Age structure for rock cod *Patagonotothen ramsayi* from Falkland Island waters:

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1. Introduction

The age structure in a fish population provides the basic information for mortality rates, recruitment and growth (Hussy et al., 2016). These parameters are essential inputs in age-structured stock assessment models that provide the basis for management advice in many world fisheries (Lorenzen, 2016; Payne et al., 2005). Due to the key role of age in stock assessment models, bias in age estimates can therefore fundamentally influence the perception of the stock and fishing mortality, resulting in erroneous predictions of stock size and related management advice.

Rock cod *Patagonotothen ramsayi* is the most abundant species of the genus *Patagonotothen* in the Southwest Atlantic, inhabiting the Argentinean and Patagonian shelves from 35°58'S to the Burdwood Bank (55°S; Brickle et al., 2006). After the collapse of southern blue whiting (*Micromesistius australis*) between 2004 - 2007, its role was overtaken by rock cod, which exhibited a 20–30 fold increase in catches and CPUEs (Laptikhovsky et al., 2013). Although catches have since started to decline, the species remains an important component within the Falkland Islands ecosystem and forms the basis of extensive finfish fisheries within the region. Significant amounts of juveniles are also captured as bycatch in squid fisheries, although the true extent of this requires further investigation.

This annual report, presents a reliable ageing methodology for the construction of age length keys and estimation of growth and mortality parameters from samples samples obtained in the Falkland Islands during 2017. It also aims to provide estimates of inter- and intra-reader bias and precision in the age estimation in order to establish the reliability of the age estimation protocol and their potential use in stock assessment and subsequent management advice.

2. Methods

2.1. Data Collection

Rock cod were sampled by scientific observers and other scientific staff of the Falkland Islands Government Fisheries Department. Data were collected on board commercial fishing vessels operating bottom trawls under various license types. In

addition data were collected on board RV 'Castello' operating bottom trawls during research cruises.

Randomly sampled rock cod were measured to the nearest cm (TL), sexed and the stage of reproductive maturity assigned according to an eight-stage scale (I and II – immature, III and IV – maturing, V – mature, VI – running, VII – post spawning and VIII – spent). Each annual collection of otoliths are stored in paper envelopes in four quarterly time periods (A: Jan – Mar, B: Apr – Jun, C: Jul – Sep and D: Oct – Dec).

Otoliths for ageing are selected to cover the length distribution of sampled fish from each quarterly otolith collection. This ensures that sufficient otoliths are aged for all lengths on a temporal basis.

2.2. Preparation of otoliths

Otoliths were embedded in rows of five in blocks of amber coloured polyester resin and left to set for 24 hours. Fully dried blocks are ground in order to provide smooth linear surfaces and the nucleus marked using a pencil. This is undertaken in order to guide the cutting angle and ensure that sections are cut precisely at right angles. Resin blocks were subsequently sectioned using a Buehler Isomet Low Speed Saw. Between two and five sections of 0.35mm were taken per resin block and mounted on microscope slides under coverslips with clear polyester resin.

2.3. Reading methodology

Sections were viewed under reflected light at 20 to 40 times magnification. All sections of each row of otoliths were inspected and the section closest to the primordium was used for subsequent ageing. Images were taken for the best section for each otolith and enhanced to provide assistance in ageing.

Following previous work on age estimation of this species, the sector from the primordium to the proximal edge of the section, on the ventral side of the sulcus was chosen as the area in which to count increments. However, for some preparations, increments formed on the dorsal side were at least as clear as those on the ventral side. Each otolith was aged at least twice by the primary reader. For otoliths where the first two readings did not agree, a third reading was undertaken. All counts of

annuli were made without prior knowledge of fish size, date of capture or previous age estimates.

2.4. Precision of the age estimates

Repeated readings of the same otoliths provide a measure of intra-reader or interreader variability. They do not validate the assigned ages but provide an indication of size of the error to be expected with a set of age estimates, due to variation in interpretation of an otolith. Beamish and 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:

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

Where N is the number of fish aged, R is the number of times fish are aged, Xij is the ith determination for the jth fish, and Xj is the average estimated age of the jth fish.

