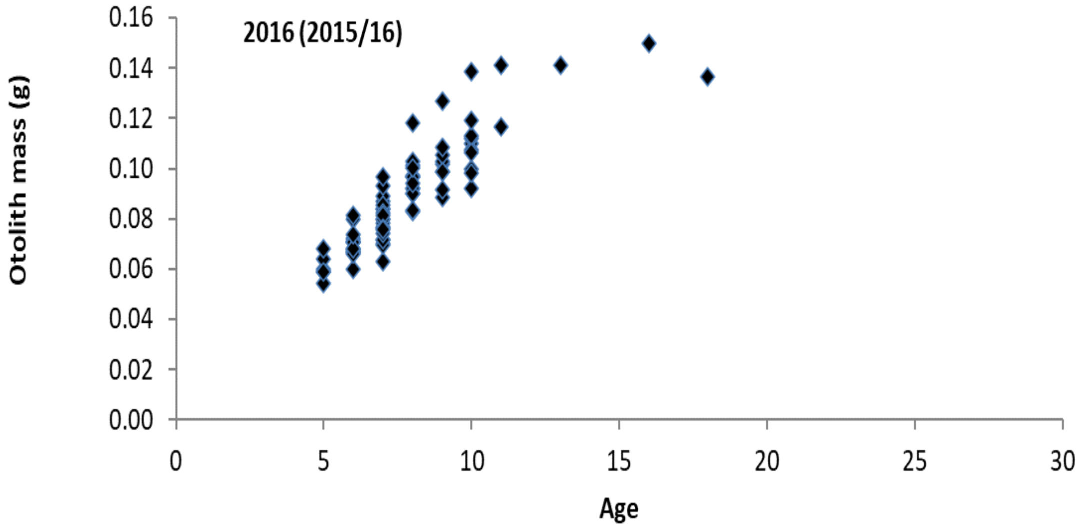


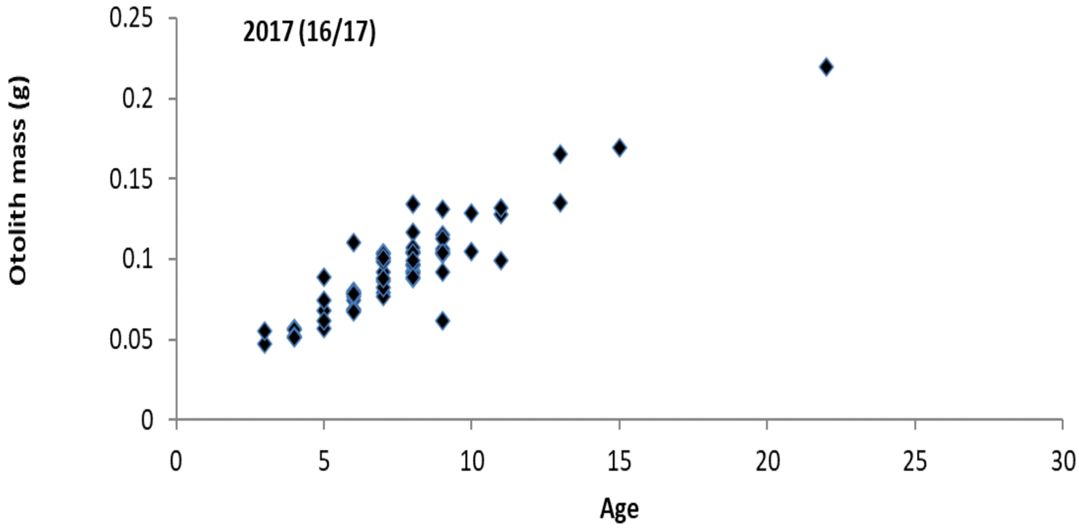


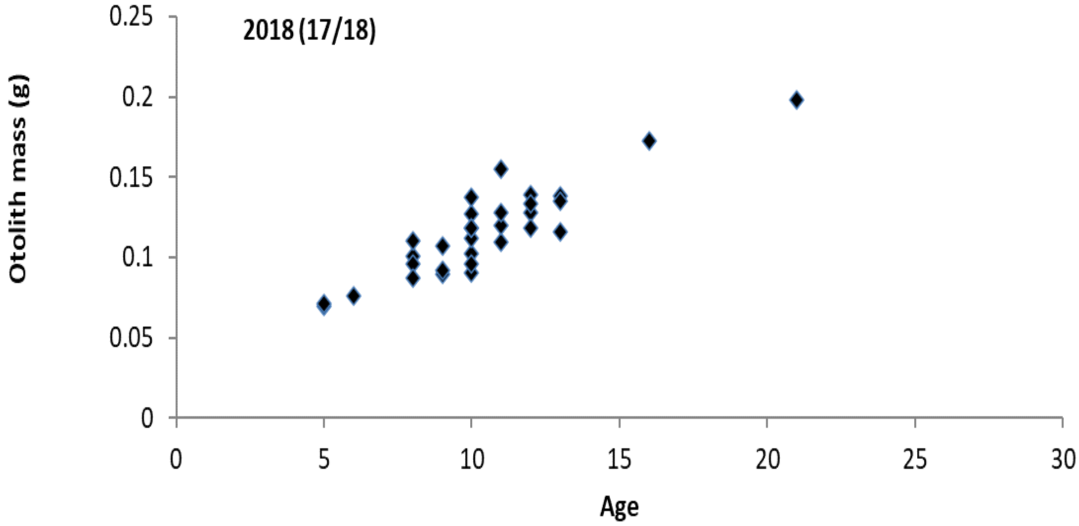
APPENDIX 3: Age-otolith weight and age-fish length relationships (2014/15 age otolith weight and age fish length relationships include previous age estimates from project SEA2015/19)

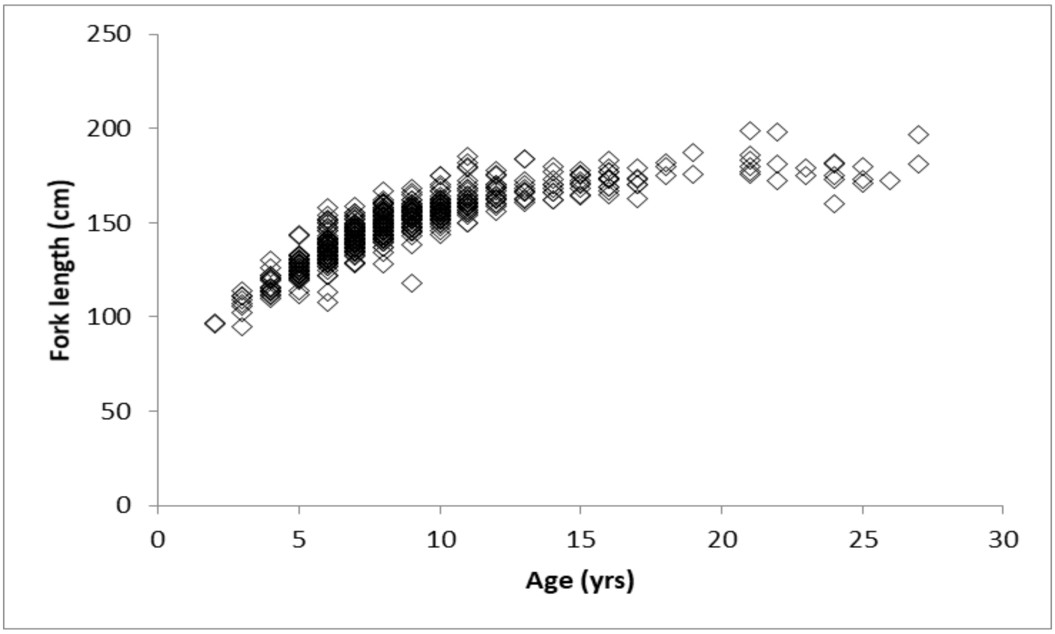
(Continued on next page)








2018 (17/18)
Otolith mass (g)
0.25
0.2
0.15
0.1
0.05
0
0
5
10
15
20
25
30
Age


Fork length (cm)
250
200
150
100
50
0
0
5
10
15
20
25
30
Age (yrs)

APPENDIX 4: The age difference between paired readings from the same otolith by the same reader

Age 1

Difference (Age 1 – Age 2)	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20+	N	% Agree	% +/1
-3						1			1							1			1		1	5		
-2						3	1		1		2	2			1						2	12		
-1				1	7	5	9	11	13	12	3	5	1		1	1	1				4	74		
0	**0**	**0**	**0**	**2**	**11**	**24**	**28**	**30**	**31**	**14**	**14**	**5**	**4**	**1**	**0**	**0**	**4**	**0**	**1**	**0**	**2**	**171**	**47.37**	**89.20**
1					1	3	5	15	14	5	8	8	2		6	2	2	1	2		3	77		
2								1	1	1	2	1	4	2	1		1			1	3	18		
3										1		1			1		1					4		

APPENDIX 5: Adjusted age composition 2009–2014 seasons

Note: 2009 to 2011 age composition sourced from Krusic-Golub (2012) and 2012 to 2014 from Krusic-Golub (2015).