

**Age structure of Patagonian
toothfish**

Dissostichus eleginoides

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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 fisheries yield models that provide the basis for management advice in many world fisheries (Ashford, 2002; Horn, 2002).

Methodology for age determination should be applied across the catch and over a sequence of years while minimizing sources of error (Morison et al., 1998). In addition, quantitative estimates of variability in age determination should be provided for incorporation into decisions on analyses and modelling and interpretation.

This annual report, therefore presents a reliable ageing methodology for Patagonian toothfish, *Dissostichus eleginoides*, to calculate growth parameters from samples obtained in the Falkland Islands during 2016. It also presented an assessment of the bias and precision in order to ensure error does not exceed a quality threshold.

2. Methods

2.1. Data Collection

D. eleginoides were sampled by scientific observers and other scientific staff of the Falkland Islands Government Fisheries Department. Data were collected on board licensed longliners as well as commercial trawlers operating bottom trawls under various license types. In addition data were collected on board RV 'Castello' operating bottom and semi-pelagic trawls during research cruises.

Randomly sampled toothfish were measured to the nearest cm (Lt), 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 six 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 10 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 saved for image enhancement and 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 preparation, increments formed on the dorsal side were at least as clear as those on the ventral side. A readability index of 1 – Easy, 2 – Medium and 3 – Difficult was assigned to each otolith. Each otolith was aged twice by the primary reader. All counts of annuli were made without prior knowledge of fish size, date of capture or previous age estimates.

2.4. Estimation of von Bertalanffy parameters

Von Bertalanffy growth parameters were estimated for male and female fish from the length-at-age data using non-linear least squares regression procedure using R software (R core Team, 2015). 95% confidence limits were calculated for the von Bertalanffy parameters using bootstrap methods with 1000 iterations. Differences in growth between male and female fish were estimated using likelihood ratio tests. The age structure of the total sample of *D. eleginoides* captured in 2016 was estimated by constructing an age-length key (ALK) using the FSA package in R (Ogle, 2016).

2.5. Precision of the age estimates

Repeated readings of the same otoliths provide a measure of intra-reader or inter-reader 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 (1987) have developed an index of average percent error (IAPE), which has become a common method for quantifying this variation. The IAPE is calculated as:

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

Where N is the number of fish aged, R is the number of times fish are aged, X_{ij} is the i th determination for the j th fish, and X_j is the average estimated age of the j th fish.

Chang (1982) suggested that precision should be measured by the mean coefficient of variation (ACV) which is defined as:

$$ACV = 100 * \frac{\sum_{j=1}^n \frac{S_j}{x_j}}{n}$$

where s_j is the standard deviation of the R age estimates for the j th fish.

An IAPE and ACV were calculated for all repeated readings undertaken by the primary reader. The distributions of the differences between repeat readings were also inspected as another indicator of ageing errors, and of any bias between readings. Precision of repeated age estimates was also examined using an age bias plot (Campana *et al.*, 1995).

3. Results and discussion

3.1. Distribution of Samples

During 2016, biological information was obtained from a total of 14 233 *D. eleginoides* samples. Of these, 10410 were collected from within the trawl based fishery while 3820 were collected from within the longline fishery. Otoliths were extracted from a total of 2667 fish of which 342 and 459 were processed for age determination from the trawl and longline fisheries respectively.

D. eleginoides captured in the trawl fishery over the Falkland Islands shelf, occurred at depths between 92 and 609 m depth (Mean = 231.52 m; Figure 1). Comparatively, longline caught *D. eleginoides* were captured at an average depth of 1304.40 m, ranging between 807 and 1827 m.

