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Age Readings of *Sebastes mentella* Otoliths: Bias and Precision Between Readers

by

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Abstract

This study presents comparative otolith-based age readings of pelagic redfish (*Sebastes mentella*) from the Irminger Sea within an otolith exchange program between institutes in Germany, Iceland and Norway. 213 otoliths were thin-sectioned and read independently in the participating labs. Age reading results were compared between readers in terms of bias and precision, using a set of statistical tests and graphical methods. Significant bias was observed between readers, mainly caused by deviations between age scores in the higher ages (> 20 years). Precision estimates, considering the high longevity of *S. mentella*, were relatively good compared to age readings of other long-lived species. In contrast, the age dependent percent agreement was poor (< 20%) for a tolerance level of ± 0 years, particularly for the age range 21-40 years, which represents the major fraction of the fished stock. A tolerance level of ± 5 years, however, lead to around 90% agreement for the age ranges up to 20 years. The fit of age reading scores with the von-Bertalanffy growth curve was relatively good, showing good correspondence between readers. A comparison of growth parameters with age-length relationships reported for demersal *S. mentella* in shelf areas in the Northeast Arctic, around Greenland and on the Flemish Cap showed that pelagic *S. mentella* generally exhibits slower growth. In the age range up to 10 years, however, relatively large fast-growing juveniles seem to be present, probably having recruited from the higher productive shelf areas off East Greenland and Iceland. The observed problems in bias and precision of age readings should to be improved by continuing with similar *Sebastes* otolith exchange programs and setting up a further age reading workshop to harmonise the interpretation of growth structures.

Keywords : Redfish, deep-sea redfish, *Sebastes mentella*; age reading, age determination; bias, precision, percent agreement; longevity.

Introduction

Age determinations provide essential input data for the stock assessment of marine fish stocks. The age-based stock assessment of redfish (*Sebastes* spp.) in the North Atlantic, however, proved to be difficult due to the lack of a sufficient amount of reliable age readings. The reliability of a set of hard body structures of the fish was addressed several times in the past. Various studies (e.g. Chilton and Beamish 1982, Nedreaas 1990) and workshops (e.g. ICES 1991, ICES 1996) have shown that the otoliths are the preferred structure for age readings

on North Atlantic *Sebastes* species due to an underestimation of older ages using scales and difficulties in the interpretation of other structures such as fin rays or vertebrae. Most laboratories are still reluctant to implement routine age readings on otoliths of *Sebastes* since there are concerns about the error observed in age reading results.

Age reading error has two major elements: bias and precision. The **bias** of age readings is caused by a consistent deviation of reading results between readers and is skewed from the mean to one side or the other, while **precision** of age readings measures the closeness of repeated independent age estimates (Wilson *et al.* 1987, ICES 1996). Precision reflects the degree of agreement among readers and is not to be confused with accuracy, which relates to the agreement with the true age of the fish.

Although there are routine testing systems and procedures for the assessment of bias and precision of age readings available (Kimura and Lyons 1991, ICES 1994, Campana *et al.* 1995, Hoenig *et al.* 1995, C.A.R.E. 2000), a broad-scale application of these methods in the laboratories carrying out redfish age readings is still missing. The 1995 ICES Workshop on Age Reading of *Sebastes* spp. (ICES 1996) showed a considerable bias between readers, which was shown to be improved after discussion of general interpretation of growth structures on the sectioned otoliths. Thus, the need for further exchange of material and knowledge on age reading methods was stressed.

As part of the four-year research project “Population structure, reproductive strategies and demography of redfish (Genus *Sebastes*) in the Irminger Sea and adjacent waters (ICES V, XII and XIV; NAFO 1)”, funded by the European Union (QLK5-CT1999-01222), an otolith exchange between redfish age reading experts of the participating institutions is being carried out to evaluate differences in age readings between readers and otolith preparation methods. The otolith exchange in 2000 was based on *Sebastes marinus* from the Icelandic shelf (Stransky *et al.* 2001). In 2001, the otolith readers comparison focused on pelagic *S. mentella*, sampled in the Irminger Sea during summer 1999. The age reading results of this otolith exchange are presented in this study, with regard to bias and precision between readers.

