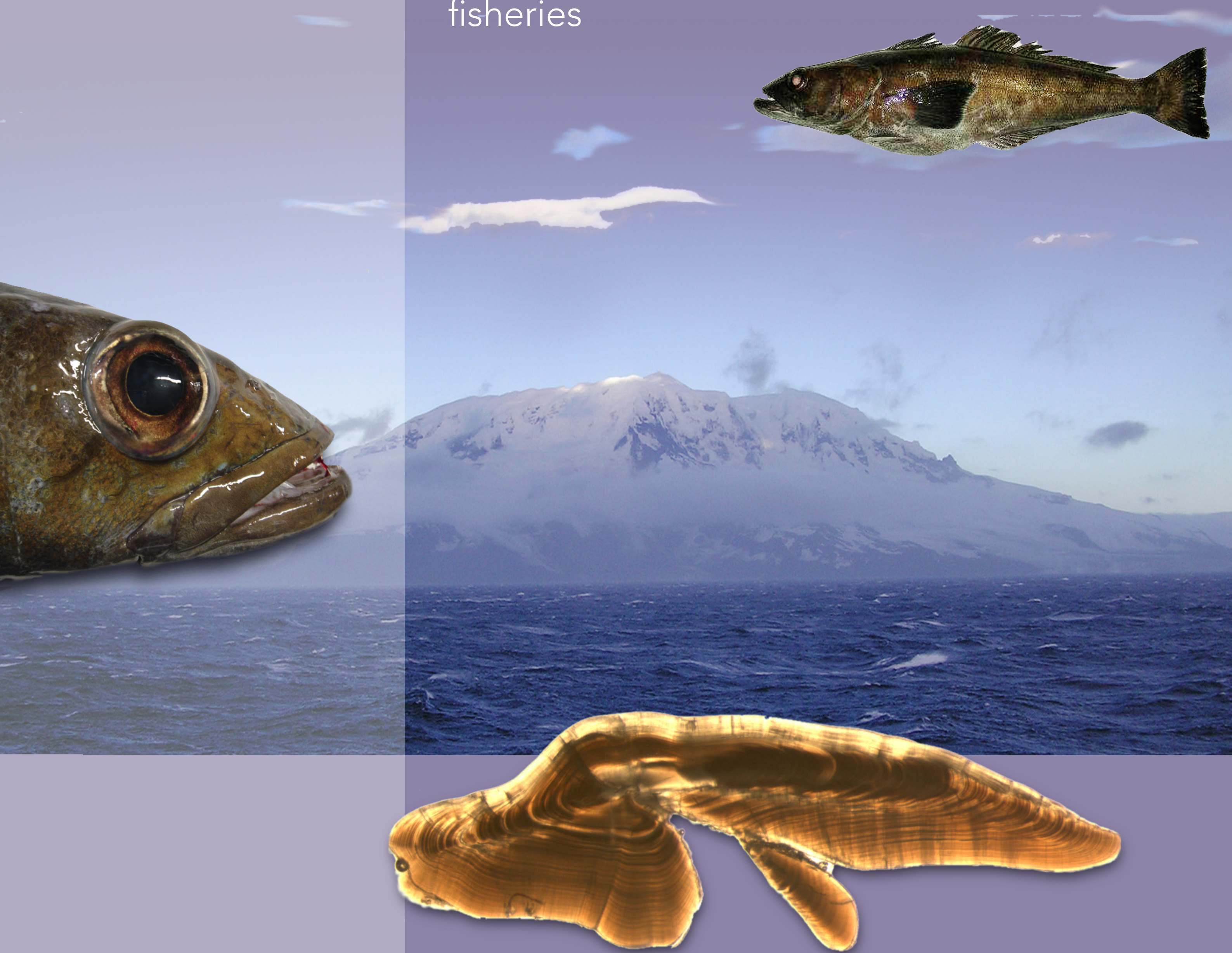
Robust characterisation of the age structure, growth and recruitment of toothfish in the Macquarie Island and Heard Island and McDonald Islands fisheries



AFMA project 2009/839
FINAL REPORT

D. C. Welsford, S.G. Candy, J.J. Verdouw and J.J. Hutchins

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The Department of Sustainability, Environment, Water, Population and Communities, Australian Antarctic Division and the Australian Fisheries Management Authority

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PRINCIPAL INVESTIGATOR Dr Dirk C. Welsford

ADDRESS Department of Environment, Water, Sustainability, Population and Communities

Australian Antarctic Division

203 Channel Highway

Kingston TAS 7050

Telephone 03 6232 3608

Fax 03 6232 3449

Email dirk.welsford@aad.gov.au

Objectives

1 Characterise the age structure of toothfish captured in the Heard Island and McDonald Island and Macquarie Island commercial fisheries

2 Characterise the age structure of toothfish captured in the Heard Island and McDonald Island annual random stratified trawl survey

3 Provide catch at length estimates in the form of age length keys for incorporation into assessment models for Macquarie Island and HIMI toothfish

4 Evaluate the performance of assessments with the inclusion of age length data and the need for ongoing ageing analysis for Australia's toothfish fisheries

Non-Technical Summary

Patagonian toothfish (*Dissostichus eleginoides*) has been targeted by Australian vessels at Macquarie Island (in the Pacific sector of the Southern Ocean) and at Heard Island and McDonald Islands (HIMI, in the Indian Ocean sector of the Southern Ocean) since the mid-1990s. These fisheries are currently changing, from most of the catch being taken by trawl and comprising juveniles, to multi-gear fisheries with longline catches dominating, taking larger, older fish from deeper water.

The AAD conducts the stock assessments for the HIMI toothfish fishery. The assessment integrates a number of datasets, including commercial catch at age as well as abundance at age estimated from an annual random stratified trawl survey, which aims to sample juveniles prior to their entry into the commercial fishery. Recent surveys have seen a decline in catch rates; however it has been difficult to determine if this is due to a decline in the abundance of juveniles, or other factors such as variation in catchability.

The Macquarie Island Toothfish Fishery (MITF) is assessed by CSIRO. The assessment has recently changed to integrate a larger number of datasets, including commercial length-at-age and tag recapture data. The model now includes the facility to incorporate more information on length and catch at age; however the quantity of such data was limited, and included little information on the larger fish that dominates the catches since longlining commenced in 2007. Catch rates in the fishery have also varied considerably, and with similar difficulty in interpretation as to the cause as for the HIMI fishery.

Scientific observers have been aboard toothfish fishing vessels on all their cruises since 1997 and have collected large numbers of otoliths in most years these fisheries have operated, across all the size classes captured by the different gear types. The AAD has developed a highly consistent and efficient method to cut thin slices from large numbers of toothfish otoliths and estimate their age from the rings that are laid down in the otoliths each year. Catch at age data has the potential to enable the more accurate estimation of which age classes of fish are present in the population, the quantities of fish recruiting each year, and which age classes are most vulnerable to the different fishing gears. This enables a much more detailed picture of the impact of fishing on these populations, and enables managers to make decisions on catch limits that are more likely to result in sustainable fishing.

This project was established to process a large number toothfish otoliths, including those collected from recent longlining (2007-2010) and earlier trawling (2000-2009) at Macquarie Island, as well as those from the four Random Stratified Trawl Surveys at HIMI (2009-2011). These data were then incorporated into the most recent assessments for those fisheries.

The 2011 assessment for HIMI included ages from 2775 toothfish captured in trawl surveys. The revised assessment estimated that the abundance of at least two year classes born between 2001 and 2006 were well below average, which is likely to have reduced catches in the trawl survey between 2005 and 2009 when these fish became vulnerable to the survey. Catch rates in recent surveys are gradually increasing, and catch at age data indicate that recent year classes, while still variable, are closer to average abundance, however as toothfish at HIMI may live up to 28 years, the impact of these low recruitments may affect future yields for many years to come. This model was reviewed by international scientists at the annual meetings of the Commission for the Conservation of Antarctic Marine Living Resources (CCAMLR) and it was agreed that the catch limit for the next two seasons could increase to 2730 tonnes from 2550 tonnes on the basis of this model. Excluding recent catch-at-age data from the assessment model leads to a reduced ability to estimate the future trajectory of the HIMI stock and therefore the less certainty as to the catch levels that would be sustainable. Therefore the incorporation of contemporary catch at age data in future stock assessment models is recommended.

An additional 1737 ages from longline were also available to be incorporated into the most recent Macquarie Island stock assessment. The data from larger fish captured by longline, some estimated to be in excess of 40 years old, enabled refinement to key parameters used in the new integrated assessment, including re-estimation of the growth model to provide more plausible estimate of growth for older age classes, and estimation of selectivity ogives for the longline fishery. A further 1283 ages for fish caught in trawls between 2000-2009 will be incorporated into the assessment being developed for early 2012, and is expected to allow a more accurate picture of the impact of fishing on this stock just prior to transitioning to a primarily longline fishery.

Overall, the results of this project support the value of processing contemporary and archived otoliths. The stock assessments that have used the datasets generated by this project have substantially improved the robustness of estimates of sustainable harvest for these important Australian fisheries. For long-lived species such as toothfish, where the consequences of interannual variability in recruitment may persist for decades, and because the HIMI and Macquarie Island fisheries that are undergoing important changes in the parts of the population they catch, collecting and ageing otoliths, and developing statistical methods for incorporating this data fully into stock assessments should remain a research priority.

Outcomes Achieved

The primary outcome of this project has been the successful inclusion of large amounts of otolith based age-length data in the recent assessments of the Heard Island and McDonald Islands (HIMI) and Macquarie Islands (MITF) toothfish fisheries. The inclusion of these datasets have improved the estimates of key parameters population dynamics of toothfish including growth rates, mean recruitment levels and interannual variability, as well as allowing more precise estimates of the selectivity of the different fishing gears used in these fisheries. Therefore stock assessment scientists, resource managers, industry and other stakeholders are in a much stronger position to understand the historical impact of these fisheries, and evaluate the likely long term sustainable harvest.

The results of this project firmly establish that consistently high-quality; high-throughput otolith processing is a cost-effective method of providing age estimates for toothfish of all sizes. The protocols used in this project were able to produce large datasets in a timely manner, ensuring stock assessments were able to be based on the most up-to-date estimates of population status. For example, the HIMI stock assessment presented at CCAMLR in October 2011 included length-at-age data from more than 500 fish captured during the annual Random Stratified Trawl Survey in April in the same year, and age length keys that were based on an overall dataset of more than 10200 aged individuals.

The project was also able to use the large number of archived otoliths collected in these fisheries to fill critical data gaps. For example little catch-at-age data was available in the MITF between 2000 and 2011, which was a period of significant change in the fishery, both in catches as well as the main gear type used. However archived otoliths from this period were processed during this project, and the data included to date have enabled the structure of the population and the impact of historical fishing to be reconstructed with unprecedented detail.

Keywords

Patagonian toothfish, *Dissostichus eleginoides*, age-based assessment, length-based assessment, Heard Island and McDonald Islands, Macquarie Island, integrated assessment, otoliths

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Our sincere thanks also, to the Australian Fisheries Management Authority scientific observers and Data Collection Officers from Fisheries Audit Services and Capfish P/L, for collecting all of the otoliths used in this project. Many thanks also to Dick Williams for establishing the original otolith collection program, Tim Lamb, Gabrielle Nowara, Troy Robertson and Steve Cameron for database management.

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Background

This project was developed through consultations with all key stakeholders in the HIMI and MITF, informally as well as at meetings of the Sub Antarctic Resource Assessment Group (SARAG) between 2007 and mid 2009. The application was developed to address several strategic research issues that have been identified over recent years, relating to AFMA objectives for sustainable resource management; some specific to either HIMI or MITF, others common to both, but all of which have the potential to be resolved using age-based population modelling approaches (see below).

Fishing at HIMI and MITF commenced in the mid 1990s, primarily as trawl fisheries targeting juvenile toothfish. Since 2003, longlining has grown to become an important capture method at HIMI, leading to an increase in the range of depths fished and sizes of toothfish captured (Welsford *et al.*, 2011; Welsford *et al.*, 2009). Since the commencement of an experimental longline fishery in the MITF in 2007, this method has developed to become the primary capture technique on the Macquarie Ridge, leading to fishing in areas that were inaccessible to trawling. Further details on the development of the fisheries and assessments at HIMI can be found in Patterson & Skirtun (2011a), Welsford *et al.* (2011; 2009) and Constable and Welsford (2011), and for MITF in Tuck (2007, 2009), Patterson & Skirtun (2011b) and Fay and Tuck (2011).

Both assessments now use a Bayesian integrated assessment frame work; CASAL for HIMI (Bull *et al.*, 2005) and Stock Synthesis for MITF (Methot, 2005). However, the principle index of abundance that the assessments models currently use differs. The HIMI assessment is strongly influenced by the results of an annual trawl survey which provides abundance data for pre-recruits (Candy and Constable, 2008), while the primary input into assessments of the MITF is mark recapture data (Fay and Tuck, 2011; Tuck, 2009). However, in both fisheries, variability in catch rates is common, and attribution of this variability to selectivity, changes in year class strength and/or availability of fish in the main grounds has been difficult.

Completion of a recent Fisheries Research and Development Corporation Tactical Research Fund project using subsamples of otoliths collected from the HIMI (Welsford *et al.*, 2009), showed that time series of catch-at-age derived from age length keys significantly improved estimates of gear selectivity and year class strength in stock assessment models for that region. Industry and AFMA agreed that due to the success in applying age length keys to assessment of the HIMI fishery, similar work should be conducted to provide key inputs into the new MITF integrated assessment concurrently developed by CSIRO (Fay and Tuck, 2011). Further, as a result of materials and expertise developed in the tactical research fund project, the capacity to perform the large amounts of otolith analyses required existed, and the project would a have a high probability of achieving its objectives. The project commenced in January 2010.

Need

Accurate estimates of size-at-age and recruitment variability, as well as fishery specific catch-at-age and gear selectivity are critical to the integrated stock assessments for toothfish at HIMI and in the MITF. Otolith analysis represents a powerful method for improving these estimates. Recent work on otoliths collected from HIMI between 1997 and 2007 shows that age length keys significantly improve the biological plausibility and precision of year class strength estimates, which are critical to predicting the productivity and sustainable yield from these fisheries. Commercial trawl and survey catch rates at HIMI indicated that recruitment may have been very low in the most recent seasons. Hence there was an urgent need to analyse otoliths from recent surveys (2008-2011) to better understand the role of variability in recruitment in survey catch rates, and its likely impact on long term yields for the fishery at HIMI. Analyses of otoliths from larger fish are also required to better characterise the catch at age and selectivity from the longline and pot fisheries. Prior to the commencement of this project, the MITF assessment did not incorporate any recent catch at age data collected from the commercial trawl or experimental longline fishery. With the development of an integrated assessment framework, the inclusion of age length data is likely to improve the precision and accuracy of the assessment of this fishery as it has developed to include longline and trawl methods.

Methods

Otolith sampling and sub-sampling

Using previous toothfish ageing work done by researchers at the AAD as a guide, it was estimated that approximately 6000 otoliths samples could be processed and aged by two researchers during the 18 month timeframe of this project. Before the project commenced, otoliths from particular fisheries, seasons and gear types were identified as priority samples to be aged. These were (in order of priority): otoliths from fish captured in the MITF by longline (2007 to 2010); otoliths from fish captured during the HIMI Random Stratified Trawl Surveys (2008 to 2011) and archived samples collected from fish captured in the MITF trawl fishery (2000 to 2009). There were many more than 6000 otoliths in the AAD otolith collection from these fisheries (Appendix 3), and as a result sub-sampling of otoliths for several seasons from the HIMI RSTS and MITF trawl fishery was required.

Macquarie Island longline fishery otoliths were not sub-sampled, and instead all otoliths were processed and aged. This decision was based on the fact that the number of samples available totalled less than 2000, and was therefore tractable within the scope of the project. Furthermore, the length frequency distribution (Appendix 3, Table A3.3) showed a relatively even spread of samples over a broad size range, and was therefore was deemed likely to contain a broad range of age classes. In addition, samples from this fishery were of particular importance to the study since no age information had been obtained from this fishery previously. The total number of otoliths processed and aged from this fishery was 1737 (from 353 to 517 otoliths from each of the four seasons) (Table 1).