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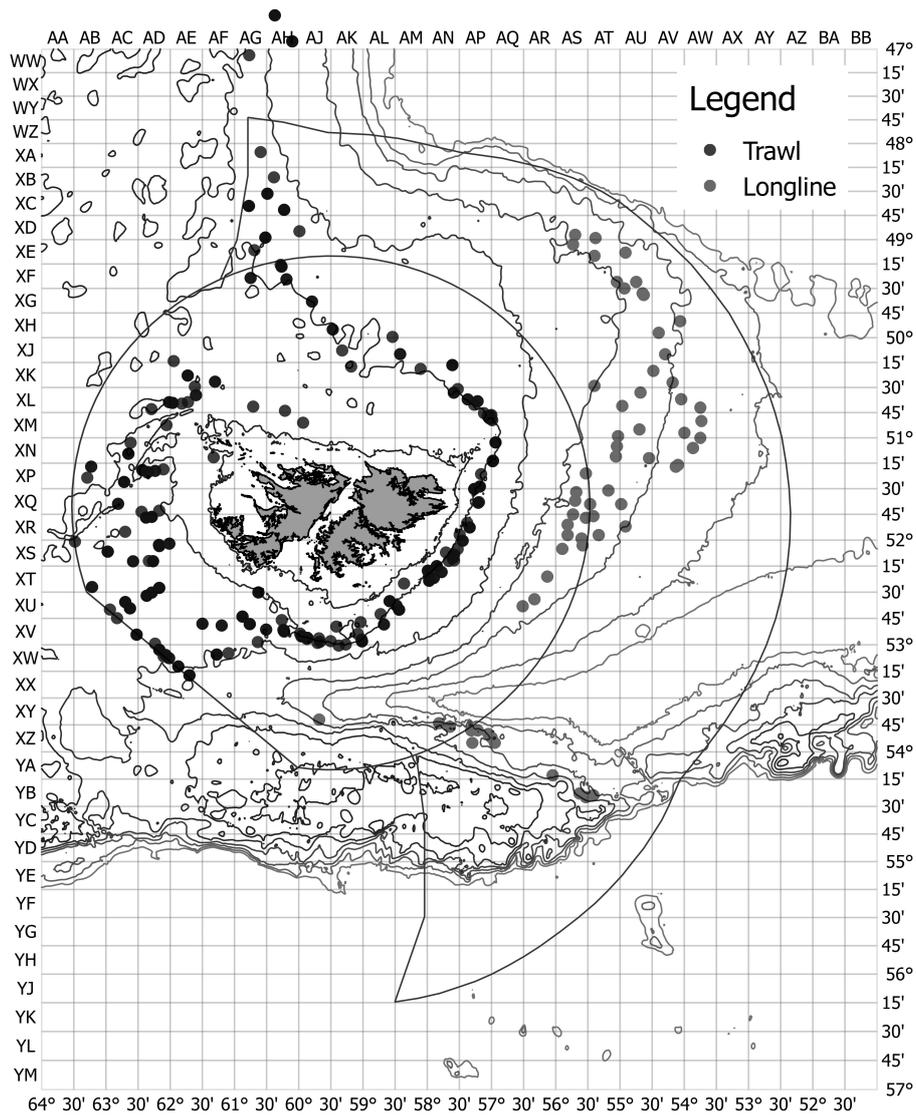


Figure 1: Positions from which *D. eleginoides* samples were obtained over the Falkland Islands shelf for 2016.

3.2. Length and Age composition

The length frequency distribution of *D. eleginoides* sampled from within the trawl-based fishery displayed two clear modes, occurring at 34 and 48 cm TL (Figure 2A). These two modes represented 1+ and 2+ aged fish (Figure 2B) A mode of 12 cm, consisting of juveniles, reflected 0+ aged fish being recruited into the trawl-based fishery. The largest fish captured was 106 cm TL, although only few fish larger than 74 cm TL (+4-5 years old) were captured within the trawl-based fishery.