Materials and Methods

The otoliths used in this study were collected from *S. mentella*, caught within a commercial sampling program (EU Study 97/004 “Sampling of 8 German commercial fisheries”) in July 1999, onboard the German F/V *Fornax*. The otoliths were taken from twelve hauls in the Irminger Sea (ICES sub-area XII) at mean depths of 235-285 m. 213 otolith pairs were selected for age determination, covering fish of 22.5-41.5 cm in total length. From each pair, one otolith was prepared for age reading using the thin-sectioning technique described by Bedford (1983), while the other otolith was kept for later radiometric age validation (*e.g.* Campana *et al.* 1990).

The otolith thin-sections were carried out at the Institute for Sea Fisheries of the Federal Research Centre for Fisheries in Hamburg/Germany. A diamond-covered saw blade of 0.3 mm thickness and 100 mm diameter, rotating at 6000 rotations/min, was used to cut cross-sections of 0.5 mm thickness. The cross-sections were embedded onto glass plates with translucent polyester resin and read with a magnification of 25-30x using polarised transmitted light. These otolith cross-section plates were read at the Marine Research Institute in Reykjavík/Iceland with transmitted light, magnified 40x. At the Institute of Marine Research in Bergen/Norway, the plates were read through transmitted light using a magnification of 20-35x.

For the comparison of bias and precision between readers, a set of statistical tests and graphical methods were applied. **Bias** estimates were based on simple linear regression analysis, the parametric paired *t*-test and the nonparametric Wilcoxon matched-pairs rank test (Conover 1998, Hollander and Wolfe 1999). The slope and intercept of simple linear regressions are tested for significant differences ($\alpha = 0.05$) from 1.0 and 0, respectively. The parametric paired *t*-test and the nonparametric Wilcoxon matched pairs rank test are used to detect significant differences from a paired difference of 0. Error terms are 95% confidence limits. Age bias plots (ICES 1994, Campana *et al.* 1995, Eltink 1997) visualise the deviation of the age scores of two readers or methods from the 1:1 equivalence line and also allow the detection of non-linear bias patterns, *e.g.* the underestimation of ages by one reader in one part of the age range and overestimation in another part of the age range. The mean age assigned by one reader for all fish assigned a given age by the second reader is presented including the standard deviation around the mean.

Various measures for **precision** were suggested for the comparisons of age readings. One of the more common indices is the percent agreement that compares the percentage of age determinations that are in agreement within a specified number of years. This index, however, does not evaluate the degree of precision equally for all species. If for example 95% of the age readings agree within a range of ± 1 year for cod (*Gadus morhua*), this could be a very poor precision since there are just few year-classes in the fishery. For *S. mentella*, a 95% agreement within a tolerance range of ± 5 years could represent a good precision, given 70-80 years longevity and 30-40 age groups present in the fishery. Beamish and Fournier (1981), therefore, suggested an average percent error, which is dependent on the average age of the fish species observed. Chang (1982) modified this index to a coefficient of variation, substituting the absolute deviation by the standard deviation from the mean age. Besides these indices, the correlation coefficient r^2 is given to evaluate the fraction of variation explained by the linear relationship between readers or otolith preparation methods.

Results

Three age readers from institutes in Germany, Iceland and Norway were participating in the *S. mentella* otolith exchange in the year 2001 (Table 1).

As indicated by the age bias plots (Figure 1), all between-reader comparisons are subject to a certain degree of bias. In all three cases, the deviation from the 1:1 equivalence line is non-linear. Particularly in the age range 20-30 years, readers 2 and 3 assign higher ages than reader 1. The comparison of readers 2 and 3 shows high variation in ageing scores for fish assigned ages of 45 years and more. Table 2 presents the statistical tests applied to the comparison of readers in terms of bias. Regression analysis as well as the nonparametric Wilcoxon test and the parametric paired *t*-test show high significance levels, indicating bias between readers in all three comparisons, apart from the intercept for reader 1 versus reader 3 which is close to zero. The overall bias between readers 1 and 2 amounts to -2.2 years (mean paired difference). This overestimation of ages by reader 2 relative to reader 1 is also indicated by the positive intercept (1.9 years). The opposite is true for the comparison of readers 2 and 3, where reader 3 assigns ages 1.5 years higher than reader 2 for the same fish, coinciding with a negative slope (-1.6 years). In all three reader comparisons shown in Figure 1, no general trend in increasing standard deviations around the mean with increasing age was visible, as would have been expected from the *S. marinus* otolith exchange (Stransky *et al.* 2001).