The number of otoliths collected from HIMI RSTS from the four seasons from 2008 to 2011 totalled 4806 (ranging from 523 to 2372 per season) (Table 1). The length frequency distributions showed that samples from each of the seasons were dominated by smaller (<600mm) with few larger (>1000mm) fish (Appendix 3, Table A3.1). As the majority of fish fell into similar length classes, and based on previous experience were likely to be mostly form a narrow range of age classes, the decision was made to sub-sample otolith samples from each season, with the exception of the 2011 season where only 523 otoliths were collected (Table 1). The sub-sampling process for HIMI RSTS otoliths from seasons 2008 and 2009 involved dividing fish into 10mm length bins and then capping the number of fish to be randomly sampled from each length bin to 20, the minimum number estimated to adequately quantify the distribution of age classes within a length bin (McKinlay, 2009). In the 2010 season, two surveys were conducted, and a much larger number of samples were collected compared with other years and the number of fish selected from each length bin was capped to 10 per survey. The result was that a total of 2775 otoliths from the four HIMI RSTS seasons were processed and aged, providing age data which covered the entire size range of fish collected from each survey, and between 492 and 656 individuals per survey (Table 1).

Otoliths from the Macquarie Island trawl fishery from all seasons (1995/96 to 2009) were checked against database records. Some samples from seasons 1995/96 to 1998/99 had previously been processed and aged by the Central Ageing Facility (CAF) and the Australian National University for previous studies (He and Furlani, 2001; Kalish and Timmiss, 2001) and consequently priority was placed on otoliths collected from 2000 to 2009 seasons. Approximately 2400 otoliths were accounted for in total from the aforementioned seasons. The scope of the project meant that only half of these could be processed and as a result otoliths were sub-sampled from each season. In seasons where less than 200 otoliths were accounted for, all otoliths were selected for processing and ageing. However, where samples collected in a given season exceeded 200, otoliths were sub-sampled by capping each 10mm length bin to 3 or 5 samples per bin. This resulted in a total of 1268 Macquarie Island trawl caught toothfish otoliths selected for sampling (Table 1).

Table 1. Otolith samples processed during the current project. (LL = Longline fishery, RSTS = Random Stratified Trawl Survey)

Fishery	Cruise	Season	Number of otolith samples		
			In database	*Sampled*	*Processed & aged*
Macquarie Island (LL)	AC03	2007	431	427	427
Macquarie Island (LL)	AC04	2008	365	353	353
Macquarie Island (LL)	JA10	2009	548	517	517
Macquarie Island (LL)	JA11	2010	543	440	440
				sub total	1737
HIMI RSTS (Trawl)	SC50	2008	992	656	656
HIMI RSTS (Trawl)	SC53	2009	919	654	654
HIMI RSTS (Trawl)	SC57	2010a	1347	450	450
HIMI RSTS (Trawl)	SD59	2010b	1025	492	492
HIMI RSTS (Trawl)	SC61	2011	523	523	523
				sub total	2775
Macquarie Island (Trawl)	AL22	2000	53	27	27
Macquarie Island (Trawl)	AL25	2000	311	182	182
Macquarie Island (Trawl)	AL27	2001	120	104	104
Macquarie Island (Trawl)	AL38	2002	73	63	63
Macquarie Island (Trawl)	AL44	2004	672	230	230
Macquarie Island (Trawl)	SC40	2006	662	231	231
Macquarie Island (Trawl)	SD45	2007	37	20	20
Macquarie Island (Trawl)	SC49	2008	490	226	226
Macquarie Island (Trawl)	SD52	2009	412	185	185
				sub total	1268
		TOTAL	9523	5780	5780

Preparation of otolith sections, interpretation and quality assurance

The preparation method and ageing protocol followed that described in Nowara *et al.* (2009), and is summarized below. Each otolith was cleaned, if necessary, and then weighed to ±0.0001g on a Mettler Toledo balance. One otolith of each pair, chosen at random, was set in a polyester resin block, the primordium marked under a low magnification stereomicroscope and three to five 0.35mm sections were cut around the primordium using either a Buehler Isomet low speed saw or a Gemmasta high speed saw, both with diamond impregnated blades. The sections were rinsed in water and ethanol before being mounted on a slide. The three to five sections from one otolith were placed in order of cutting onto a single slide and covered in clear casting resin under a cover slip. Images were taken of the best section and the number of increments was read from these images and entered into the database along with a readability index. Each otolith was rated for readability, using a five point scale from 1 (unreadable) to 5 (very good).

All 5780 otoliths were read once by two separate readers. In addition the AAD Patagonian toothfish reference collection (some 200 examples) was read by both readers three separate times during the project to assess possible drift in age estimates over time. The first reference collection read was done on commencement of the project, the second midway through the project and the third read was done on completion of the project. Within reader bias plots and between reader bias plots from all three reference collection reads showed no significant deviation from a 1:1 relationship, indicating results over time and from different readers can be pooled without any need for bias correction. Evaluation of consistency within and between readers when reading the reference collection is detailed in Appendix 4.

Incorporation of catch-at-age data into stock assessments

The most recent stock assessment for the MITF (June 2011) successfully incorporated the additional 1737 ages derived from longline catches between 2009 and 2010, as detailed in Fay *et al.* (2011), which builds on the integrated assessment model described in Fay & Tuck (2011). The 2011 HIMI assessment incorporated the 2775 ages from the recent RSTS, as well as all previous age-length data in season and sub-fishery age length keys (Welsford *et al.*, 2009 and below). The assessment model and input data is described in detail in Candy & Welsford (2011) as well as below.

Evaluation of the benefit of incorporating ageing data in the HIMI assessment

Model structure

To investigate the effect of the age data available due to this project on the interpretation of stock status and productivity, and in particular the recommended long-term sustainable catch, a comparison was made between the results of the CASAL assessment model presented to CCAMLR in 2011 (Model 1, denoted *a2-2011-alkall-PE* in Candy & Welsford (2011)), and a model (denoted Model 2) that replaced the data for commercial catch-at-age proportions for each subfishery and year and abundance-at-age data for the RSTS from 2006 with catch-at-length proportions and abundance-at-length data.

Models 1 and 2 in this study differ primarily in how catch at length data, collected by observers measuring the catch, is converted into predicted catch-at-age (for commercial catches) or abundance-at-age (for catches in the RSTS). Model one uses all the available ageing data to predict the distribution of ages in each length class. In contrast, Model 2 requires CASAL to derive catch at age from catch-at-length to using a single von Bertalanffy growth model (Candy *et al.*, 2007), similar to the CASAL models used prior to the implementation of age length keys for HIMI assessments in 2009. Otherwise the parameters and model structures were identical. The methods used and detailed description of the CASAL models, gear types and subfisheries and data types used for model calibration are described in Candy & Constable (2008), Candy & Welsford (2009, 2011) and SC-CAMLR (2011).

Input data and data weighting

For Model 1, commercial catch-at-age proportions were obtained using age length keys (ALKs), derived from the aged samples of otoliths, applied to the catch-at-length, and input into CASAL. The ALKs were pooled across subfisheries[1] but disaggregated across fishing years, following a log-linear contingency table analysis that confirmed that there was no substantial loss of information by pooling across subfisheries(Candy and Welsford, 2009). A separate ALK was applied to the RSTS for each year of the survey from 2006 to 2011 (Candy and Welsford, 2011). For Model 1, abundance by year and proportions by age class from the RSTS were combined to give abundance-at-age data. The coefficient of variation for abundance-at-age numbers was also derived as described in Appendix 5. An ageing error matrix, based on multiple re-reads of the reference collection was also incorporated using the method of Candy *et al.* (2012) was also estimated and included in the CASAL input for Model 1 (Appendix 6, Table A6.1).

Effective samples sizes (ESS) for commercial age-at length data, incorporating ALK sampling and random ageing error for Model 1 and haul level variability in commercial catch-at-length proportions for Model 2 were also calculated (Candy, 2008, 2009). For all commercial catch-at-age and catch-at-length proportions, the ESS also incorporated the effect of process error to avoid giving too much weight to these datasets. No process error component was calculated for the abundance-at-age and abundance-at-length data, as a heuristic way of giving statistical weight to the fisheries-independent survey (Francis, 2011).

[1] The CASAL assessment model used for HIMI is non-spatial, however to capture some of the spatial structure of the fishery, which has tended to focus on discrete fishing grounds, data is partitioned into subfisheries denoted as follows: f1=Trawl survey; f2= Trawl, Ground B; f3= Trawl, Ground C, f5= Longline Ground C, f6 = long line Ground D, f7 = Longline Ground E, f8= Trawl Ground F, f9 = Longline Ground F, f10 = Pot.

The resulting input datasets are summarized in Appendix 7 Tables A7.1-A7.3. Table A7.1 shows the age length data for each subfishery in each season[2]. For Model 1, where the trawl sub-fisheries are subdivided into periods within each season, the same season ALK was applied to the length frequency (LF) across all periods. Table A7.2 gives the sample sizes for length measurement for each fishery and for the RSTS (i.e. f1) carried out in each year. For 2009 to 2011 where insufficient fish were aged the catch-at-length proportions were retained but for a given sub-fishery the same selectivity function and parameter values were logically applied to both types of data.

Table A7.3 gives the ESS applied in the CASAL assessment for catch-at-age and catch-at-length proportions. Table A7.3 also shows the ESS after 4 iterations of the method described in Candy (2008) for accounting for process error. This number of iterations was required for the ESS to stabilise with no further reductions of practical significance. In a small number of instances, insufficient data was available to estimate the ESS values, so a value of either 200 (or 400 where greater than 1500 fish were measured in any season) was assigned. For Model 2 the ESS for catch-at-length proportions was also reduced for process error using 3 iterations.

Selectivity functions

The fishing selectivity functions fitted are those described in Candy & Constable (2008) and Candy & Welsford (2009) with one exception. The Double Normal Plateau (DNP) function was replaced by a Double Normal (DN) function for the main survey group (Group 1, years 2001, 2002, 2004-2011) since the plateau length, parameter a2, was typically estimated to be very small (≈0.1 yr), thus collapsing to a DN function and causing numerical problems when calculating the profile likelihood for B0. The reduction in goodness of fit to the survey abundance data was not detectable when this parameter was dropped (i.e. set to zero) by fitting the DN function.

Projection trials

The harvest strategy within CCAMRL toothfish fisheries seeks to set catch limits that satisfy the CCAMLR decisions rules (Constable *et al.*, 2000), which can be described as follows:

1. choose constant annual catch γ_1 so that the probability of the toothfish spawning biomass being depleted below 20% of its pre-exploitation median level over a 35-year harvesting period is 10%;

2. choose constant catch γ_2 so that the median escapement in the toothfish spawning biomass over a 35-year period is 50% of the pre-exploitation median level (the ‘escapement’ rule); and

3. select the lower of γ_1 and γ_2 as the total allowable catch.

[2] The CASAL assessment model used for HIMI subdivides each assessment year into 3 periods, s1 (1 December- 30 April), when trawling and some potting occurs, s2 (1 May- 30 September) when trawling and longlining occurs, and s3 (1 October-30 November) when trawling dominates.

The key output from the CASAL models that significantly affects the calculated allowable catch under the CCAMLR decision rules are the estimates of year class strength (YCS) parameters and their coefficient of variation (CV_R). The depletion probability component of the decision rules states that no more than 10% of projected trajectories for spawning stock biomass, SSB, may go below 20% of the pre-fishery estimate of median SSB (i.e. B_0) over a 35 year projection of the current stock. Previous studies (Welsford *et al.*, 2009) show that ageing the catch and surveys provides sufficient additional information on YCS parameters that results in a reduction in their between-year (CV_R), and therefore a reduction in uncertainty regarding future recruitment, and a reduced likelihood that the depletion rule will constrain catch limits.

In CASAL projection trials, uncertainty surrounds the estimates of the parameters in the model as well as in how recruitment will vary in the future. In order to integrate across uncertainty in the parameters, sets of parameters were sampled from the results of the stock assessment in CASAL. The sampling method obtained independent multivariate normal (MVN) samples of the parameter set using the maximum posterior density (MPD) estimates of parameters and their estimated variance-covariance matrix.

Recruitment variability in each trial was modelled as a log-normal recruitment function. A random set of time series (1996 to 2008) of estimated number of age-1 recruits (corresponding to estimates of YCS for 1995 to 2007) were obtained using CASAL's projection procedure. To do this 1000, independent multivariate normal (MVN) samples of the parameter set were drawn using the parameter estimates and their approximate variance-covariance matrix, then used by CASAL to obtain 1000 samples of the recruitment time series. The samples of age-1 recruit numbers were then analysed in R (R Development Team, R Development Core Team, 2006) using a linear mixed model (LMM). The random effect variance for year was obtained using the LMM fit to the logarithm of the number of recruits obtained using the *ASReml* package (Butler *et al.*, 2007) within R. The square root of the variance of the year random effects gives a robust estimate (i.e. based on a 1st order Taylor series expansion of the log transformation) of CV_R required for the lognormal random recruitment facility in CASAL. Using estimates of YCS prior to 1995 was not considered for inclusion to the series for calculating CV_R since the reliability of these values is subject to a substantially greater degree of uncertainty compared to more recent years were larger number of fish have been observed.

Results

Incorporation of catch-at-age data into stock assessments - MITF

The data from larger fish captured by longline, some estimated to be in excess of 40 years old, enabled refinement to key parameters used in the new integrated assessment, including re-estimation of the growth model to provide more plausible estimate of growth for older age classes, and estimation of separate selectivity ogives for the longline and trawl fisheries, as well as the estimation of year class strengths in the population from 1985-2003 (Fay and Tuck, 2011; Fay *et al.*, 2011). The stock assessment was reviewed by SARAG and the recommended catch limit was accepted. A further 1283 ages for trawl caught fish will be incorporated into the assessment being developed by the CSIRO for early 2012.

Incorporation of catch-at-age data into stock assessments – HIMI

Stock assessments for the HIMI fishery occur every two years. As HIMI falls within the boundaries of the area managed under the Convention for the Conservation for Antarctic Marine Living Resources, assessments are reviewed the CCAMLR Scientific Committee's Working Group for Fish Stock Assessment (WG-FSA) as well as SARAG. The assessment developed for the 2011 meeting of WG-FSA (Candy and Welsford, 2011) was found to be appropriate to provide management advice and was used to set the catch limits for the next two seasons.

Evaluation of the benefit of incorporating ageing data in the assessment

Key parameters from the fit of Model 1 and Model 2 are given in Table 1.

Table 1. Results of assessments of stock status of *Dissostichus eleginoides* at Heard Island and the McDonald Islands, comparing the 2011 assessment model (Model 1) with a model with catch-at-age and abundance-at-age data replaced by catch-at-length and abundance-at-length data (Model 2). B_0 is the maximum posterior density (MPD) estimate of the pre-exploitation median spawning stock biomass (SSB), SSB status 2011 is the ratio of the model prediction of SSB in 2011 to B_0, R_0 is the MPD estimate of mean Age 1 recruitment prior to exploitation (1981), and CV_R is the coefficient of variation of the annual recruitment series (1996-2008).

Model	Description	B_0 (tonnes) (SE)	SSB Status 2011	R_0 (mil.)	CV_R
1	Model in Candy and Welsford (2011), presented to CCAMLR	86 400 (1 915)	0.629	5.765	0.78
2	Model 1 without applying age length keys to catch-at-length data	104 881 (2 886)	0.631	6.999	1.28

Figure 1 shows abundance-at-age observations from the RSTS and model 1 predictions for the main survey group. Figure 2 shows abundance-at-length observations from the RSTS and Model 1 predictions for survey years for the main survey group prior to 2006. Figure 3 shows abundance-at-length observations from the RSTS and Model 2 predictions for all years for the main survey group.