An IAPE was calculated for all repeated readings undertaken by the primary reader.

2.5. Estimation of von Bertalanffy parameters

A von Bertalanffy growth function was fitted to the observed length-at-age data:

$$L_t = L_\infty \left(1 - e^{-K(t-t_0)} \right)$$

where L_t is length (TL in cm) at time t (years), L_{∞} the asymptotic length, K is the rate (year⁻¹) by which L_{∞} is approached, and t_0 is the theoretical age at length zero. Growth curves were fitted for males and females by non-linear least-square regression in R (R Core Team, 2018). Likelihood ratio tests were used to estimate whether a combined sex growth model or sex-separated models better described the length-at-age data. A parametric bootstrapping procedure with 1,000 iterations was employed to estimate 95% confidence intervals for final parameter estimates (Baty et al., 2015).

2.6. Mortality estimates

Catch-at-age frequency plots for each sex and fishery were obtained from representative length frequency data, by applying age-length keys derived from the aged otoliths (Ogle et al., 2018). The annual survival rate (S) and instantaneous rate of total mortality (Z) were estimated from catch-at- age data on the descending limb of the catch-curve using the Chapman- Robson approach (Chapman and Robson, 1960) with the variance estimate corrected for overdispersion as recommended by Smith et al. (2012). Initial ascending points representing fish that were not fully recruited to the fishery were excluded from the analysis.

3. Results and discussion

3.1. Distribution of samples

Biological information was obtained from a total of 70927 rock cod samples. Of these, 33915 were females 35444 were males, 1537 were juveniles and 31 were not identified for sex. Rock cod were sampled from across the shelf at depths between 58 and 470 m depth (Mean = 196.07 m; Figure 1).



Figure 1: Positions from which rock cod were sampled around the Falkland Islands during 2017 (n=70927).

3.2. Length and Age composition

Lengths for rock cod ranged between 6 and 38 cm TL for males and between 6 and 40 cm TL for females. A clear modal peak occurred at 15 cm TL and a minor peak at 24 cm TL (Figure 2). A second minor modal peak occurring at 5 cm reflected new recruits entering the fishery at inshore regions to the south of the Falkland Islands.



Figure 2: Length frequency distribution for male (n=35444), female (n=33915), juvenile (n=1537) and unidentified sex (n=31) rock cod sampled around the Falkland Islands.

The age frequencies derived from the age length keys showed a unimodal distribution with a peak occurring for 2 year old fish (Figure 3). Ages ranged from 0 to 15 years for females and 0 to 13 years for males. There were few fish caught with ages greater than 6 years old. While there was an increase in the proportion of fish between 3 and 5 years old, this was not to the extent seen in samples prior to 2016 when a clear second modal peak was prevalent at 4 years of age.



Figure 3: Age frequency distribution for male (n=35444), female (n=33915), juvenile (n=1537) and unidentified sex (n=31) rock cod sampled around the Falkland Islands.

3.3. Otolith interpretation

Rock cod otoliths were variable in the deposition patterns of their annuli. Otoliths could occasionally be straightforward to age, with a clear first annuli, identified from juvenile fish (Figure 4A and B) and distinct banding extending to the margin. However, this was not always the case, and many section displayed what can be considered check or false annuli (Figure 4C).

Once the fish becomes a bit older, the outer annuli frequently become a bit more discernible and the earlier increments could subsequently be interpreted based on these later patterns (Figure 4D). The variability in otolith patterns may be due to differences in the environmental conditions encountered during their life; or differences in life-history strategies between different groupings of rock cod in the Falkland Islands waters (Brickle et al., 2006). Alternately, it may simply be a result of the actual otolith preparation or the angle that the otolith was sectioned.