From the precision estimates between readers (Table 3), the correlation coefficient, the coefficient of variation and the average percent error show relatively good agreement, whereas the percent agreement value is comparatively low. As pointed out earlier, the percent agreement index does not take account of the mean age, as the average percent error does. A 7-19% agreement of age determinations between readers, as in this case, might be very low compared to values achieved for age readings of species with a shorter life span. For redfish, however, a wider age range has to be considered when comparing percent agreement results with age readings for other fish species. By increasing the tolerance level of agreement between readers, as illustrated in Figure 2 ("All"), a percent agreement of 73% is reached with a tolerance of ± 3 years, and around 90% are achieved with ± 5 years, considering the whole age range. When dividing the percent agreement plots into age ranges of 10 years (Figure 2), the fractions of the life span of *S. mentella*, where most of the ageing errors occur, become visible. The curves for all reader pairs are changing from asymptotic to linear with increasing age range, showing that for ages of >20 years, tolerance levels of $\pm 1-2$ years only lead to moderate improvements in the percent agreement and even tolerating $\pm 4-5$ years does not cause more than 90% agreement. The comparison of precision indices between pairs of readers (Table 3) shows considerably better values for reader 1 versus reader 3 than for the other reader pairs. This better agreement is particularly present in the age ranges up to 20 years, as visualised in Figure 2.

Discussion

All reader comparisons in this otolith exchange showed a considerable error in terms of bias and precision. Major differences in ageing scores between readers (standard deviations up to 6.5 years) were observed within the 21-40 years age range, representing most of the fished stock of *S. mentella* in the Irminger Sea. Although the mean difference in age readings reached up to 2.2 years, an age-based assessment of this stock still seems to be difficult. Particularly the interpretation of growth zones around the nucleus and following the transition zone is probably causing the observed error.

Considering age reading comparisons of previous otolith exchanges (*e.g.* Villamor 1995, Eltink 1997, Bergstad *et al.* 1998), the age-dependent precision estimates (coefficient of variation, average percent error) applied in this study show relatively good reproducibility. The observed CV and APE values are slightly higher than for the previously conducted *S. marinus* otolith exchange (Stransky *et al.* 2001) and indicate medium precision, compared with studies on other long-lived fish species (*e.g.* Kimura and Lyons 1991, Stevenson and Secor 1999, Horn 2002). Within an otolith exchange between Canadian and US ageing labs for Pacific *Sebastes* species with similar longevity, mean CVs of 8.2-12.2% and APE of 5.7-9.1% were calculated (C.A.R.E. 2002). In contrast, the percent agreement does not exceed 20% which is relatively poor compared to age reading results of short-lived species (*e.g.* Corten 1993). Only at a tolerance of ± 5 years, around 90% agreement is reached, looking at the whole age range. 100% agreement is only attained in the age range 0-10 years with a tolerance level of ± 4 years. This represents a relatively poor fit and should be sought to be improved in future. It should be noted that there were no data available for the age range of 0-4 years to allow a conclusive evaluation of the age reading precision in the younger ages. These juvenile redfish are lacking in the Irminger Sea completely, since they most likely inhabit demersal nursery grounds on the East Greenland and Iceland shelf before recruiting into the pelagic occurrences (Stransky 2000).

Age-length relationship and growth

The fit of the von-Bertalanffy growth functions shows relatively good correspondence between readers, indicating that the observed error in ageing scores do only lead to small deviations in the age-length relationships. Although maximum ages of around 70 years were reported for *S. mentella* (Campana *et al.* 1990, Nedreaas 1990), only a “window” of ages seems to be present in the Irminger Sea, consisting of fish with a length range of about 20-50 cm (*e.g.* Rätz 2001, ICES 2002). The lack of data for fish <22 cm in this case contributes largely to relatively low t_0 values of the age-length relationships (Figure 3). An additional error might be introduced by the presence of relatively large fast-growing juveniles that have been recruiting from the higher productive shelf areas off East Greenland and Iceland. Due to our results, a 25 cm pelagic *S. mentella* would be around 6 years old, while in shelf areas of the Flemish Cap (NAFO 3M), in the Northeast Arctic (ICES I, II) and off East Greenland (ICES XIVb), demersal *S. mentella* of the same size would be 7, 8 and 8.5 years old, respectively (Saborido-Rey 1995 and 2001, Nedreaas 1990, Kosswig & Rätz 1995). Since the L_{inf} and k values were reported to be higher for the shelf areas, faster growth of demersal *S. mentella* compared to pelagic *S. mentella* was observed for ages of >10 years. This recruitment effect was not that pronounced in a set of age readings carried out by reader 3 onboard the Norwegian research vessel *G.O. Sars* during the international survey on pelagic redfish in the Irminger Sea in June/July 2001 (Fig. 4). Age readings by reader 2 on *S. mentella* otoliths collected on the 1999 international survey, divided by “oceanic” and pelagic “deep-sea” types, illustrate that the material exchanged in this study most probably represents the “oceanic” type inhabiting shallower depths (Fig. 5).