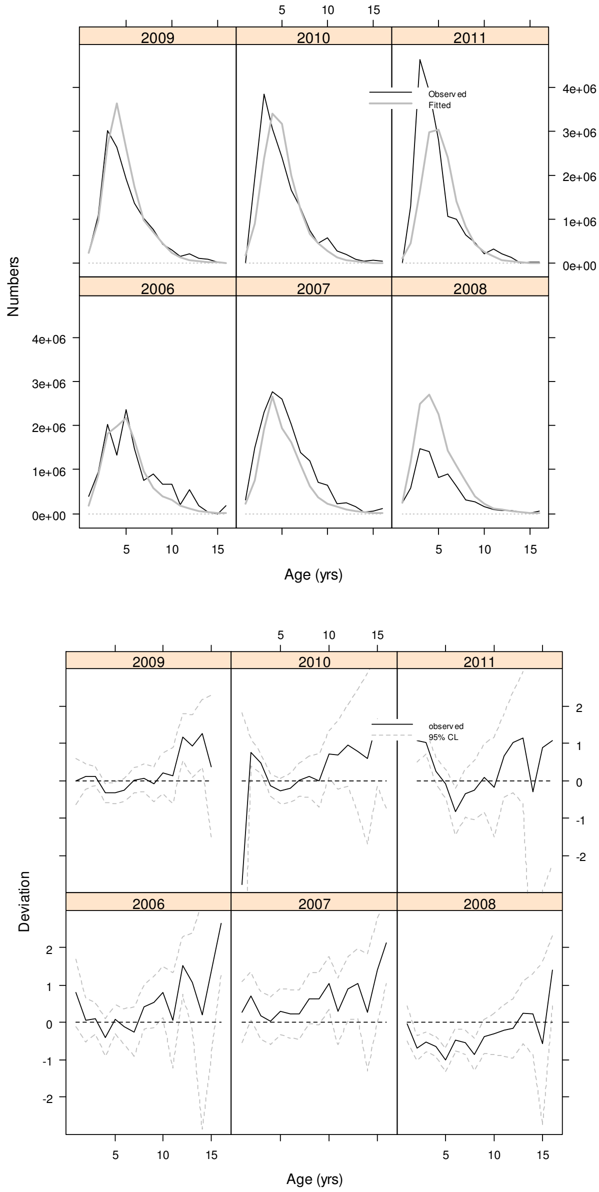


Figure 1. Abundance-at-age observations from RSTS and Model 1 predictions for years with substantial ageing data (2006-2011). The upper panel shows observed vs. fitted numbers and the lower panel deviations about predictions showing 95% confidence limits based on abundance-at-age CVs.

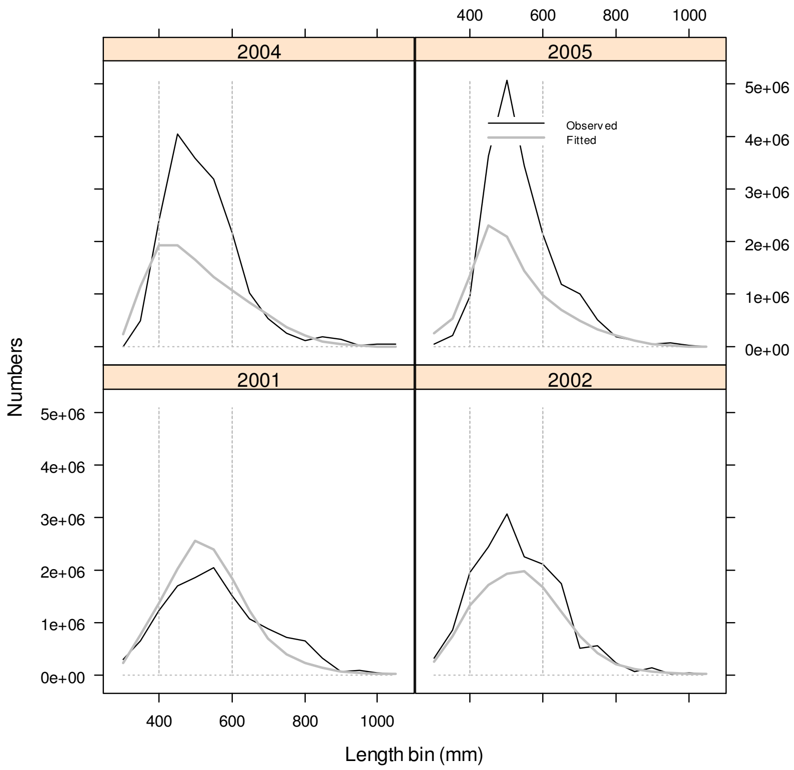


Figure 2. Abundance-at-length observations from RSTS Model 1 predictions for years with insufficient ageing data (2001-2, 2004-05).

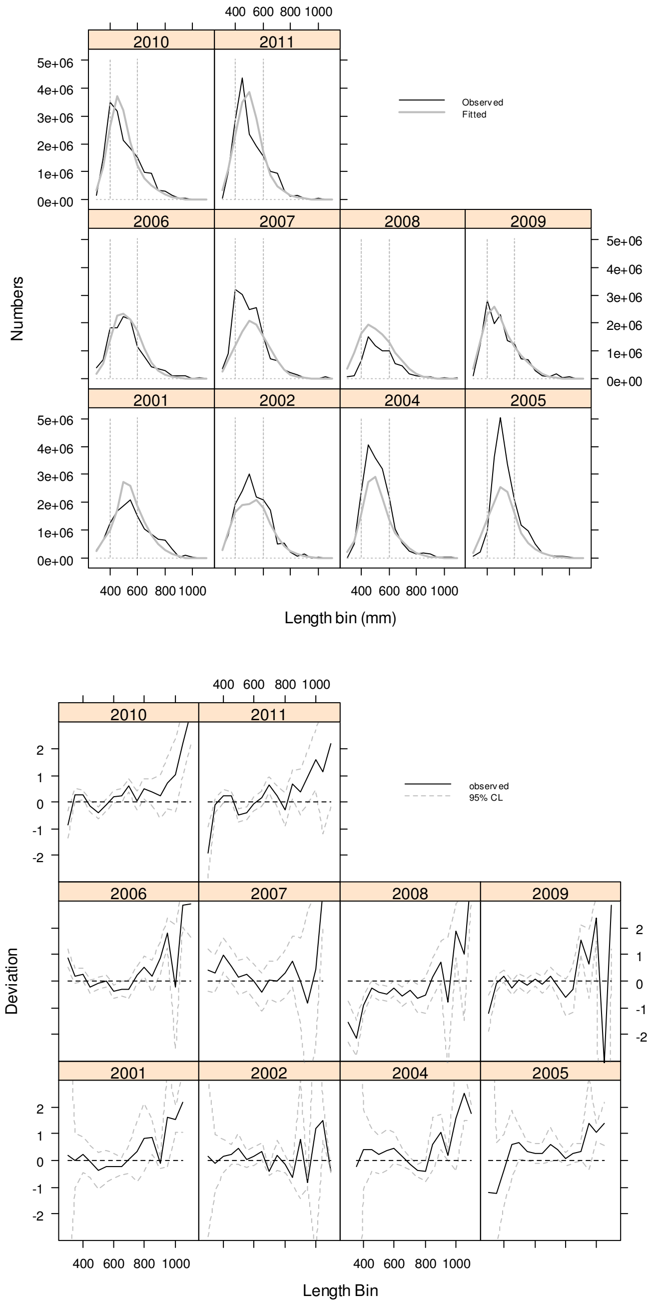


Figure 3. Abundance-at-length observations from RSTS and Model 2 predictions. Upper panel shows observed vs. predicted numbers, and the lower panel shows deviations about predictions showing 95% confidence limits based on abundance-at-age CVs.

Figure 4 shows observed and predicted proportions-at-age from Model 1 for sub-fishery f2-s2 (the main trawl ground in season 2) while Figure 5 shows observed and predicted proportions-at-length for this subfishery and season for years 2009-2011. Figure 6 shows the corresponding observed and predicted proportions-at-length from Model 2 for all years for f2-s2.

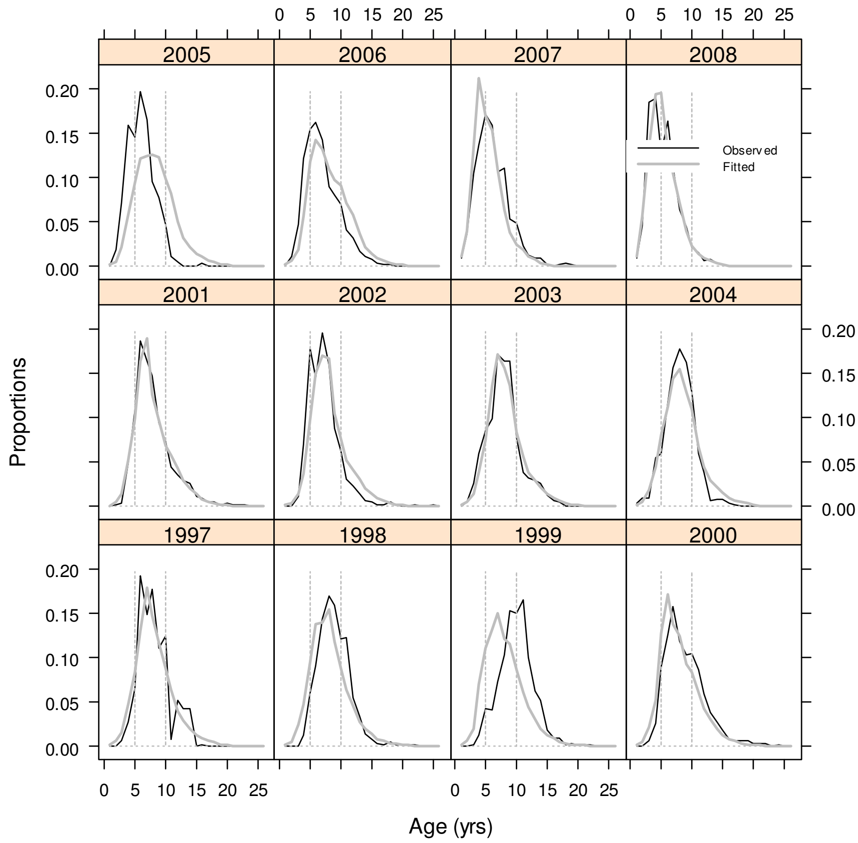


Figure 4. Observed and fitted proportions-at-age sub-fishery f2-s2 from Model 1.

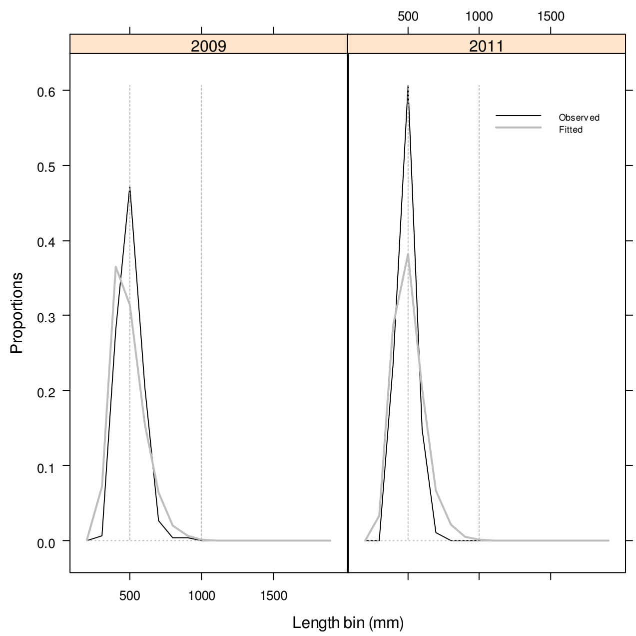


Figure 5. Observed and fitted proportions-at-length sub-fishery f2-s2 from Model 1.

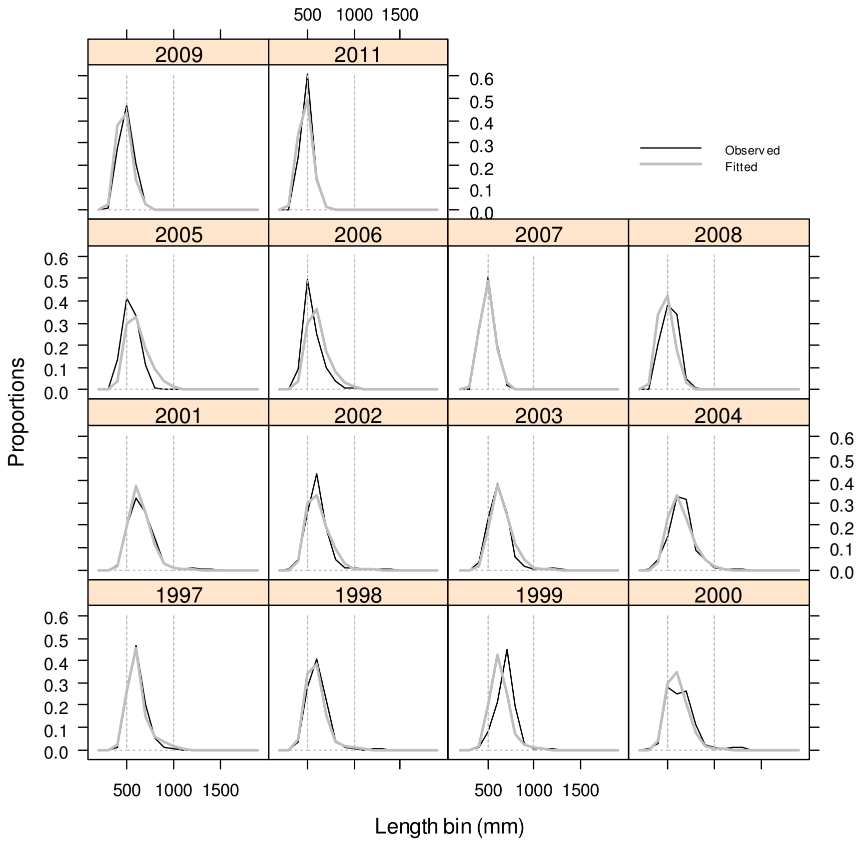


Figure 6. Observed and fitted proportions-at-length sub-fishery f2-s2 from Model 2.

Figures 7-9 show corresponding results for subfishery f5 (longline ground C, season 2). Similar, generally good fits were obtained for all other catch-at-age and catch-at-length proportion datasets form both models, with the exception of subfishery f8 (trawl ground) which has unusually noisy catch-at-length and corresponding catch-at-age distributions (see Candy and Welsford, 2011).

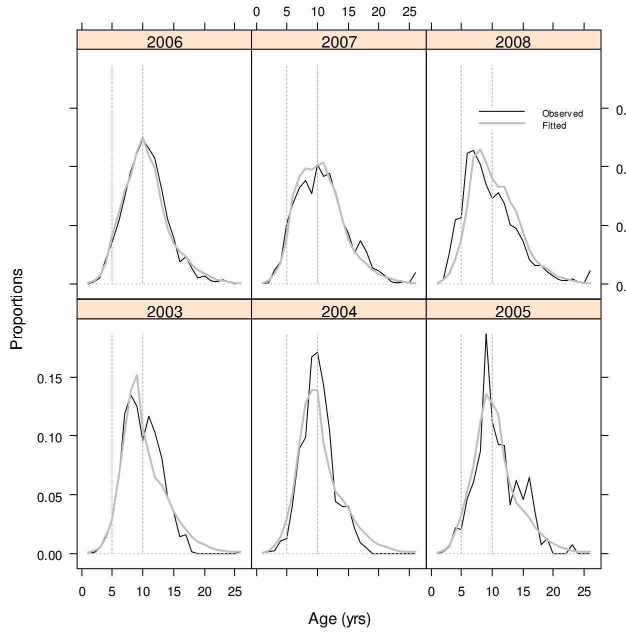


Figure 7. Observed and fitted proportions-at-age sub-fishery f5-s2 from Model 1.

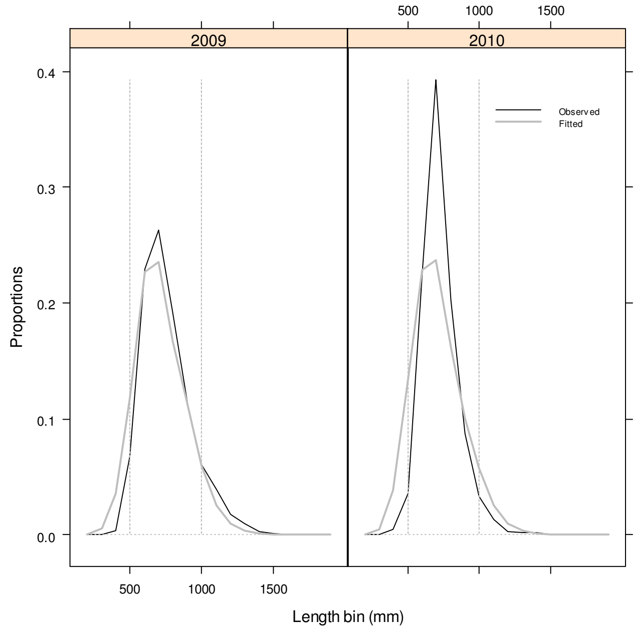


Figure 8. Observed and fitted proportions-at-length sub-fishery f5-s2 from model 1.