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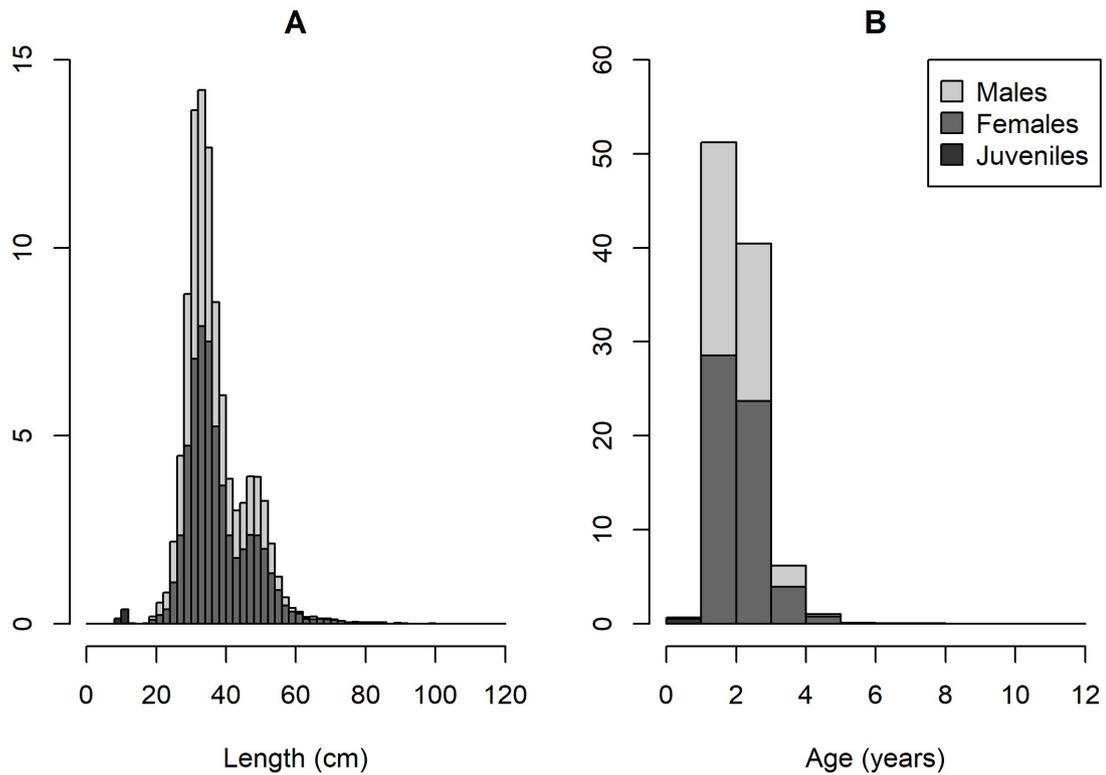


Figure 2: Length (A) and age frequencies (B) estimated from the total (aged and unaged) sampled catch of *D. eleginoides* captured in the trawl-based fishery as estimated from the age-length key (n = 10396).

The longline-based fishery appeared to target a different part of the *D. eleginoides* stock with lengths generally being greater than 60 cm TL (44 – 202 cm TL; Figure 3A) and ages greater than 5 years Figure 3B. The length frequency distribution for longline based catch was unimodal (95 cm TL) reflecting the slower growth occurring in the older (modal age = 8 years) more mature fish.