Further redfish otolith exchange

The redfish otolith exchange in 2002 is based on *S. mentella* otoliths, collected during the German groundfish survey off East Greenland in autumn 1998 and on the international survey on pelagic redfish in the Irminger Sea in July 1999. This material is of particular interest, since a considerable recruitment of juvenile *S. mentella* from East Greenland into the Irminger Sea has been observed (Stransky 2000) and the corresponding year-classes still have to be identified. One otolith of each pair will be thin-sectioned at the Institute for Sea Fisheries in Hamburg, while the other one will be used for radiometric age validation (*e.g.* Campana *et al.* 1990). Digital pictures of the sectioned otoliths will be exchanged together with the otolith plates to allow readers to set annulus marks and to evaluate interpretation differences.

As stated in the report of the most recent *Sebastes* age reading workshop (ICES 1996), there is further need for exchange of otoliths and knowledge on the interpretation of growth structures. The best forum to do so would be a further age reading workshop in the frame of ICES, which is planned for 2003 or 2004.

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Table 1. Participating age readers.

Reader ID	Name	Institute
1	Christoph Stransky	Federal Research Centre for Fisheries, Institute for Sea Fisheries, Hamburg, Germany
2	Sif Guðmundsdóttir	Marine Research Institute, Reykjavík, Iceland
3	Svend Lemvig	Institute of Marine Research, Bergen, Norway

Table 2. Statistical tests for the detection of **bias** for age readings of *S. mentella* between readers.

Statistic	Age reader		
	Reader 1 versus Reader 2 (N = 191)	Reader 1 versus Reader 3 (N = 213)	Reader 2 versus Reader 3 (N = 191)
	Regression		
Slope	1.016 ± 0.017	1.055 ± 0.017	1.006 ± 0.017
<i>P</i>	0.000	0.000	0.000
Intercept	1.903 ± 0.396	-0.348 ± 0.408	-1.632 ± 0.448
<i>P</i>	0.000	0.395	0.000
	Wilcoxon test		
<i>P</i>	0.000	0.001	0.000
	Paired <i>t</i>-test		
Mean paired difference	-2.215 ± 0.412	-0.808 ± 0.431	1.492 ± 0.443
<i>P</i>	0.000	0.000	0.000

Table 3. Measures of **precision** for age readings on *S. mentella* between readers.

Statistic or index	Age reader		
	Reader 1 versus Reader 2 (N = 191)	Reader 1 versus Reader 3 (N = 213)	Reader 2 versus Reader 3 (N = 191)
Correlation coefficient (r^2)	0.951	0.950	0.947
Coefficient of variation (%) ^a	11.22	8.16	11.06
Average percent error ^b	7.93	5.77	7.82
Percent agreement	6.81	19.25	11.52

^a from Chang (1982)^b from Beamish and Fournier (1981)