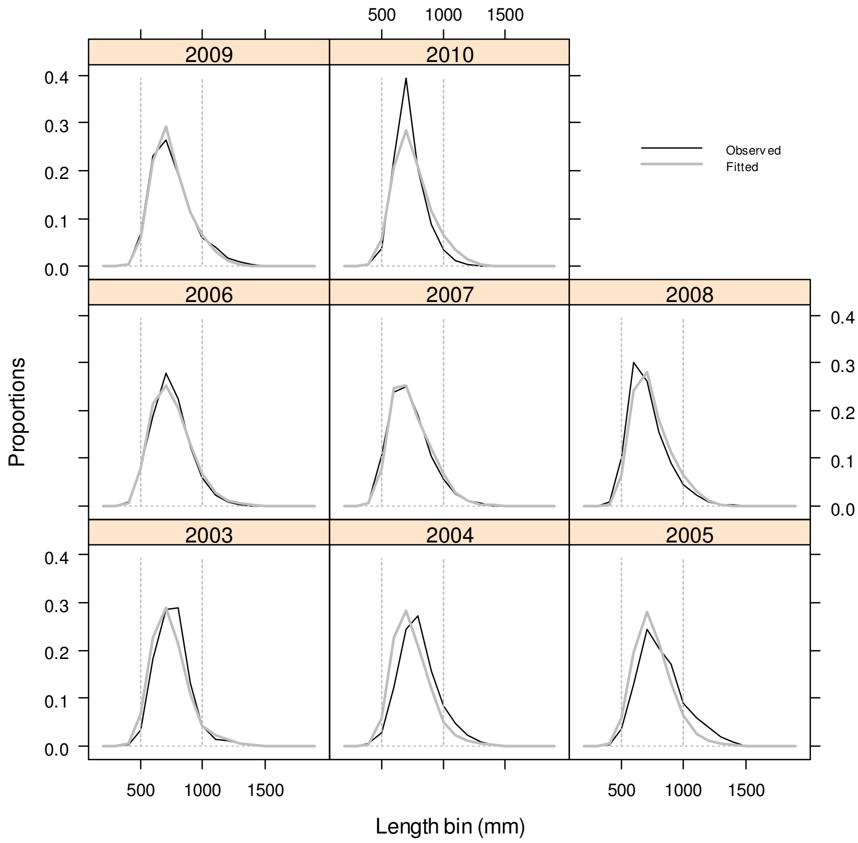
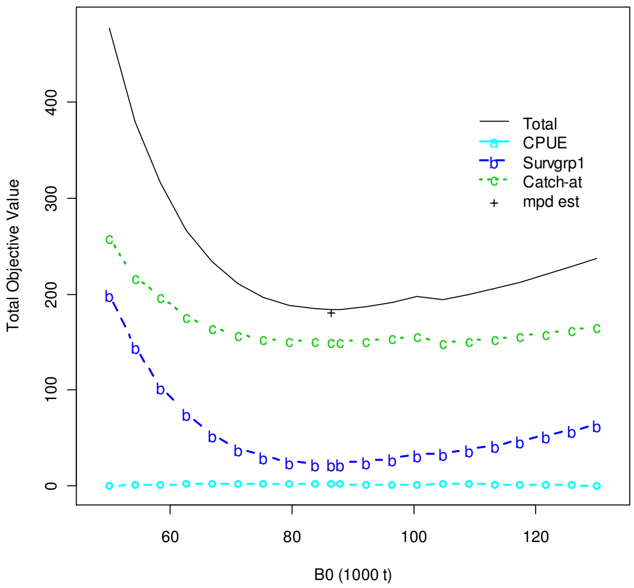


Figure 9. Observed and fitted proportions-at-length sub-fishery f5-s2 from Model 2.

Figures 10 and 11 show profile plots of the objective functions (i.e. -2 log-likelihood) versus B_0 disaggregated to the three types of observational data and separately for the total objective for Models 1 and 2, respectively. The maximum posterior density (MPD) estimate (i.e. corresponding in this case to the maximum likelihood estimate) of B_0 is also shown and appears as the minimum point of the profile in each case indicating estimation is well-behaved with respect to B_0.



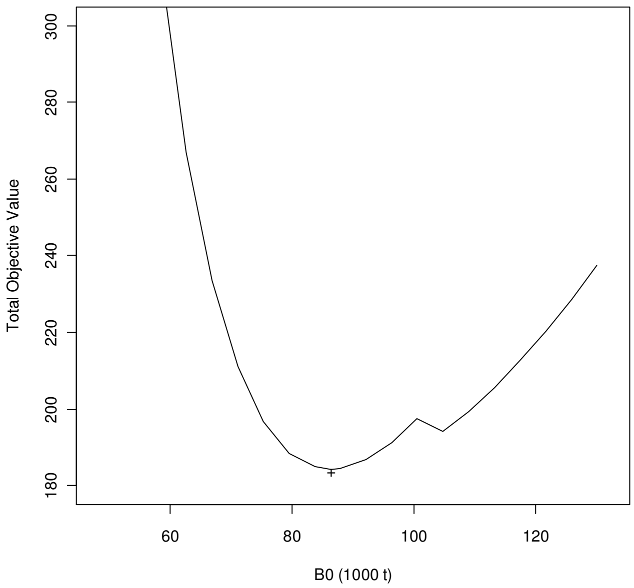
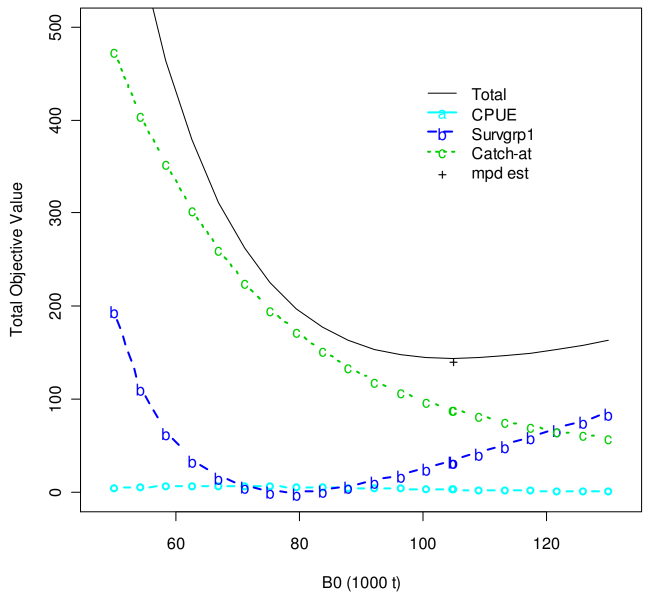


Figure 10. Profile of objective function (-2 log-likelihood) across grid of values of B0 showing the maximum posterior density (MPD) estimate for Model 1. The lowest point in the curve indicates the B0 estimate with the highest likelihood according to the model. The upper panel shows the MPD contribution by data source (catch per unit effort, survey abundance and commercial catch at age) and the total value and lower panel shows the total value presented on a restricted ordinate scale.



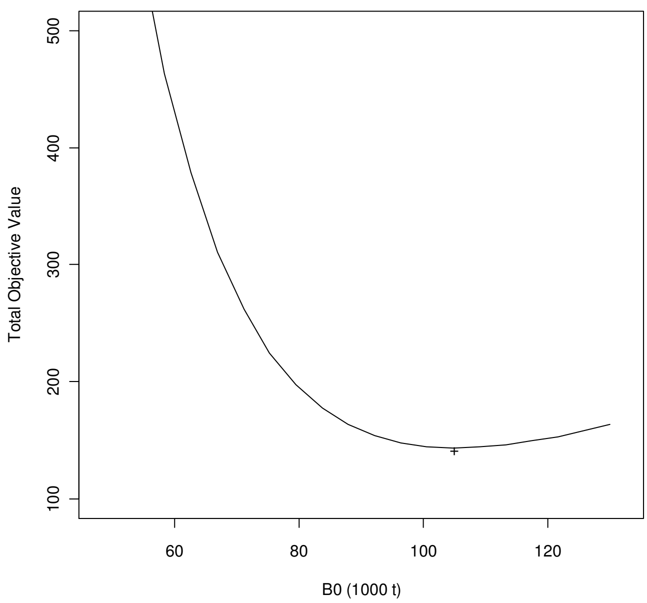


Figure 11. Profile of objective function (-2 log-likelihood) across grid of values of B_0 showing MPD estimate for Model 1. The lowest point in the curve indicates the B0 estimate with the highest likelihood according to the model. The upper panel shows the MPD contribution by data source (catch per unit effort, survey abundance at length and commercial catch at length) and the total value and lower panel shows total value presented on a restricted ordinate scale.

Figures 12 and 13 show the fitted selectivity functions for each of the survey groups and for each commercial sub-fishery along with 95% confidence bands based on the 1000 MVN samples of parameters. Table 2 gives selectivity function parameter estimates for the main survey group and catchability constants for the other survey groups for each of the models.

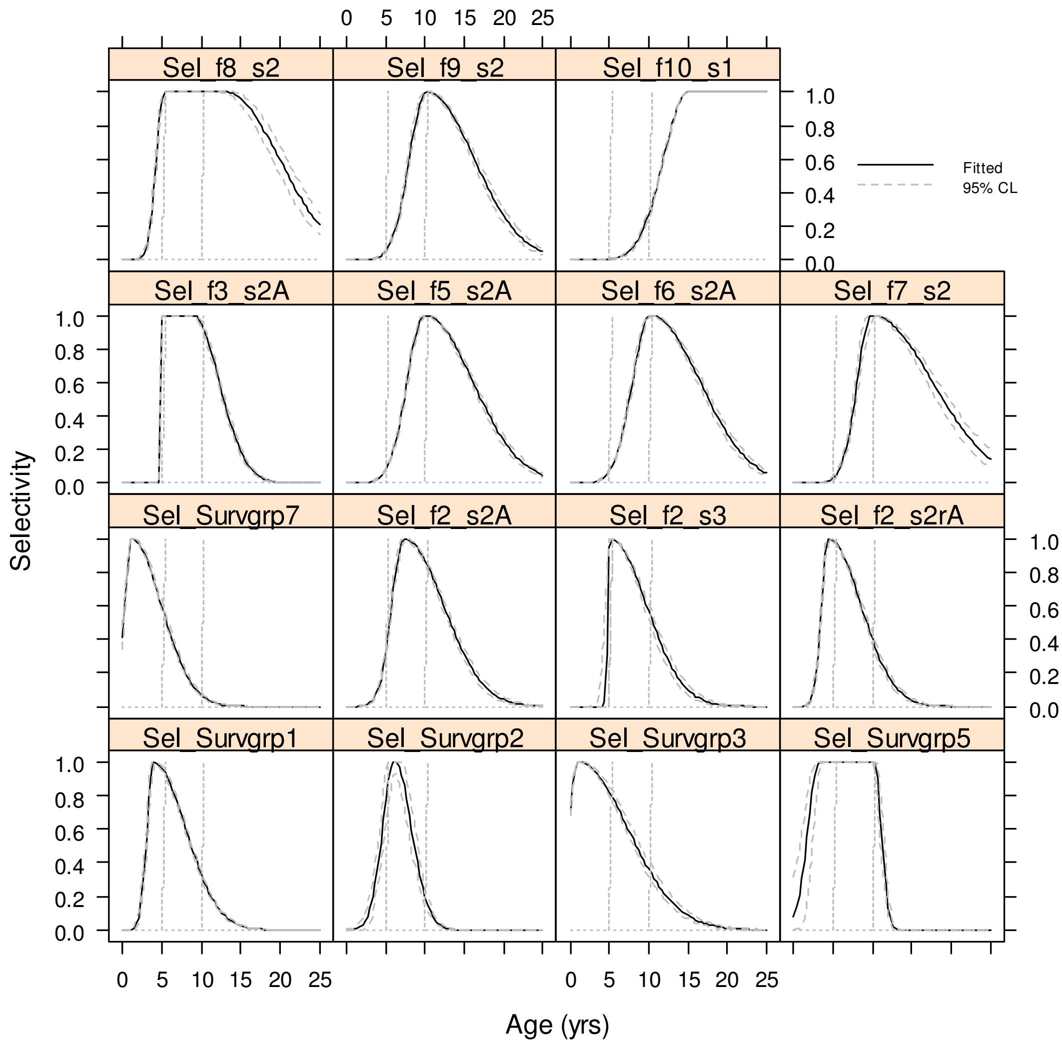


Figure 12. Double-normal-plateau (DNP) and double-normal (DN) fishing selectivity curves from the fit of Model 1 showing 95% confidence bounds obtained from the MVN sample. Panel headings: Survgrp1 (survey years 2001, 2002, 2004-2011), Survgrp2 (survey year 1999), Survgrp3 (survey year 1990), Survgrp5 (survey year 1993), Survgrp7 (survey year 2003), f2_s2, f2_s3 (trawl Ground B, seasons 1&2, season 3), f2_s2r (trawl Ground B 2006, 2007 all seasons), f3_s2 (trawl Ground C, all seasons), f5_s2 (longline Ground C, season 2), f6_s2 (longline Ground D, season 2), f7_s2 (longline Ground E, season 2), f8_s2 (trawl Ground E, all seasons) , f9_s2 (longline Ground F, season 2), f10_s1 (pot, seasons 1&2). Reference lines are shown at ages 5 and 10.

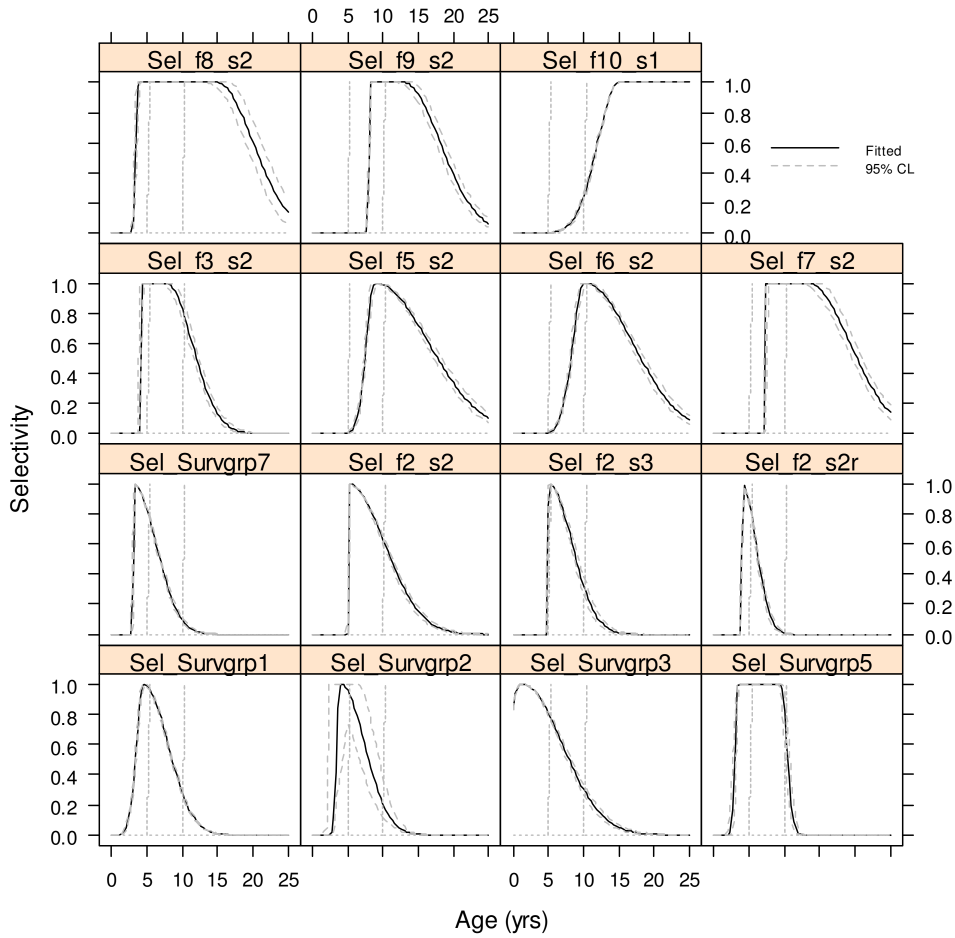


Figure 13. Double-normal-plateau (DNP) and double-normal (DN) fishing selectivity curves from fit of Model 2 showing 95% confidence bounds obtained from the MVN sample. Panel headings: Survgrp1 (survey years 2001, 2002, 2004-2011), Survgrp2 (survey year 1999), Survgrp3 (survey year 1990), Survgrp5 (survey year 1993), Survgrp7 (survey year 2003), f2_s2, f2_s3 (trawl Ground B, seasons 1&2, season 3), f2_s2r (trawl Ground B 2006, 2007 all seasons), f3_s2 (trawl Ground C, all seasons), f5_s2 (longline Ground C, season 2), f6_s2 (longline Ground D, season 2), f7_s2 (longline Ground E, season 2), f8_s2 (trawl Ground E, all seasons) , f9_s2 (longline Ground F, season 2), f10_s1 (pot, seasons 1&2). Reference lines are shown at ages 5 and 10.

