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

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TABLE OF CONTENTS

1. I	ntroduction	1
2. N	1ethods	1
2.1	Data Collection	1
2.2	Preparation of otoliths	2
2.3	Reading methodology	2
2.4	Precision of the age estimates	3
2.5	Estimation of von Bertalanffy parameters	3
2.6	Mortality estimates	4
3. F	esults and discussion	4
3.1	Distribution of samples	4
3.2	Length and Age composition	5
3.3	Otolith interpretation	7
3.4	Age and growth	8
3.5	Mortality estimates	9
3.6	Precision of the age estimates 1	0
4. C	Conclusion 1	1
Refer	ences 1	2
Ackn	pwledgements 1	3

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). The species has therefore become 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 2015. 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 48130 rock cod samples. Of these, 23653 were females 24292 were males, and 185 were juveniles for which the sex could not be determined. Rock cod were sampled from across the shelf at depths between 75 and 403 m depth (Mean = 193.44 m; Figure 1).



Figure 1: Positions from which rock cod were sampled around the Falkland Islands during 2015 (n=48130).

3.2. Length and Age composition

Lengths for rock cod ranged between 7 and 40 cm TL for males and between 8 and 41 cm TL for females with two clear modes, occurring at 14 and 28 cm TL (Figure 2).



Figure 2: Length frequency distribution for male (n=24292), female (n=23653), juvenile (n=184) and unidentified sex (n=1) rock cod sampled around the Falkland Islands.

The age frequency distribution was derived from the age length keys. The age distribution for rock cod was bimodal with a major peak of 2 year old fish, likely from the 'Loligo' fishery and a secondary peak for 4 year old fish captured in the finfish related fisheries (Figure 3). Ages ranged from 0 to 15 years for females and 0 to 14 years for males. There were few fish caught with age's greater than 11 years old.



Figure 3: Age frequency distribution for male (n=24292), female (n=23653), juvenile (n=184) and unidentified sex (n=1) 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 and distinct banding extending to the margin (Figure 4A and B). 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) a 11 cm and (B) 25 cm male, and a (C) 26 cm and 29 cm female rock cod with estimated ages of 1, 3, 4 and 7 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 =3.61; *P*=0.307; AIC=4511.11). Likelihood ratio tests indicated no significant differences in the *L*[∞] (χ^2 =2.35; *P*=0.125; AIC=4501.05), *K* (χ^2 =3.43; *P*=0.064; AIC=4509.67), or the *t*₀ (χ^2 =3.34; *P*=0.068; AIC=4508.99) parameter estimates for male and female fish (AIC=4482.37 for null model).

Calculated von Bertalanffy growth parameters and their 95% confidence intervals for the combined sex rock cod 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 2015 (n=564).

Table 1: Von Bertalanffy parameters (with 95% confidence intervals) for rock cod sampled during 2015 (n=564).

Parameters	Estimate	LCI	UCI	
L∞	36.78	35.70	38.00	
κ	0.31	0.28	0.35	
t_O	-0.56	-0.71	-0.40	
n	564			

3.5. Mortality estimates

The threshold ages for the mortality estimates for rock cod was 1, respectively (Figure 6). The total annual survival (S) and mortality (Z) rates were 60.11% and 0.51 year-1 (Table 2, Figure 6).

Table 2: Estimates of survival rate and total mortality for rock cod (n=48130) around the Falkland Islands during 2015 using the Chapman-Robson catch-curve estimator.

Parameter	Estimate	Std. Error	95% LCI	95% UCI
S	60.11	0.14	59.83	60.39
Z	0.51	0.039	0.43	0.59



Figure 6: Catch at age for rock cod (n=48130) sampled around the Falkland Islands during 2015. 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 65.60 to 86.52% of the fish otoliths, while between 12.41 and 32.27% differed by one year (Table 3). The APE was 3.74% and the ACV was 6.48% (Table 4). These indicate that ageing precision was reasonable in rock cod.

	Age difference (%)				
	0	1	2	3	4
Age v. Age1	78.01	21.10	0.71	0.18	0.00
Age v. Age2	86.52	12.41	0.53	0.35	0.18
Age1 v. Age2	65.60	32.27	1.60	0.35	0.18

Table 3: Percentage table of raw differences between multiple readings of rock cod otoliths (n=564).

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
564	3	65.60	0.22	6.48	0.17	3.74

4. Conclusion

Results of the current study provide biological parameters for rock cod in the Falkland Islands for 2015. Our findings indicate that the prescribed ageing protocol provides a reliable method for age estimation for the successful application of empirical age-length keys for the assessment of the rock cod stock. Results indicate some degree of concern regarding the quantities of juvenile fish being captured in the squid fisheries, although further investigation is required.

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