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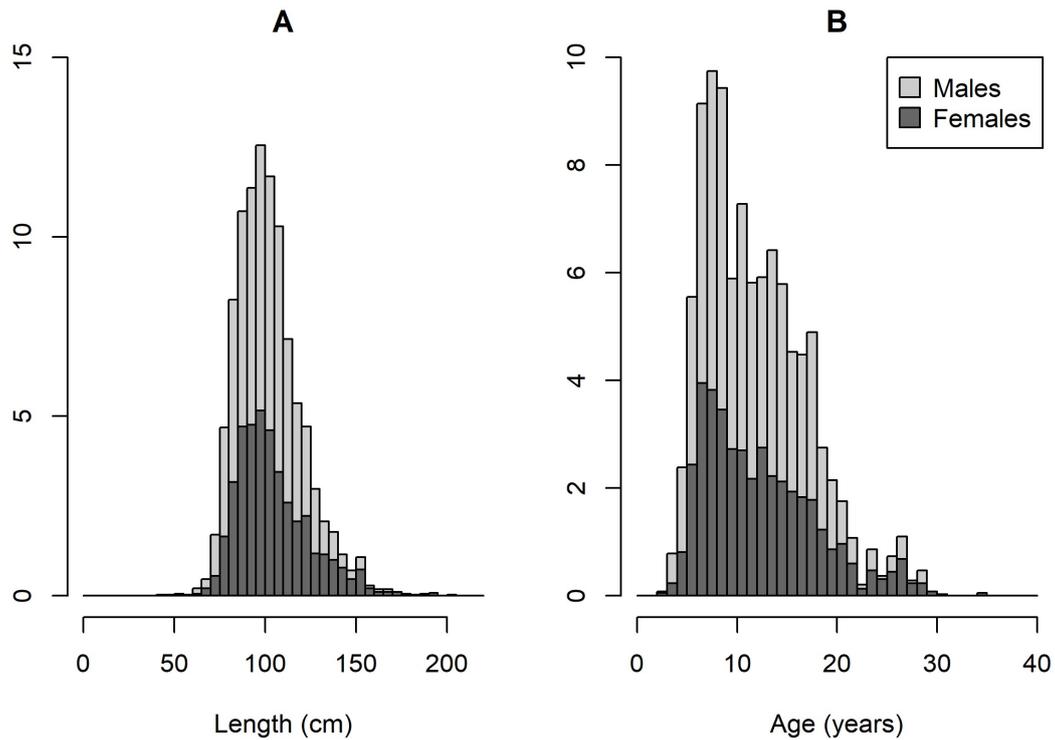


Figure 3: Length (A) and age frequencies (B) estimated from the total (aged and unaged) sampled catch of *D. eleginoides* captured in the longline-based fishery as estimated from the age-length key (n = 3820).

3.3. Size and Age

D. eleginoides ages ranged from 0 to 34 years Figure 4. Likelihood ratio tests indicated significant differences in growth between male and female *D. eleginoides* ($\chi^2=83.76$; $P<0.01$). Likelihood ratio tests indicated significant differences in the Linf ($\chi^2=81.94$; $P<0.01$) parameter for male and female fish. A representation of the age-length key for *D. eleginoides*, smoothed using a multinomial logistic regression model is presented in Appendix A. Calculated von Bertalanffy growth parameters and their 95% confidence intervals for male and female fish are presented in Table 1 and Figure 4.

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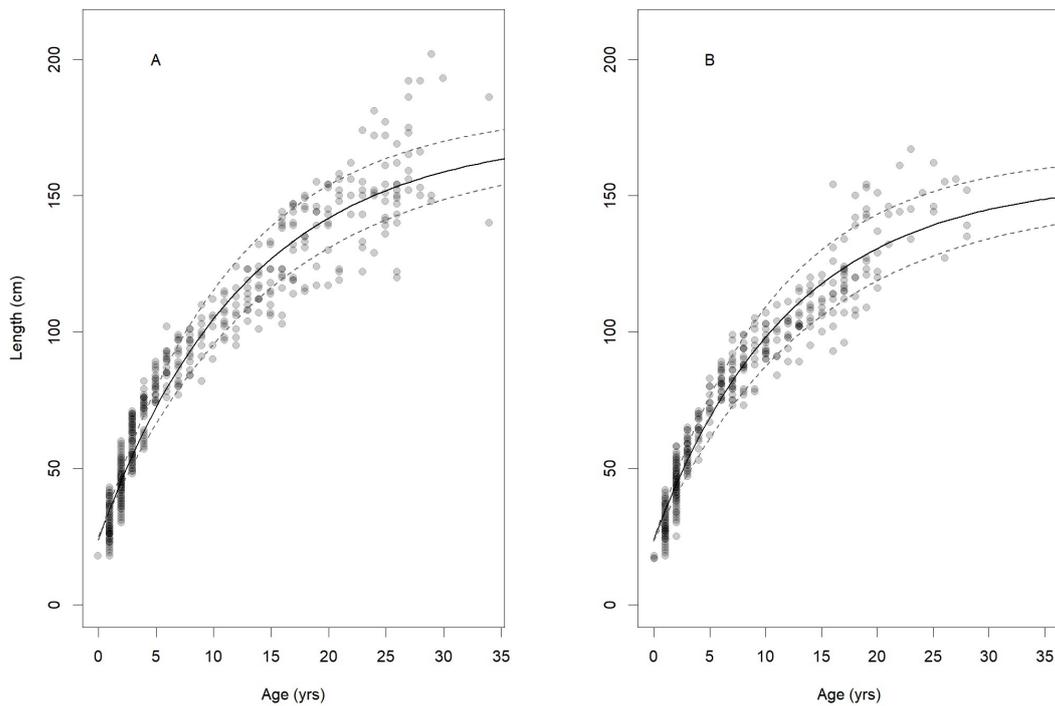