Table 2. Estimates of selectivity parameters for Double Normal function for Survey Group 1 and catchability of the other survey groups in CASAL assessments of stock status of *Dissostichus eleginoides* at HIIMI comparing the 2011 assessment model (Model 1) with a model with catch-at-age and abundance-at-age data replaced by catch-at-length and abundance-at-length data (Model 2). s_L, s_U and a_1 relate to the width of the left hand limb of the selectivity ogive, the width of the right hand limb of the selectivity ogive and the age at maximum selectivity respectively, estimated in CASAL (see Bull *et al.,* 2007).

Model	Selectivity parameter estimates, Survey Group 1 (SE)			Survey group *q* estimate[a]			
	s_L	s_U	a_1	Group 3 (1990)	Group 5 (1993)	Group 2 (1999)	Group 7 (2003)
1	0.9449 (0.1006)	4.8183 (0.0908)	3.8640 (0.1290)	0.187	0.187	1.982	0.748
2	1.1755 (0.0638)	4.0402 (0.0523)	4.5056 (0.0904)	0.248	0.248	1.665	0.466

[a]Catchability *q* set to 1 for Survey group 1 (2001, 2002, 2004-2011).

Catch per unit effort has little influence on the model fit. However it was considered informative to see how well the trends in CPUE predicted by CASAL compared to those observed in the fishery. Figure 14 shows that standardised CPUE in the RSTS has been relatively low between 2006 and 2009 and is gradually increasing since, and there is some indication that Model 1 is able to capture this more closely than Model 2.

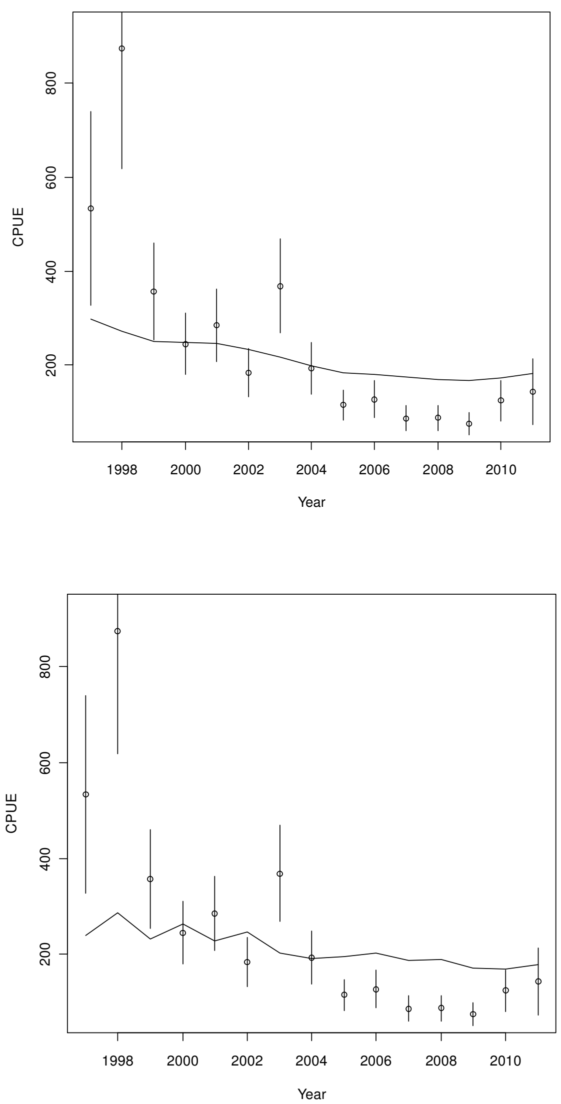


Figure 14. Standardised catch per unit effort series (SE bars shown) and fitted CASAL trend line for the Random Stratified Trawl Surveys from Model 1 (upper panel) and Model 2 (lower panel).

Figure 15 shows the estimated time series of Year Class Strength (YCS) for each of model. Prior to 1984 and after 2008 YCS parameters were fixed to 1.0 in CASAL as YCS would not be expected to be estimated with any degree of confidence from the current dataset, and when YCS for 2009 was estimated it resulted in numerical instability. Random MVN draws of YCS values (based on the parameter estimates from CASAL, combined with the estimate of B_0 (Table 1), as well as parameters which determine the selectivity functions (Figures 12 and 13), combined with the Hessian matrix), were used to determine random recruitments (the product of YCS estimates and median pre-exploitation recruitment (R_0 ; Table 1)). Figure 15 shows box-and-whisker plots of these random recruitments generated from the MVN sample of CASAL model parameter estimates for the year range (1996-2008) for each model.

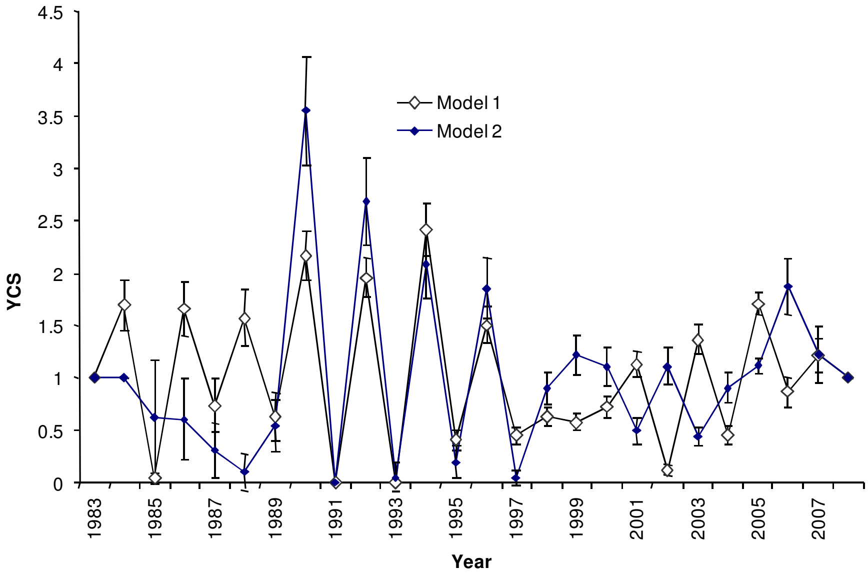


Figure 15. Estimates of Year Class Strength (YCS) estimates (showing +/-SE bars) comparing model 1 (with ageing data) and model 2 (with no ageing data).

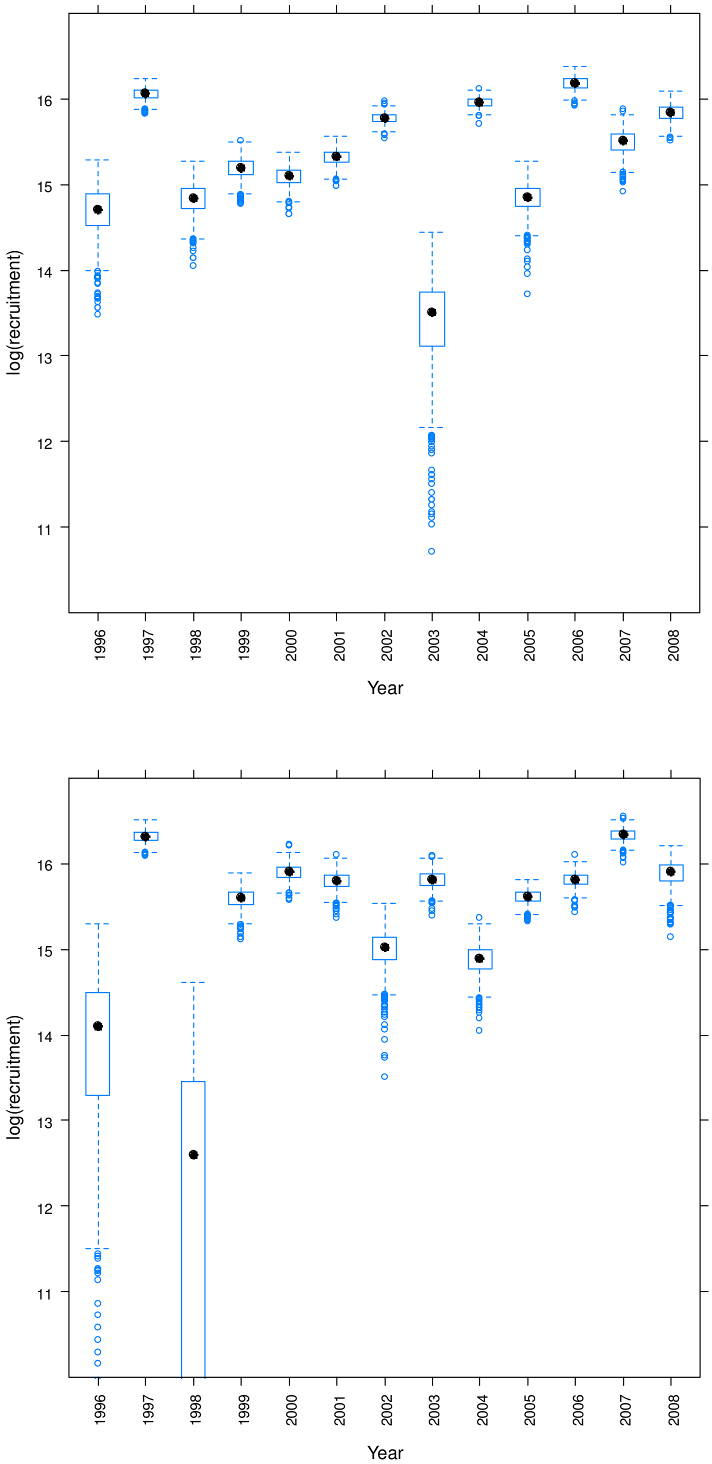


Figure 16. Box plots of estimates of log of recruitment for 1986-2005 with variability generated using 1000 MVN samples for Model 1 (upper panel) and model 2 (lower panel).

Each of the yearly values for numbers of recruits for the 1000 MVN samples were fitted by a LMM to the logarithm of these values using the *asreml* package within R. From the yearly number of recruits for Model 1, the estimate of yearly process error σ_R^2 (*sigma_r*=σ_R) was 0.6053 (SE= 0.247) (i.e. $\sigma_R \equiv CV_R$ = 0.78) while the amount of variance accounted for by within-year estimation error gives a σ^2 component of 0.0865 (SE= 0.001). Future recruitment random lognormal CV was therefore set to 0.78. For Model *2* the estimate of yearly process error σ_R^2 (*sigma_r*=σ_R) was 1.6405 (SE= 0.669) (i.e. $\sigma_R \equiv CV_R$ = 1.2808) while the amount of variance accounted for by within-year estimation error gives a σ^2 component of 0.513 (SE= 0.006). Future recruitment random lognormal CV was therefore set to 1.28.

The future annual catches used in the projection trials were divided amongst the sub-fisheries according to best estimates of future splits in the catch across major subfisheries (Candy and Welsford 2011). Figure 16 shows the summary results for projections for a long-term yield of 2 730 t. The upper panel of Figure 16 shows boxplots of the SSB in terms of status for Model 1, while the lower panel shows the corresponding boxplots for Model 2. The CCAMLR escapement is met in each case with 50.0% and 54.7% median escapement respectively. The CCAMLR depletion rule is also not triggered for either model. However Model 1, with 0.9% of the 1000 trajectories falling below the 20% level has a much lower chance of depletion to Model 2 with 6.5% of the 1000 trajectories falling below the 20% level.

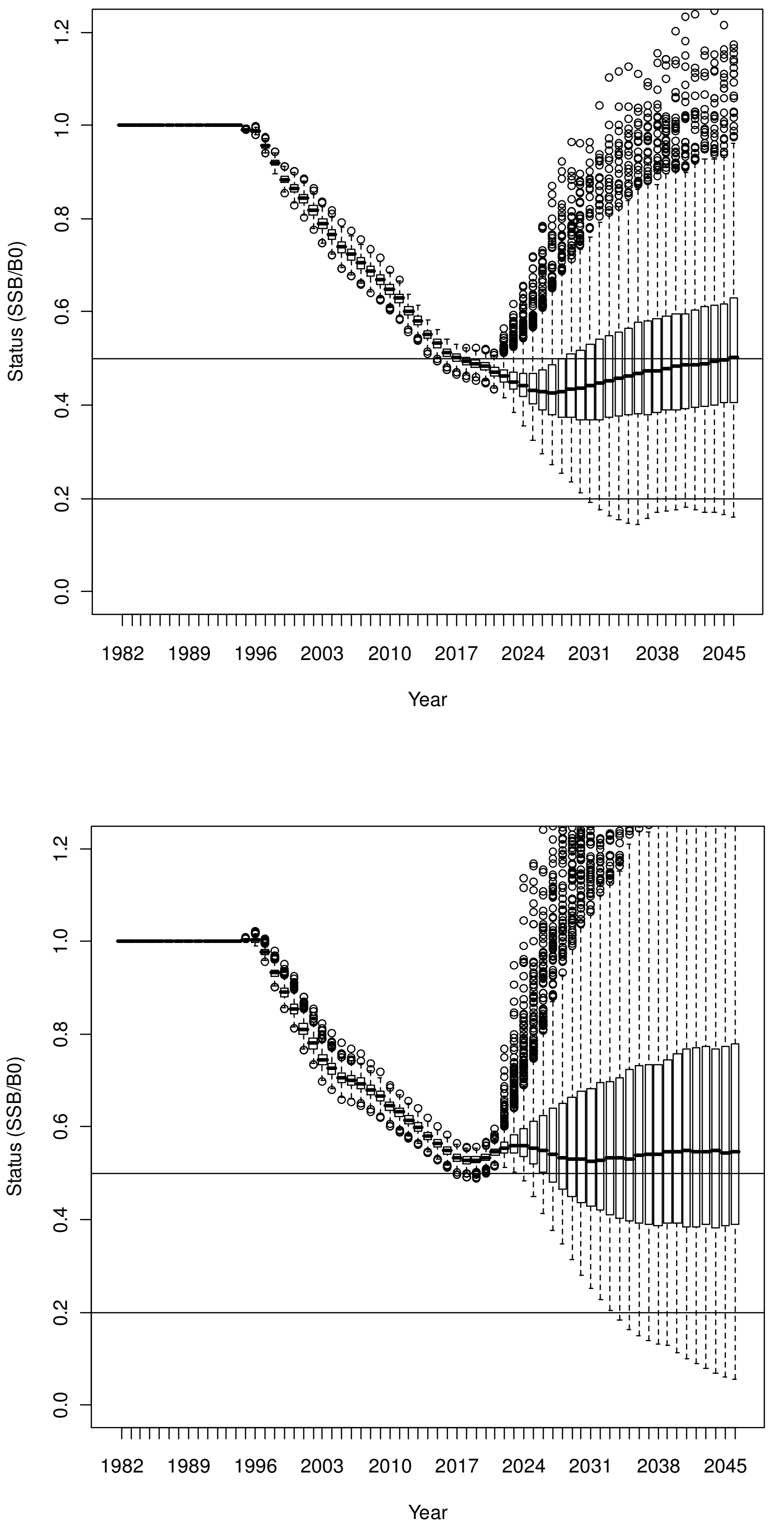


Figure 17. Projection results using future random lognormal recruitment from 2009 with an annual catch of 2 730 tonnes between 2012 and 2046 distributed among sub-fisheries based on best estimates of future expected catches. Boxplots represent the distribution of the status of spawning stock biomass in a trial relative to B_0 in that projection trial across 1000 projection trials for that year (used in CCAMLR decision rules - lines show the 50% and 20% status levels for reference). Upper panel Model 1(escapement= 0.50, depletion= 0.009), lower panel Model 2, (escapement= 0.547, depletion= 0.065).

Discussion

The availability of expanded age datasets for the MITF and HIMI fisheries has had a significant impact on the advice derived from stock assessment models. In the HIMI assessment tested in this study, excluding ageing data gave a higher estimate of pre-fishery median spawning stock biomass, B_0, and corresponding recruitment compared to model 1, by 21% in each case. However the estimated status at 2011 was almost identical (62.9% in model 1 against 63.1% in model 2). These values are key drivers of median stock status projections to 2046, however, the escapement rule gave a higher value for model 2 for an allowable catch of 2 730 t per year, indicating that if this model had been used for management advice a higher, possibly, catch limit may have been recommended if ageing data had not been available.