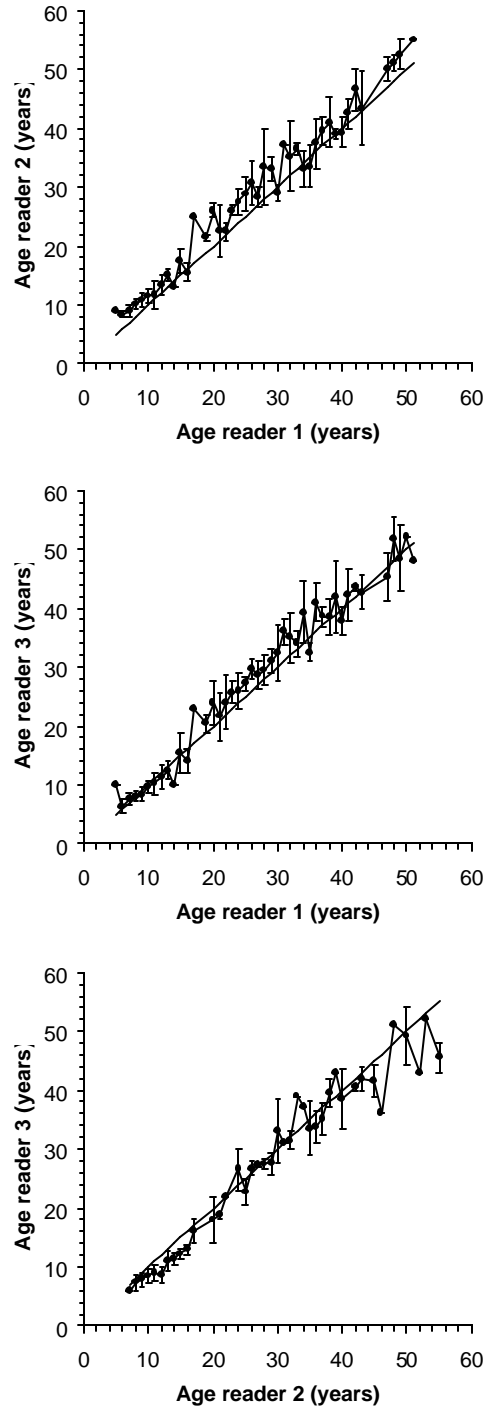


Fig. 1. Age bias plots for the reader comparisons given in Table 2. Each error bar represents the standard deviation around the mean age assigned by one reader for all fish assigned a given age by the second reader. The 1:1 equivalence (straight line) is also indicated.