Figure 4: Length versus age with superimposed best-fit von Bertalanffy growth model and 95% confidence bands (dashed lines) for (A) female and (B) male *D. eleginoides* sampled during 2016

Table 1: Von Bertalanffy parameters (with 95% confidence intervals) for *D. eleginoides* sampled during 2016.

Parameters	Estimate	Std Error	LCI	UCI
Females				
Linf	172.97	3.36	166.57	180.63
K	0.078	0.0042	0.070	0.087
t0	-1.96	0.17	-2.33	-1.61
n	437			
Males				
Linf	156.55	4.58	148.89	165.27
K	0.082	0.0060	0.072	0.092
t0	-1.99	0.20	-2.37	-1.67
n	365			

3.4. Precision of the age estimates

The percentage agreement table indicates that multiple estimates of ages by the primary reader agreed for 61.22% of the fish, 19.33% differed by one year and 7.36% differed by two years (Table 2). Figure 5 shows that bias between the two age readings was normally distributed across the age range. The exception occurred in older fish in which the second age estimate was generally higher in comparison with the accepted age estimate. The APE was 3.80% and the ACV was 5.38% (Table 3). These indicate that ageing precision was high in *D. eleginoides* although the age-bias plot indicates difficulty in ageing of older fish.

Table 2: Percentage table of raw differences between multiple readings of *D. eleginoides* otoliths for 2016.