Another significant effect on allowable catch is due to between-year variation in year class strength estimates with model 2 giving a coefficient of variation of 1.28 compared to 0.78 for Model 1 as calculated for age-1 recruitment for years 1996 to 2008. The depletion rule gave less than 1% of trajectories falling below 20% of B_0 for model 1 while for model 2 this value was 6.5%. Also there was a poor correspondence in YCS estimates between these models for 1999 to 2007 years with high and low years generally occurring one year out of synchrony. The pattern of lower than average YCS seen in mode1 appears more consistent with the pattern of persistent lower catch rates seen during the RSTS during 2006-2009, and is reflected in the better predictions of CPUE by model 1 compared to model 2.

The benefit of the ageing work carried out under this project to the utility of the integrated assessment for toothfish stocks at HIMI and the MITF can therefore be seen to have therefore been considerable. These benefits include:

(i) more realistic estimates of year class strengths in terms of trend and a lower and more realistic estimates of the annual variation in recruitment,

(ii) improved the power of assessment models to reproduce phenomena such as interannual variability in survey catch rates at HIMI;

(iii) the ability to estimate other key parameters such as growth rates of large fish in the MITF; and

(iv) the availability of large ageing datasets has enabled the development and validation of new statistical methods, such as for estimation of ageing error matrices. These methods result in assessment models at HIMI and MITF that more fully incorporate statistical uncertainty, as well have wider application to all other fisheries that use otolith derived age data.

Benefits and adoption

This study has demonstrated the feasibility of collecting large size at age datasets for toothfish using high throughput otolith processing. It has also demonstrated the benefits of including season and gear specific catch at age data into the integrated assessment for the HIMI and MITF toothfish fisheries. These achievements have particular benefit for industry and management stakeholders in these fisheries, as they can have increased confidence that the current assessment frameworks are providing a robust and plausible assessment of stock status and dynamics, while integrating across a range of datasets. This is important during a period when the HIMI has moved from being a trawl focused fishery, targeting a relatively small spatial area to a more spatially extensive multi-gear fishery. Furthermore, as increasing attention is placed on the larger scale linkages between populations of toothfish across the Kerguelen Plateau, the large datasets on the distribution and abundance of different age classes at HIMI will facilitate future analyses to determine the relative impacts of fishing by the Australian, French and illegal vessels in the past

This work also has benefits for the MITF, which has recently moved from a trawl only, tag-based assessment to a multi-gear, spatially structured integrated assessment. The outcomes of this project will assist stock assessment scientists in considering the value of age-based data in such a framework at a relatively early stage of its development in the MITF. Furthermore, the statistical methods developed for statistically representing reader error and effective sample sizes have application in any fishery using integrated assessments and/or age-length keys derived from multiple readers.

A key planned outcome, refinement of integrated stock assessments for toothfish in the HIMI and MITF through the inclusion of age-length data, has benefitted resource managers and industry stakeholders by removing key uncertainties in the data and assessment models used to estimate sustainable catch limits in these fisheries. The age-based integrated assessment models developed using the ageing data from this project are now accepted by both SARAG and CCAMLR as appropriate methods for determining sustainable harvest levels in both the HIMI and MITF fisheries. Furthermore, it is clear from the results of this and other recent studies that gear specific catch at age data will remain a key input into future assessments of both fisheries. This is due to the fact that Patagonian toothfish populations are clearly structured in space, with larger older fish tending to be found in deeper locations, and that the gears used to target toothfish tend to select distinct sections of the population. Furthermore, the fact that as toothfish are long lived (living at least 30 years at HIMI and over 40 years in the MITF) means that the impacts of fishing and extreme recruitment events are likely to continue to influence stock status for decades.

Adoption of the results of this study have been comprehensive; all of the data generated by the project have already been included in the most recent stock assessment for toothfish at HIMI, and much of the data has been or soon will be included in the annual round of stock assessments for the MITF. It is notable that the success of this project was based upon a high level of industry and AFMA support, as well as collaboration between stock assessment groups at AAD and CSIRO. This gives a strong indication of the commitment that research providers, industry, management and stakeholders have to full adoption of the implication of this project for evaluating data collection and assessment strategies and management advice based on its outcomes.

The ongoing collection of otoliths, in each season and across all size classes captured by the different sectors of the fishery represents a cost-effective use of onboard sampling efforts. Sampling by length bin is shown to be a robust method of collecting sufficient otoliths to allow an appropriate number of otoliths to be available for future ageing studies, while balancing the time required for observers to complete other important data collection task at sea.

Further development

Several aspects of this project would benefit from further development, but are outside the scope or timeframe of this project. These include:

Targeted otolith analyses of under-represented age classes

Age-length datasets from the largest and smallest fish in the population are relatively small, particularly for MITF, where longline fishing that selects larger older fish is a relatively recent development in this fishery. Processing and analyzing otoliths from under-represented parts of the population captured by future fishing activity would be likely to provide multiple observations of year age classes as they move through the fishery, will improve the precision of estimate of fishing selectivity and year class strengths. Appendix 7 includes a cost breakdown of processing otoliths using the AAD protocol to assist in developing budgets to process future otolith collections.

Use of ancillary data to improve ageing precision

Covariates of age such as otolith weight, which are available for nearly all fish sampled in this project, has been used in other studies to improve the precision of allocating individuals to age classes (Francis *et al.*, 2005). With the dataset now available, exploring methods of using otolith weight, as well as other covariates such as sex has the potential to make age length keys more precise.

International inter-laboratory comparisons

Outside of Australia, at least two other research groups routinely provide ageing data for inclusion in assessments of Patagonian toothfish fisheries, particularly in the South Atlantic. A useful further development of this work would be to exchange reference collections with other laboratories and develop error matrices which can be incorporated into other toothfish assessments using the method developed by, such as those conducted by CCAMLR. Strong relationships have also been established during this project between AAD and NIWA in New Zealand, which has the potential to expand the data available for assessment to Antarctic toothfish, as describe in Appendix 8.

Conclusion

As noted above, large quantities of contemporary and archived otoliths have been aged across an unprecedentedly broad range of seasons, gears and fish sizes from commercial and research fishing at HIMI and the MITF, thereby achieving objectives 1 and 2.

This study has demonstrated that the inclusion of age-length data results in significant refinements to the estimates of several key parameters in these assessments. These include more precise estimates of the level and variability of recruitment of juvenile toothfish to the stock relative to when size at age is estimated using a von Bertalanffy growth function alone. This highlights the value of ageing data, drawn from a broad range of seasons and all gear types used in the fishery to date, in producing precise and plausible results under the assessment framework used for the HIMI toothfish fishery and the MITF.

This project has also achieved its other objectives, relating to the development of robust and effect otolith collection, processing and interpretation. High-quality, high-throughput ageing of toothfish otoliths is shown to be feasible, such that season and gear specific age-length keys could be routinely incorporated into the HIMI assessment and MITF assessments with less than one year lag. Otoliths collection should continue across all gear types in both fisheries toothfish fishery. This is because it is clear that each gear type samples a different part of the population, and both HIMI and MITF move from being predominantly trawl to fisheries that use longline to target larger older cohorts, many of which are likely to have been vulnerable to the trawl fishery in the past.

A firm basis is therefore provided for stock assessments scientists, fishers and resource managers to see the benefits of including season and gear specific ageing data as key input into the integrated assessments at HIMI and MITF into the future, to maintain confidence in the current stock assessment approach, as well as providing statistical tools that can be readily applied in other fisheries were catch at age data or integrated assessments are used.

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Appendix 1. Intellectual Property

No intellectual property is identified as arising from this project.

The dataset generated from this project is housed in a secure database at the AAD. A metadata record describing the datasets and terms of use has been lodged with Australian Antarctic Data Centre (http://data.aad.gov.au/).

Appendix 2. Staff

Dr Steve Candy

Mr Joseph Hutchins

Mr Tim Lamb

Ms Gabrielle Nowara

Mr Jeremy Verdouw

Dr Dirk Welsford

Appendix 3. Distribution of Patagonian toothfish otolith samples by year and total length subsampled during this study

Table A3.1 Otoliths collected from Random Stratified Trawl Surveys at Heard Island and the McDonald Islands, 2008-2011. Note two surveys were conducted in 2010.

	Year				
Length Bin (mm)	**2008**	**2009**	**2010a**	**2010b**	**2011**
300-349	1	5	1	-	-
350-399	2	11	19	7	-
400-449	8	69	143	66	22
450-499	80	197	340	196	77
500-549	197	135	281	166	89
550-599	210	185	201	137	73
600-649	186	132	126	107	60
650-699	132	78	91	115	54
700749	73	41	61	70	46
750-799	43	28	43	67	34
800-849	16	20	19	27	26
850-899	16	6	9	25	14
900-949	10	3	4	17	11
950-999	7	1	4	7	6
1000-1049	4	4	1	5	1
1050-1099	3	2	2	5	4
1100-1149	1	1	-	2	2
1150-1199	2	1	1	4	2
1200-1249	1	-	1	-	1
1250-1299	-	-	-	-	-
1300-1349	-	-	-	1	1
1350+	-	-	-	2	-
Total	992	919	1347	1026	523

Table A3.2 Otoliths collected from commercial trawl fishing operations at Macquarie Island, 2000-2009. Note no fishing occurred in 2003 and no otoliths were collected in 2005.

	Year							
Length Bin (mm)	**2000**	**2001**	**2002**	**2004**	**2006**	**2007**	**2008**	**2009**
300-349	-	-	-	-	-	-	-	6
350-399	2	-	-	1	3	1	-	-
400-449	13	-	-	-	9	4	2	-
450-499	22	1	-	4	29	3	1	15
500-549	42	14	-	32	46	8	10	26
550-599	37	38	-	52	51	5	52	35
600-649	56	23	4	53	61	6	70	47
650-699	43	10	19	59	58	5	73	32
700749	49	10	11	58	65	5	63	31
750-799	39	9	9	63	65	-	60	35
800-849	25	6	11	52	58	-	54	39
850-899	13	3	6	52	57	-	52	35
900-949	9	4	4	52	57	-	27	41
950-999	5	2	3	57	53	-	-	31
1000-1049	1	-	2	51	40	-	-	30
1050-1099	1	-	-	40	8	-	-	5
1100-1149	2	-	1	11	2	-	-	3
1150-1199	-	-	1	15	1	-	-	-
1200-1249	-	-	-	10	-	-	-	-
1250-1299	1	-	-	4	-	-	-	-
1300-1349	-	-	-	3	-	-	-	-
1350+	1	-	-	3	-	-	-	-
Total	361	120	71	671	663	37	464	411

Table A3.3 Otoliths collected from experimental and commercial longline fishing operations at Macquarie Island, 2007-2010.

	Year			
Length Bin (mm)	**2007**	**2008**	**2009**	**2010**
400-449	1	-	-	-
450-499	1	-	2	-
500-549	5	-	1	1
550-599	7	-	9	6
600-649	13	-	14	6
650-699	19	3	40	17
700749	26	5	32	30
750-799	48	12	35	49
800-849	39	28	39	39
850-899	50	35	51	56
900-949	40	35	47	71
950-999	35	36	49	72
1000-1049	18	29	49	60
1050-1099	28	33	46	53
1100-1149	16	28	31	32
1150-1199	25	29	30	16
1200-1249	18	27	15	10
1250-1299	11	16	17	7
1300-1349	5	15	16	3
1350-1399	8	18	11	1
1400-1449	9	6	6	5
1450-1499	2	4	3	3
1500-1549	2	2	3	1
1550-1599	1	1	1	-
1600-1649	-	2	-	1
1650-1699	2	1	1	2
1700+	2	1	-	2
Total	431	366	648	543

Appendix 4. Assessments of within and between reader bias in repeated readings of the AAD Patagonian toothfish otolith reference collection.

Accurately interpreting the structures within otoliths, and ensuring consistency across large datasets requires a great deal of technical skill as well as strict adherence to the protocols developed at the AAD for this purpose (Nowara *et al.*, 2009). To assess consistency in the way that otoliths were read throughout the duration of this project, and with otolith reads conducted in previous projects (Welsford et al. 2009; Candy *et al.* 2011), the AAD Patagonian toothfish otolith reference collection[3] was read on three separate occasions by both of the project staff that read all of the otoliths used in this project. These reads were then assessed to evaluate any drift or bias in the way otoliths were being interpreted. Age bias plots (Campana *et al.* 1995; Campana 2001) were used as the primary method for assessing consistency, both between readers and between readings of the reference collection by the same reader. Plots were scrutinized for evidence of systematic bias, where a fitted linear relationship would have a different slope and/or have a different intercept to a 1:1 line. Spread of points around the fitted line was also scrutinised to ensure that no pattern ageing errors was evident e.g. a fanning out of points from left to right may indicate that older fish are more difficult to age consistently than younger fish.

First reading versus readings from previous projects

At the commencement of the project, the project staff read the reference collection. Comparisons between these reads and those obtained in previous projects showed very good consistency (Figure A4.1), and also indicated minimal bias between readers (Figure A.4.2).

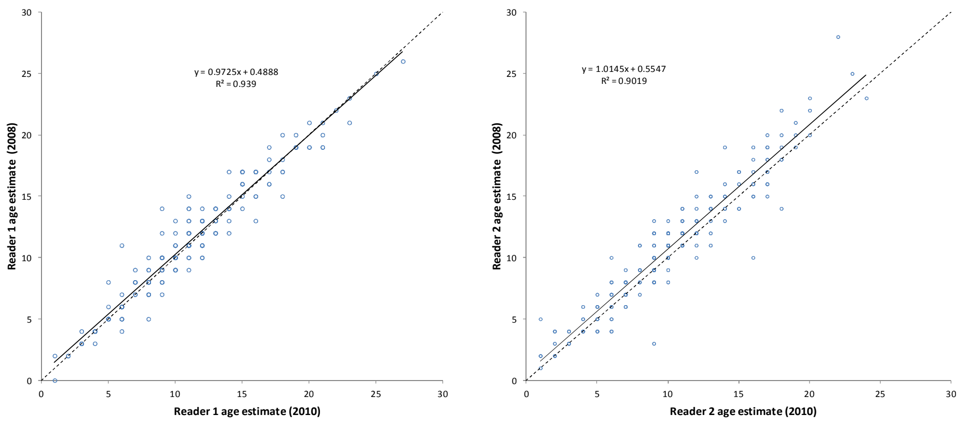

Figure A4.1. Within-reader Age bias plots comparing age estimates for the AAD toothfish reference collection from 2008 and 2010 as read by reader 1 (left panel) and reader 2 (right panel). Note that the fitted relationships (solid lines) are close to a 1:1 relationship (broken lines), indicating minimal systematic bias between 2008 and 2010 reads.

[3] The AAD otolith reference collection consists of 200 otoliths, selected from across a wide range of ages (1-25 years old, with each age estimate initially agreed by consensus between the readers) and incorporates fish from both the Heard Island and McDonald Islands, and Macquarie Island toothfish fisheries. During each reading session, the otoliths are randomised and read blind.

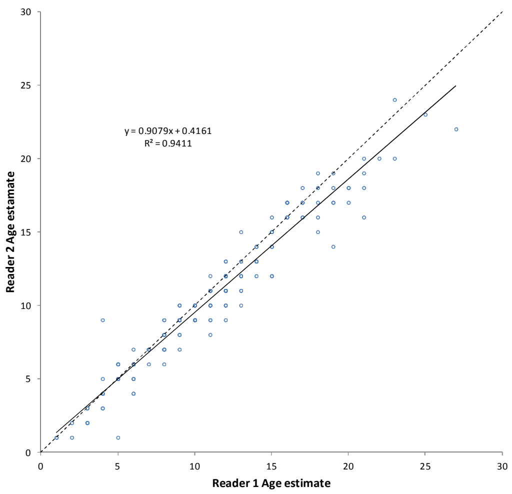

Figure A4.2. Age bias plots comparing age estimates for the AAD toothfish reference collection read by reader 1 and reader 2 in 2010. Note that the fitted relationship (solid line) are close to a 1:1 relationship (broken line), indicating no evidence of systematic bias between reader 1 and 2.

Repeat readings of the reference collection during the project

Two more readings of the reference collection where conducted; during the main production run of the project and at the end of the production run. In all instances, all age bias plots showed a good level of consistency within and between readers (Figures A4.3-A4.6).