Figure 4: Sectioned otolith from an (A) 8 cm male, (B) 14 cm male, (C) 30 cm female and (D) 32 cm female rock cod with estimated ages of 1, 2, 9 and 13 years, viewed at 25 x magnification.

3.4. Age and growth

Likelihood ratio tests indicated no significant differences in growth between male and female rock cod (χ^2 =4.37; *P*=0.224; AIC=2041.25). Likelihood ratio tests indicated no significant differences in the L^{∞} (χ^2 =3.92; *P*=0.051; AIC=2043.18), *K* (χ^2 =2.39; *P*=0.122; AIC=2041.64), or the t_0 (χ^2 =0.45; *P*=0.502; AIC=2039.7) parameter estimates for male and female fish. The AIC value for the combined sex model was 2039.62, further indicating that this was the best fitting model.

Calculated von Bertalanffy growth parameters and their 95% confidence intervals for the combined sex rock cod model are presented in Table 1 and Figure 5.



Figure 5: Length versus age with superimposed best-fit von Bertalanffy growth model and 95% confidence bands for rock cod sampled during 2017 (n=407).

Table 1: Von Bertalanffy parameters (with 95% confidence intervals) for rock cod sampled during 2017 (n=407).

Parameters	Estimate	LCI	UCI
L∞	38.73	36.96	40.74
К	0.20	0.21	0.27
t _o	-0.40	-0.56	-0.23
n	407		

3.5. Mortality estimates

The threshold age for the mortality estimates of rock cod was 2 years (Figure 6). The total annual survival and mortality rates were 51.9% and 0.66 year⁻¹ respectively (Table 2, Figure 6). Despite a slight decrease in the mortality rates from 2016 (S = 44.8%, Z = 0.80 year⁻¹), these were still higher compared to estimates obtained during 2015 (S = 60.1%, Z = 0.51 year⁻¹).



Table 2: Estimates of survival rate and total mortality for rock cod (n=45019) around the Falkland Islands during 2017 using the Chapman-Robson catch-curve estimator.

Figure 6: Catch at age for rock cod (n=70927) sampled around the Falkland Islands during 2017. The solid points were used to compute the Chapman-Robson estimates of S and Z.

3.6. Precision of the age estimates

The percentage agreement table indicates that multiple estimates of ages by the primary reader agreed for 61.18 to 70.76% of the fish otoliths, while between 25.80 and 33.41% differed by one year (Table 3). The APE was 6.23% and the ACV was 8.17% (Table 4). These indicate that precision in age estimates can be considered as reasonable. Precision estimates showed an improvement on those obtained from 2017 age estimates (APE = 11.81%).

	Age difference (%)					
	0	1	2		3	
Age v. Age1	70.7617	25.7985		3.1941		0.2457
Age v. Age2	67.8133	27.027		4.6683		0.4914
Age1 v. Age2	61.1794	33.4152		5.4054		0

Table 3: Percentage table of raw differences between multiple readings of rock cod otoliths (n=407).

Table 4: Precision indices for age estimates of rock cod. ASD = The average (across all fish) standard deviation of ages within a fish; ACV = The average (across all fish) coefficient of variation of ages within a fish using the mean as the divisor. AAD = The average (across all fish) absolute deviation of ages within a fish; APE = The average (across all fish) percent error of ages within a fish using the mean as the divisor.

n	R	Agreement (%)	ASD	ACV	AAD	APE	
407	3	51.35	0.33	8.17	0.25	6.23	

4. Conclusion

Results of the current study provide biological parameters for rock cod in the Falkland Islands for 2017. Our findings indicated an improvement in the age estimation difficulties that occurred during 2016. The prescribed ageing protocol therefore provided a reliable method for age estimation for the successful application of empirical age-length keys for the assessment of the rock cod stock. Results continue to indicate some degree of concern regarding the high number of juvenile fish and reduced numbers of larger, older rock cod within the finfish fishery. Mortality rates continue to be higher compared to estimates obtained prior to 2016.

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