Years	0	1	2	3	4	5	>=6
Frequency (%)	61.22	19.33	7.36	4.11	2.74	2.37	2.87

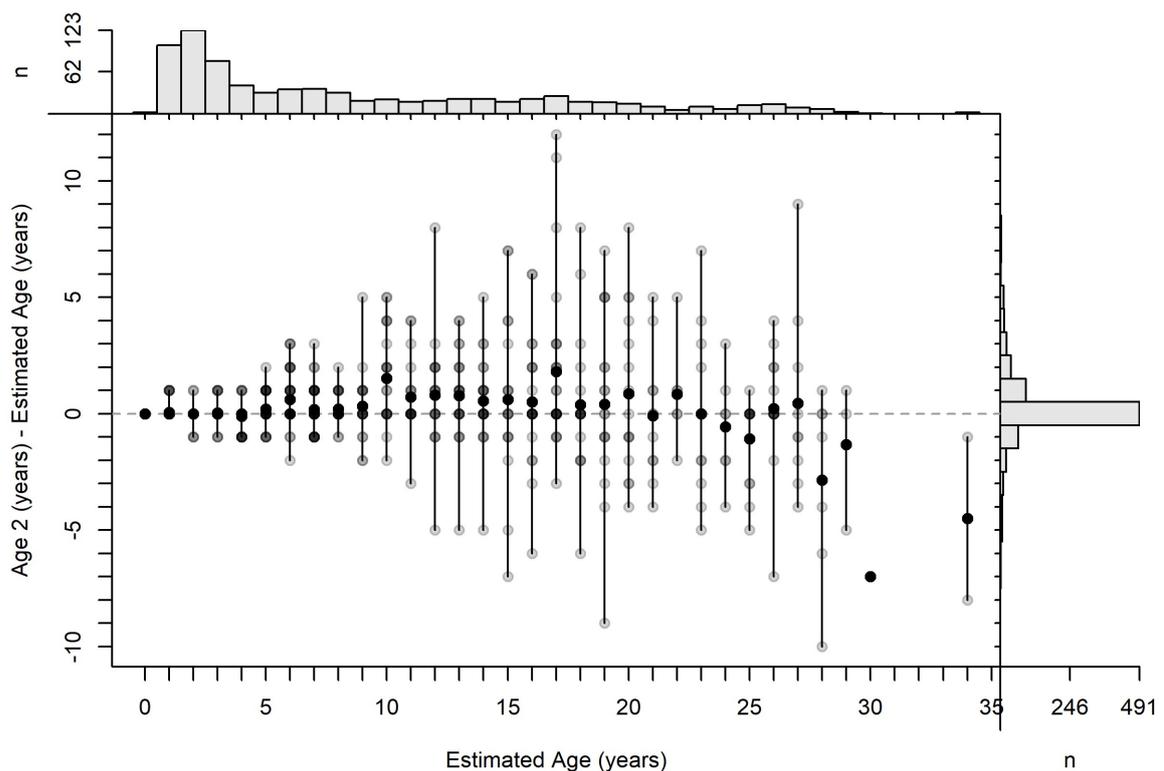


Figure 5: Age-bias plot for comparing the mean estimated ages from multiple readings of *D. eleginoides* for 2016

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Table 3: Estimates of average coefficient of variation and average percent error for multiple ageings of *D. eleginoides* otoliths for 2016.

n	Valid n	R	ACV	APE	Percent agreement
802	802	2	5.38	3.80	61.22

4. References

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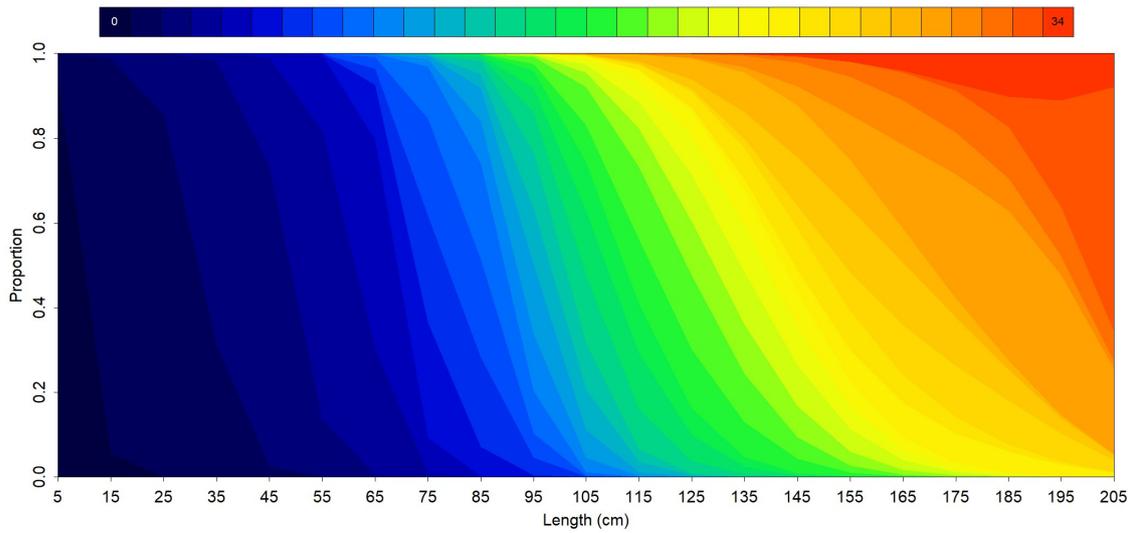
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5. Acknowledgements

Thank you to the Scientific Fisheries Observers of the Falkland Islands Government Fisheries Department for the collection of biological data.

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6. Appendix A



Area plot representation of the observed age-length key for *D. eleginoides* sampled during 2016 and smoothed using a multinomial logistic model. The area of each circle is proportional to the proportion of fish in a length interval that are a given age.