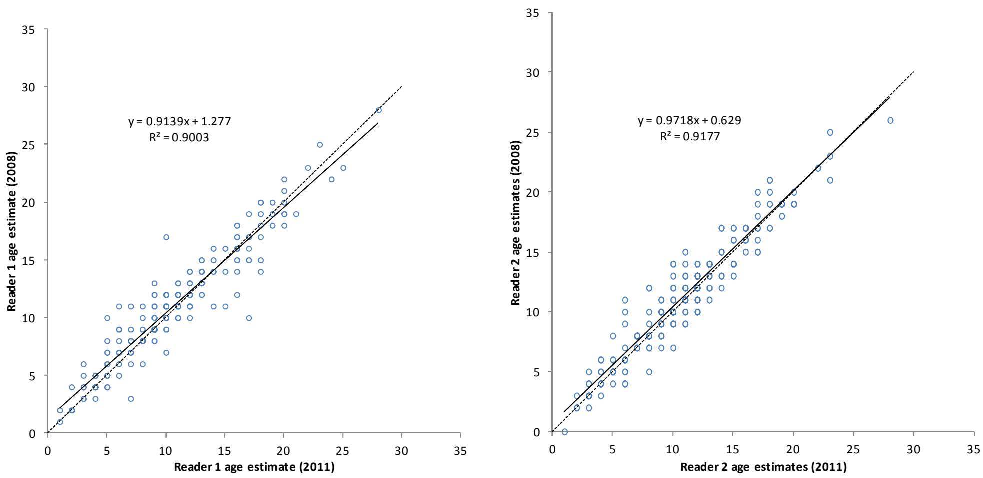

Figure A4.3. Within-reader Age bias plots comparing age estimates for the AAD toothfish reference collection from 2008 and February 2011 as read by reader 1 (left panel) and reader 2 (right panel). Note that the fitted relationships (solid lines) are close to a 1:1 relationship (broken lines), indicating no systematic bias between 2008 and February 2011 reads.

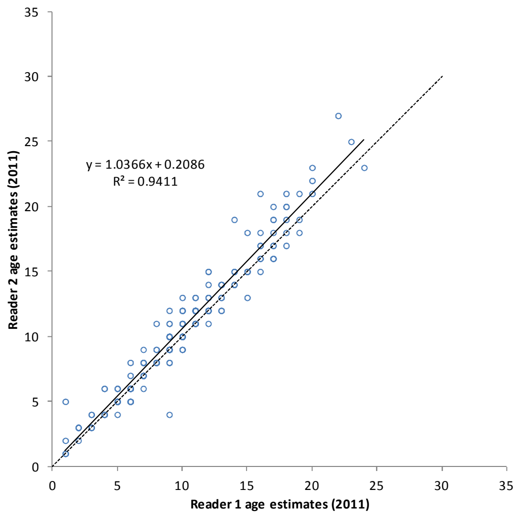

Figure A4.4. Age bias plots comparing age estimates for the AAD toothfish reference collection read by reader 1 and reader 2 in February 2011. Note that the fitted relationship (solid line) is close to a 1:1 relationship (broken line), indicating minimal systematic bias between reader 1 and 2.

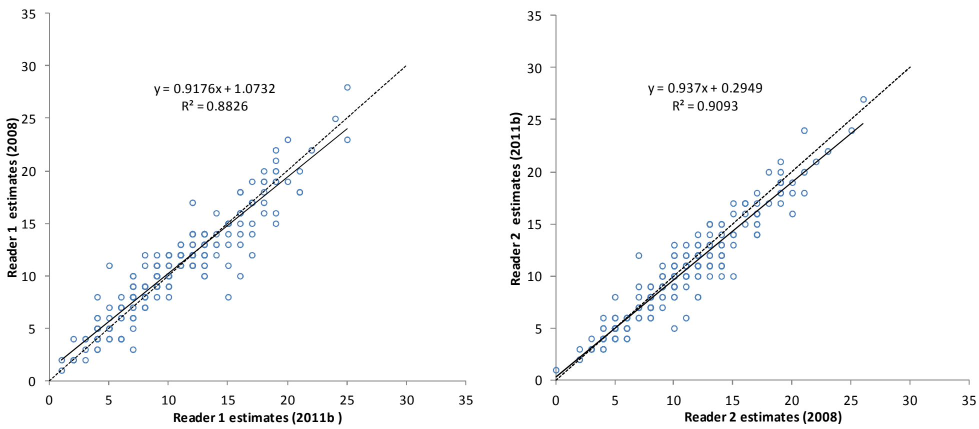

Figure A4.5. Within-reader Age bias plots comparing age estimates for the AAD toothfish reference collection from 2008 and September 2011 as read by reader 1 (left panel) and reader 2 (right panel). Note that the fitted relationships (solid lines) are close to a 1:1 relationship (broken lines), indicating no systematic bias between 2008 and September 2011 reads.

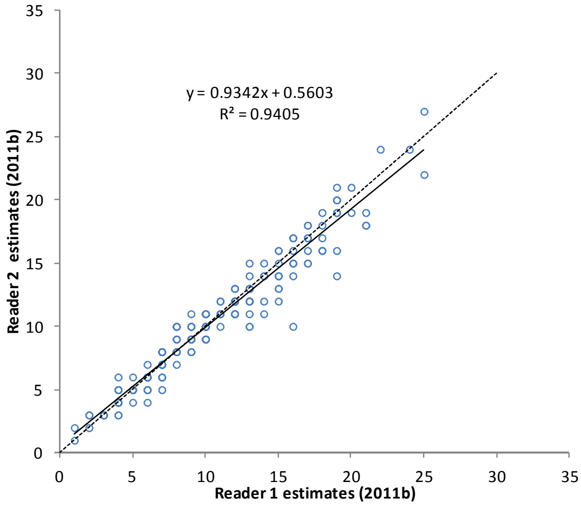

Figure A4.6. Age bias plots comparing age estimates for the AAD toothfish reference collection read by reader 1 and reader 2 in September 2011. Note that the fitted relationship (solid line) is close to a 1:1 relationship (broken line), indicating minimal systematic bias between reader 1 and 2.

The overall high consistency between readers is also shown in the fact that, in the February 2010 readings of the reference collection, more 90% of reads were less than 1 year different, and in greater than 50% of reads both readers got an identical result (Figure A4.7). Otoliths of low readability showed a higher mean level of difference than otoliths of high readability, as has been noted previously (Welsford *et al.* 2009) (Figure A4.8), however the age of the otolith had no consistent effect on the level of difference between reads (Figure A4.9). However, overall there was confidence that ages produced in the project would be suitable for adding to existing AAD datasets of toothfish age-length data without introducing any systematic errors.

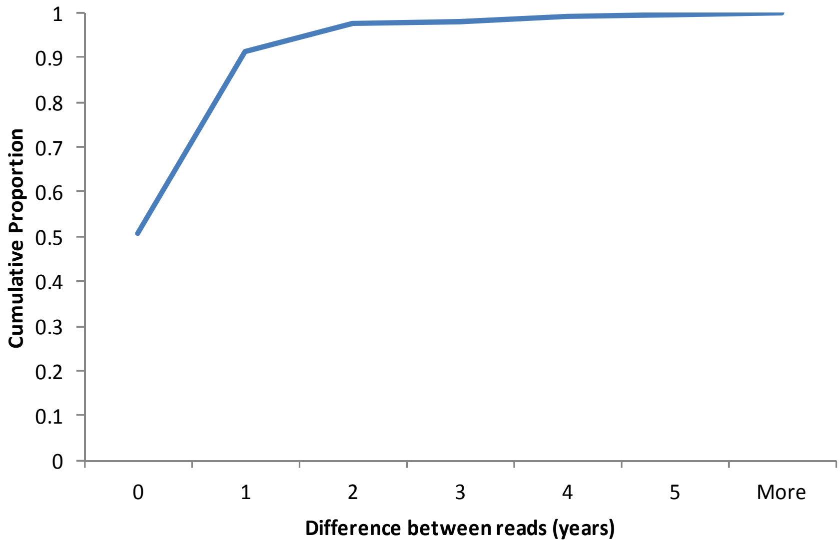

Figure A4.7. Cumulative proportion of reference collection otoliths read by reader 1 and reader 2 by the net difference in estimated age for the reference collection read in February 2010.

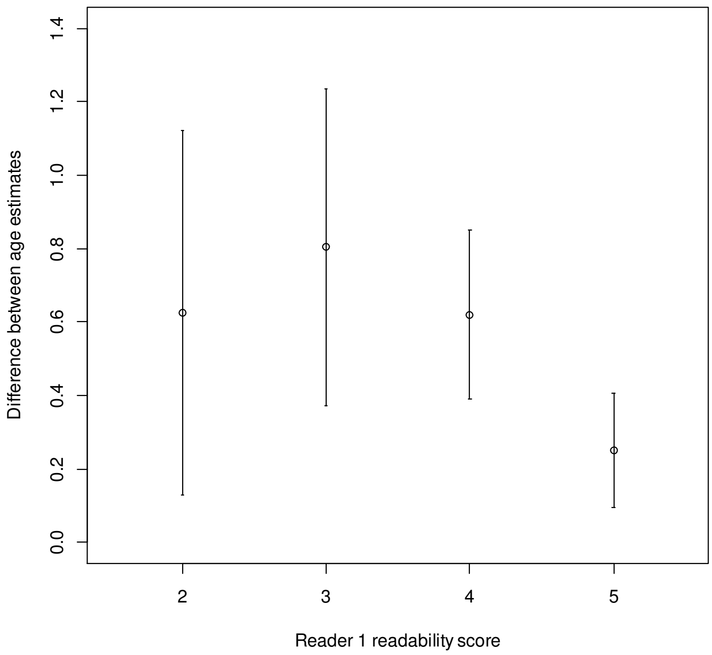

Figure A4.8. Mean difference (with standard error bars) between readers by readability score (2: poor to 5: very good) for the reference collection read in February 2010.

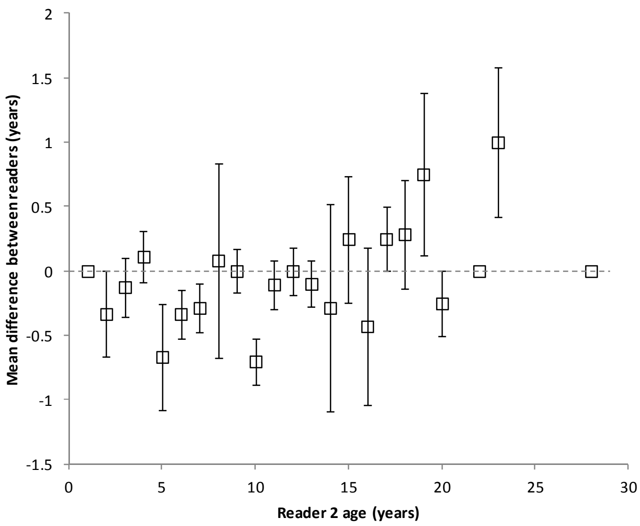

Figure A4.9. Mean difference (with standard error bars) between readers by age estimate for the reference collection read in February 2010.

Appendix 5. Calculation of the coefficient of variation (CV) of abundance-at-age data

If the estimated (i.e. 'observation' in CASAL) proportion of the survey catch of age *a* for a particular year is given by, p_a, from the application of an ALK [see equation (1) of Candy (2009)], and the corresponding estimate of the population size vulnerable to the survey is given by $\tilde{N}$ then the estimated abundance (i.e. 'observation' in CASAL) of age *a* fish is given by

$N_a = \tilde{N} p_a$. If the expected value of p_a and $\tilde{N}$ are given by μ_{p_a} and $\mu_{\tilde{N}}$, respectively and given N_a can be expressed (exactly) by a second-order Taylor series expansion about $\theta = \left(\mu_{p_a}, \mu_{\tilde{N}}\right)$ as

$$N_a = \mu_{p_a}\mu_{\tilde{N}} + \mu_{\tilde{N}}\left(p_a - \mu_{p_a}\right) + \mu_{p_a}\left(\tilde{N} - \mu_{\tilde{N}}\right) + \left(p_a - \mu_{p_a}\right)\left(\tilde{N} - \mu_{\tilde{N}}\right)$$

then

$$E\left(N_a\right) = \mu_{p_a}\mu_{\tilde{N}} \text{ and}$$

$$Var\left(N_a\right) \cong \mu_{\tilde{N}}^2 Var\left(p_a\right) + \mu_{p_a}^2 Var\left(\tilde{N}\right) + Var\left(p_a\right) Var\left(\tilde{N}\right)$$

assuming $Cov\left(p_a, \tilde{N}\right) \equiv 0$.

The variance of p_a is given by $Var\left(p_a\right) = \mu_a\left(1-\mu_a\right)/n'$ where n' is the effective sample size obtained using the method described by Candy (2009) for accounting for between-haul heterogeneity in commercial catch-at-length proportions, ALK sampling error, and random ageing error. The estimate of p_a was obtained using the standard non-parametric ALK method [equation (1) of Candy (2009)].

The only difference in calculation of n' for the survey data, compared to that for the commercial catch-at-length proportions (Candy, 2009), is that the effective sample size for the proportion-at-length data was calculated separately for each stratum (i.e. accounting for between-shot within stratum heterogeneity) and then these values were accumulated across strata to give an overall ESS for survey catch-at-length proportions. In addition, the catch-at-length proportions were obtained as stratum-area weighted estimates. The variance of $\tilde{N}$ was obtained using the standard stratified random sampling estimate (Cochran, 1977). Finally, the estimates for μ_{p_a} and $\mu_{\tilde{N}}$ of p_a and $\tilde{N}$, respectively, were substituted into the above variance formulae and the resulting estimate of the variance of N_a was expressed as a coefficient of variation for input to CASAL.

Appendix 6. Weighted average ageing error matrix E

True Age	Read Age																																		
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35
1	0.470	0.451	0.064	0.012	0.001	0.001	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
2	0.227	0.459	0.227	0.070	0.013	0.001	0.001	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
3	0.038	0.229	0.449	0.229	0.038	0.015	0.002	0.002	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
4	0.009	0.046	0.258	0.439	0.201	0.036	0.007	0.002	0.002	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
5	0.001	0.011	0.052	0.274	0.429	0.187	0.035	0.008	0.001	0.002	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
6	0.002	0.002	0.013	0.057	0.282	0.418	0.179	0.036	0.008	0.001	0.001	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
7	0.0	0.002	0.002	0.014	0.062	0.284	0.408	0.178	0.039	0.009	0.001	0.001	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
8	0.0	0.0	0.002	0.002	0.015	0.065	0.280	0.398	0.181	0.042	0.010	0.002	0.002	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
9	0.0	0.0	0.0	0.003	0.003	0.016	0.068	0.272	0.388	0.188	0.047	0.011	0.002	0.002	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
10	0.0	0.0	0.0	0.0	0.003	0.003	0.017	0.070	0.260	0.379	0.198	0.053	0.013	0.002	0.002	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
11	0.0	0.0	0.0	0.0	0.0	0.003	0.004	0.018	0.070	0.245	0.369	0.210	0.060	0.015	0.003	0.003	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
12	0.0	0.0	0.0	0.0	0.0	0.0	0.004	0.004	0.018	0.070	0.227	0.359	0.224	0.069	0.018	0.004	0.004	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
13	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.004	0.004	0.018	0.069	0.209	0.350	0.238	0.078	0.021	0.005	0.004	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
14	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.004	0.005	0.018	0.067	0.190	0.340	0.252	0.088	0.024	0.006	0.005	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
15	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.004	0.005	0.018	0.065	0.173	0.331	0.263	0.099	0.028	0.008	0.006	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
16	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.005	0.005	0.018	0.063	0.158	0.322	0.272	0.109	0.031	0.009	0.008	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
17	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.005	0.006	0.018	0.062	0.146	0.313	0.277	0.119	0.035	0.011	0.009	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
18	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.005	0.006	0.019	0.062	0.136	0.304	0.279	0.127	0.038	0.013	0.011	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
19	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.006	0.007	0.019	0.064	0.130	0.295	0.276	0.134	0.041	0.015	0.012	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
20	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.006	0.008	0.021	0.067	0.128	0.287	0.269	0.140	0.043	0.017	0.014	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
21	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.008	0.010	0.023	0.072	0.131	0.278	0.257	0.142	0.045	0.019	0.015	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
22	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.009	0.012	0.026	0.082	0.138	0.270	0.240	0.141	0.045	0.021	0.016	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
23	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.011	0.015	0.031	0.095	0.151	0.262	0.216	0.136	0.044	0.022	0.016	0.0	0.0	0.0	0.0	0.0	0.0	0.0
24	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.015	0.020	0.037	0.114	0.170	0.254	0.186	0.124	0.041	0.022	0.016	0.0	0.0	0.0	0.0	0.0	0.0
25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.017	0.024	0.041	0.123	0.173	0.246	0.173	0.123	0.041	0.024	0.017	0.0	0.0	0.0	0.0	0.0
26	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.019	0.027	0.042	0.126	0.167	0.239	0.167	0.126	0.042	0.027	0.019	0.0	0.0	0.0	0.0
27	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.021	0.030	0.044	0.129	0.161	0.231	0.161	0.129	0.044	0.030	0.021	0.0	0.0	0.0
28	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.023	0.033	0.045	0.132	0.155	0.224	0.155	0.132	0.045	0.033	0.023	0.0	0.0
29	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.025	0.037	0.047	0.135	0.149	0.217	0.149	0.135	0.047	0.037	0.025	0.0
30	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.027	0.040	0.048	0.137	0.143	0.210	0.143	0.137	0.048	0.040	0.027
31	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.058	0.044	0.049	0.140	0.137	0.203	0.137	0.140	0.049	0.044
32	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.062	0.097	0.050	0.141	0.131	0.196	0.131	0.141	0.050
33	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.067	0.106	0.101	0.143	0.125	0.190	0.125	0.143
34	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.071	0.116	0.102	0.289	0.119	0.183	0.119
35	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.076	0.126	0.103	0.291	0.227	0.177

Appendix 7. Numbers of fish aged by sub-fishery and year, length frequency sample sizes, and effective sample sizes for catch-at-length and catch-at-age proportions for the Heard Island and McDonald Islands toothfish fishery.

Table A7.1 Number of toothfish aged and used in the revised HIMI assessment by sub-fishery and year. f1=Trawl survey; f2= Trawl, Ground B; f3= Trawl, Ground C, f5= Longline Ground C, f6 = long line Ground D, f7 = Longline Ground E, f8= Trawl Ground F, f9 = Longline Ground F, f10 = Pot.

	Year														
CASAL Sub-fishery	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	Sum
f1	0	2	21	2	4	14	4	1	121	550	651	641	917	520	3448
f2	90	559	725	787	673	420	329	287	303	234	48	0	0	8	4472
f3	77	9	39	13	117	45	8	64	215	1	12	0	0	0	601
f5	0	0	0	0	0	80	1	5	103	221	27	0	0	0	437
f6	0	0	0	0	0	101	1	10	235	153	13				513
f7	0	0	0	0	0	0	0	0	0	1	0				1
f8	119	56	54	118	47	31	1	1	3	1	6	1	0	6	489
f9	0	0	0	0	0	0	0	5	76	5	2				93
f10	0	0	0	0	0	0	0	0	176	0	0				176
Sum	286	626	839	920	841	691	344	373	1232	1166	759	642	917	534	10230

Table A7.2 Number of toothfish measured for length and used in the revised HIMI assessment by sub-fishery and year. f1=Trawl survey; f2= Trawl, Ground B; f3= Trawl, Ground C, f5= Longline Ground C, f6 = long line Ground D, f7 = Longline Ground E, f8= Trawl Ground F, f9 = Longline Ground F, f10 = Pot.

CASAL Sub-fishery	Year														
	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	Sum
f1	0	0	0	0	0	0	0	0	0	2050	1281	1922	5930	2484	13667
f2	8328	13932	19095	22561	14036	17420	16707	11571	11540	12967	10459	10034	4527	2814	175991
f3	2177	517	976	1513	4192	2597	2321	3938	3231	265	2347	1566	1404	1898	28942
f5	0	0	0	0	0	1696	4793	2799	5487	3996	6339	4918	3017	0	33045
f6	0	0	0	0	0	2498	7620	6974	3514	1556	7385	11159	440	0	41146
f7	0	0	0	0	0	0	0	0	0	733	1154	581	0	4014	6482
f8	557	0	260	881	0	93	110	131	154	826	2012	1394	5454	120	11992
f9	0	0	0	0	0	0	1550	3121	896	2362	1564	13684	5188	0	28365
f10	0	0	0	0	0	0	0	0	5888	0	0	782	1500	2264	10434
Sum	11062	14449	20331	24955	18228	24304	33101	28534	30710	24755	32541	46040	27460	13594	350064

Table A7.3 Effective sample size for catch-at-age proportions (to 2008) and catch-at-length proportions (2009-2011) (numbers in brackets are result of down-weighting for process error) for sub-fishery, season, and year combinations used in model 1. CASAL sub-fisheries as described in Table A7.1. Season s1 (1 Dec - 30 April), s2 (1 May - 30 Sep), s3 (1 Oct - 30 Nov).

CASAL Sub-fishery_ season	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011
f2_s1		896 (288)	1536 (493)	2031 (653)	290 (94)	274 (88)	666 (214)	334 (107)	357 (115)	1114 (358)	650 (209)	1182 (221)	3034 (567)	1914 (358)
f2_s2	1208 (200)	889 (147)	1660 (274)	1923 (318)	1495 (248)	675 (112)	743 (123)	290 (48)	2498 (413)	2107 (348)	1096 (182)	2306 (200[1])		145 (145)
f2_s3	269 (70)		666 (172)	1395 (360)	1670 (431)	550 (142)	561 (145)		1962 (507)	1066 (275)	2426 (626)	1446 (200[1])		
f3_s1		79 (79)	225 (225)				50 (50)	141 (141)	223 (223)	37 (37)		80 (70)	149 (131)	736 (646)
f3_s2	531 (170)			405 (131)	2072 (666)	641 (206)	551 (176)	478 (153)	933 (300)	3 (1)	1440 (462)	770 (200[1])		
f3_s3	559 (460)			301 (248)			362 (297)		595 (490)	55 (46)	551 (453)	132 (132)		500 (500)
f5_s2						653 (357)	658 (360)	206 (113)	2925 (1601)	2015 (1103)	2593 (1419)	2128 (551)	2629 (681)	
f6_s2						817 (341)	654 (273)	250 (105)	2777 (1159)	1372 (572)	2206 (921)	3539 (200[1])	214 (214)	
f7_s2										643 (643)		411 (411)		2626 (400)
f8_s1			257 (114)	220 (171)			96 (74)			83 (65)	240 (186)	293 (210)	135 (97)	42 (30)
f8_s2	179 (73)		37 (15)	73 (30)		76 (31)		36 (15)	40 (40)	130 (53)	117 (47)	393 (129)	845 (275)	
f8_s3				178 (178)						275 (275)	1004 (200)	53 (39)	1587 (200[1])	
f9_s2							644 (417)	246 (159)	603 (390)	1725 (400[1])	1379 (892)	3970 (200[1])	3792 (200[1])	
f10_s1									2029 (200[1])			782 (126)		1332 (553)
f10_s2									245 (245)					

[1] Value assigned not calculated by method of Candy (2008)

Appendix 8. Cost breakdown of otolith processing and ageing

Table A8.1. List of consumables required to process toothfish otoliths at AAD. Cost is shown per 1000 otoliths.

Consumables	Price	Amount	Cost/1000 Otoliths
Microscope slides to mount sections on (25x75mm)	$240	1000	$240.00
Slide coverslips (22x50mm)	$77	1000	$77.00
Slide boxes for storage (Capacity 100)	$16	1	$160.00
Polyester resin for mounting	$70	4000mL	$35.00
Epoxy resin for embedding	$130	5000mL	$104.00
3 mL plastic pipettes for transferring resin	$65	1000	$5.40
Wooden stirrers for mixing resin	$50	1000	$4.15
70 mL specimen jars for mixing polyester resin	$50	250	$2.00
250 mL plastic jars for mixing epoxy resin	$90	100	$4.50
30 mL syringes for transferring epoxy resin	$55	50	$22.00
1 mL syringes for adding catalyst to resin	$20	100	$2.00
Ease release for lining ice cube tray moulds	$30	500g	$0.01
Disposable nylex gloves	$30	100	$6.00
Superfine point permanent marker pens	$3	1	$3.00
		Total	**$665.06**

Table A8.2. Time breakdown of otolith processing tasks (per 1000 otoliths) for a single technician.

Process	Processing Time per 1000 Otoliths (Weeks)
Data retrieval	0.4
Weighing	2.6
Embedding	2.2
Marking	1.2
Sectioning	6.4
Mounting	3.6
Image capturing	3.6
Ageing	3.6
Training. quality assurance	2.4
Total	26

Table A8.3. Processing costs per otolith. Assumes salary and on-costs totalling $70,000 for a full time technician.

Processing Cost per Otolith	
Labour cost	$35.00
Consumables cost	$0.67
Total cost per otolith	**$35.67**

Appendix 9. Investigation of ageing methods for Antarctic toothfish (*Dissostichus mawsoni*) captured from Australian vessels in CCAMLR exploratory fisheries.

Introduction

Dissostichus mawsoni is the primary species targeted in longline fisheries in the Southern Ocean in the areas of Australia's Antarctic Territory. Antarctic toothfish otoliths have otolith morphology that is quite distinct form that of Patagonian toothfish (*D. eleginoides*). This document summarises a comparison of ageing methods currently employed by New Zealand scientists form the National Institute of Water and Atmospheric Research (NIWA), who routinely age Antarctic toothfish from the Ross Sea, and the method used by AAD for Patagonian toothfish, to determine suitable methods for high throughput processing of Antarctic toothfish if required in future; for example if Australian vessels become more active in CCAMLR exploratory fisheries.

Processing methods

There are two methods currently used to process *D. mawsoni* otoliths for ageing; the bake, embed and section method as used by NIWA, and the multiple thin section method as used by the AAD. The method which provides the most easily interpretable otolith sections is multiple thin sectioning. This method involves embedding whole otoliths (left or right) in individual epoxy resin blocks. The location of the primordium on the otolith is marked on the block as well as a cutting line transverse through the sulcal groove. The embedded otoliths are then sectioned using a low speed diamond blade saw. Three sections are cut to ensure at least one of them includes the primordium of the otolith. The thin sections (0.35mm) are mounted onto a microscope slide using polyester resin after which they are viewed under transmitted light on a dissecting microscope. The best section is located and an image is captured from which an age estimate can be made. This is the same method used by the AAD to process *D. eleginoides* otoliths (For further details see Nowara *et al.* 2009).

Ageing

<u>*D.mawsoni* otolith section - features</u>

When viewed under transmitted light, *D.mawsoni* otolith sections usually have a dark inner area which in most cases consists of the first 4 to 7 annuli or rings. Outside the dark inner zone, the increments become narrower and more evenly spaced and it may be easier to count increments towards the proximal surface. These outer zones often have split zones, where a single zone appears to separate into two zones at a particular point along the ring. If split zones are encountered, the reader should count the zone as a single zone unless the zones stay separated right around the otolith section.

<u>*D.mawsoni* otolith section - interpretation</u>

Interpretation of *D.mawsoni* otoliths to obtain an estimate of age differs to that of *D.eleginoides* in a few distinct ways. Unlike *D.eleginoides* otoliths, the earliest increments on a *D.mawsoni* section are small and compressed on the ventral lobe and can be difficult to distinguish (Figure A7.1). However, when viewed on the dorsal side the early rings appear darker and are easier to identify.

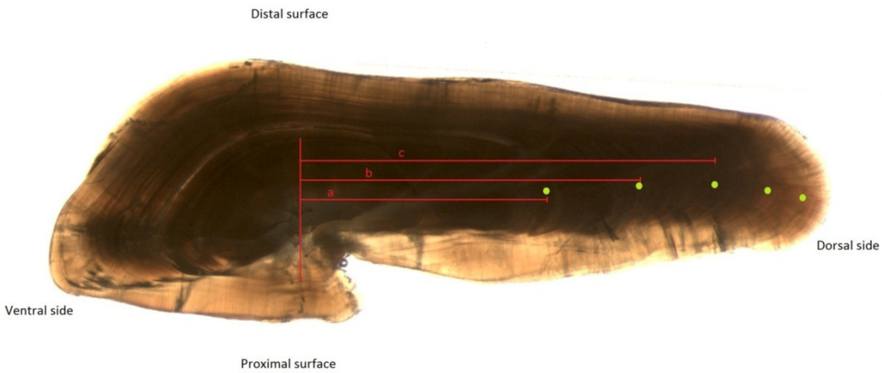

Figure A9.1. Transverse otolith section from *D.mawsoni* viewed under transmitted light and estimated to be 5+ years old. Green dots mark the position of the opaque zones (annuli) on the dorsal side of the section. Red lines indicate the average distance of the first three zones from the primordium a = 1.5mm, b = 2.1mm and c = 2.6mm.

In order to make sure early rings are included in age estimations it is preferable to conduct counts along the dorsal lobe starting from the primordium (or nucleus) and moving to the outer dorsal edge for the first 3 to 9 increments and then down towards the proximal surface (Figure A7.2) (Preliminary age estimates of *D.mawsoni* otoliths counted on the ventral lobe were found to consistently underestimate age (Figure A7.3) and this can largely be attributed to the readers missing early rings in their counts). On otoliths where it is particularly difficult to identify the first increment, the measurements seen in figure A7.1 can used to help locate the rough position of these early zones.

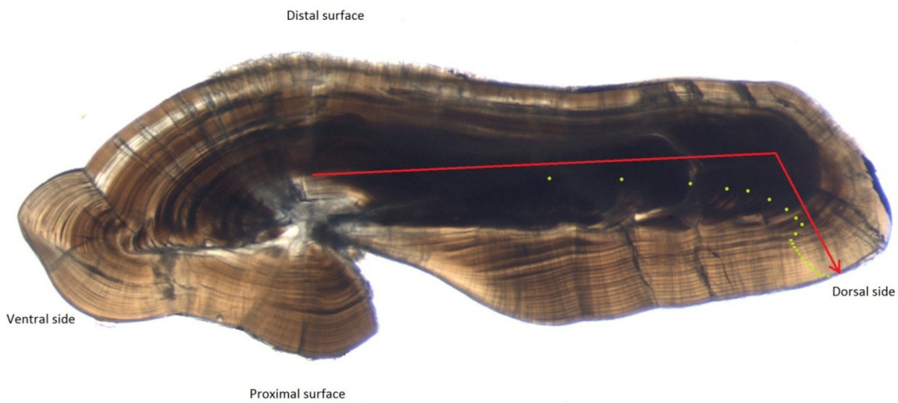

Figure A9.2. Transverse otolith section from *D.mawsoni* viewed under transmitted light and estimated to be 24+ years old. Green dots mark the position of the opaque zones (annuli) on the dorsal side of the section. Red line indicates the preferred path for counting increments.

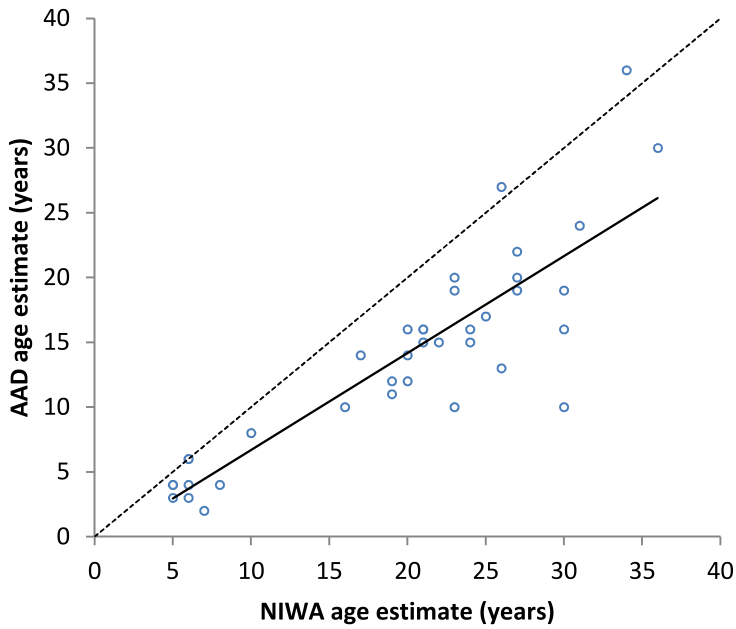

Figure A9.3. Age bias plot showing preliminary age estimates produced by AAD to be heavily biased towards under ageing *D.mawsoni* otoliths compared with age estimates from Peter Horn (NIWA).

Conclusions:

The AAD method produces images of Antarctic toothfish otolith structure that are equivalent to those used routinely by NIWA. Increments structures are more difficult to interpret consistently in the samples of this species. However, the development of a reference collection and standardized protocols to interpret sections is likely to lead to some improvements. Access to a broad range of fish size and ages would assist in this process, as would routine exchange of material between NIWA and AAD if high throughput processing for this species becomes a priority.