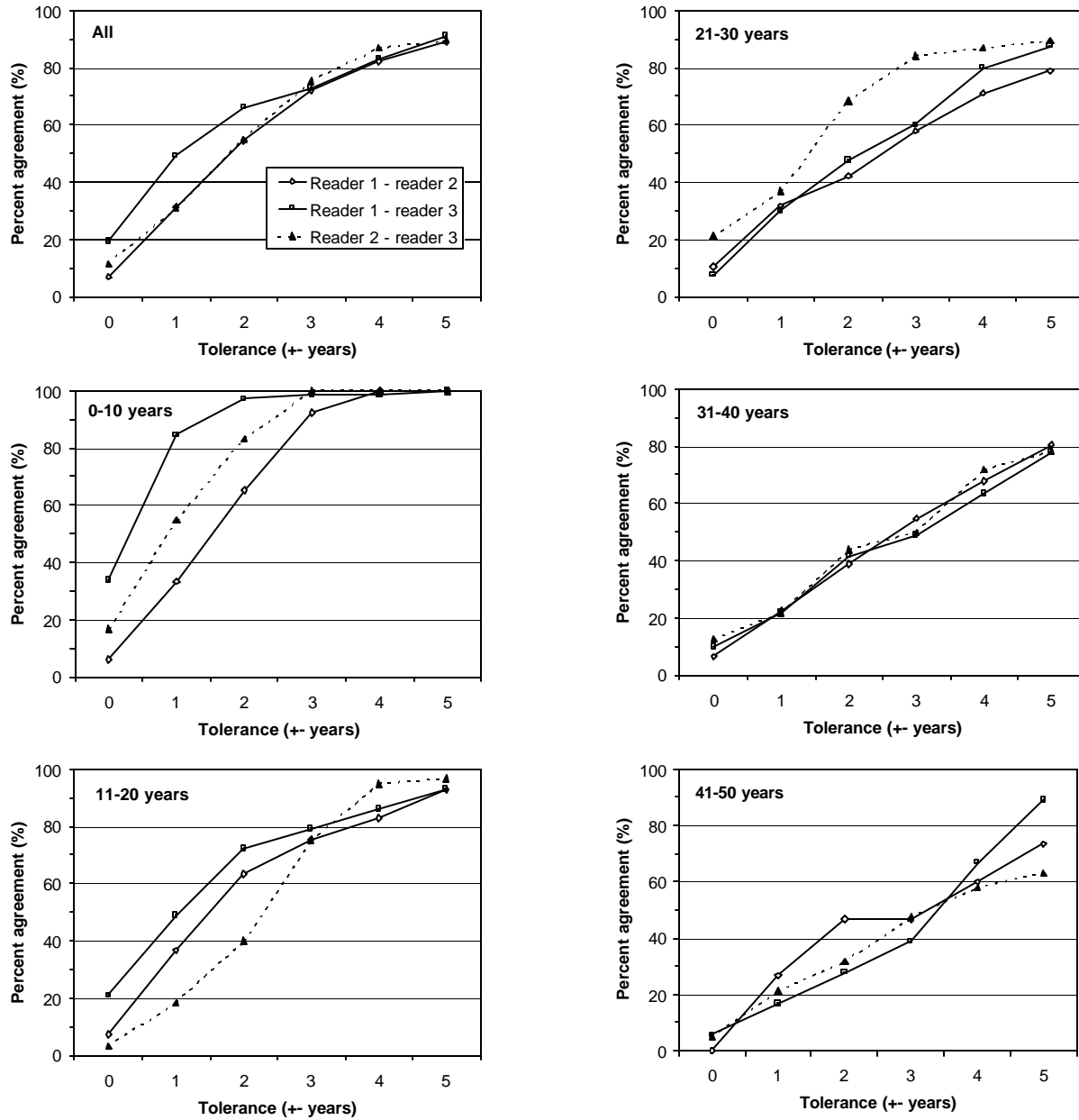


Fig. 2. Percent agreement for the reader comparisons given in Table 3 for a tolerance level (deviation of assigned ages between both readers) of ± 0 (total agreement) to ± 5 years, applied to all age groups and sub-sets of age ranges assigned by the first reader.

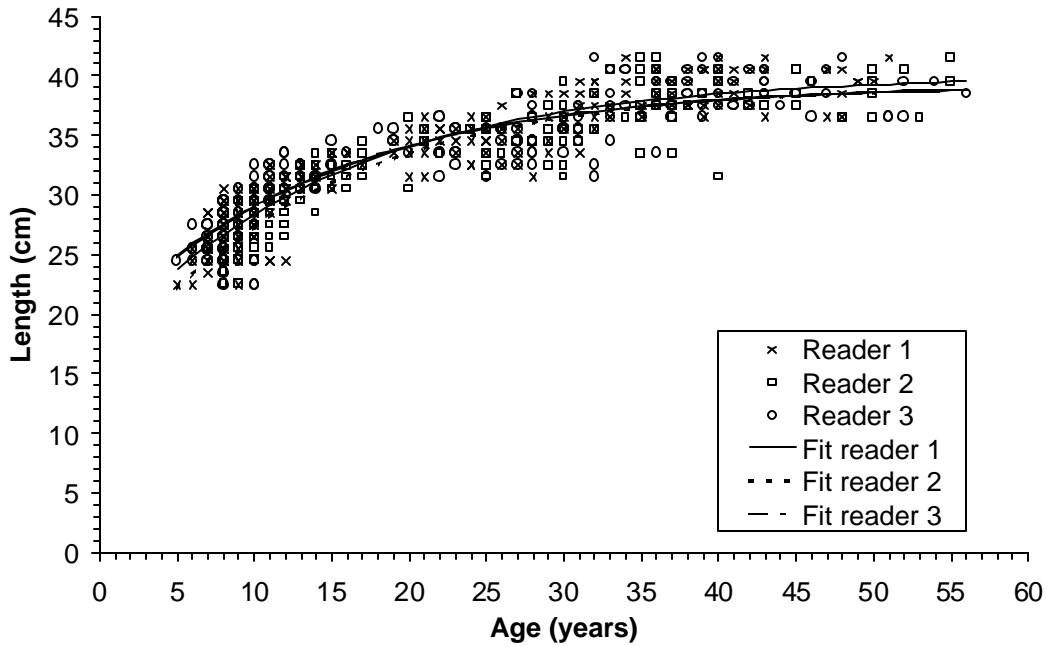


Fig. 3. Age-length relationship and fitted von-Bertalanffy growth curves of the comparison of readers. The fitted growth parameters are:

Reader 1: $L_{\text{inf}} = 40.081$ cm, $k = 0.066$, $t_0 = -8.531$ years

Reader 2: $L_{\text{inf}} = 39.227$ cm, $k = 0.073$, $t_0 = -6.379$ years

Reader 3: $L_{\text{inf}} = 39.274$ cm, $k = 0.069$, $t_0 = -9.635$ years.

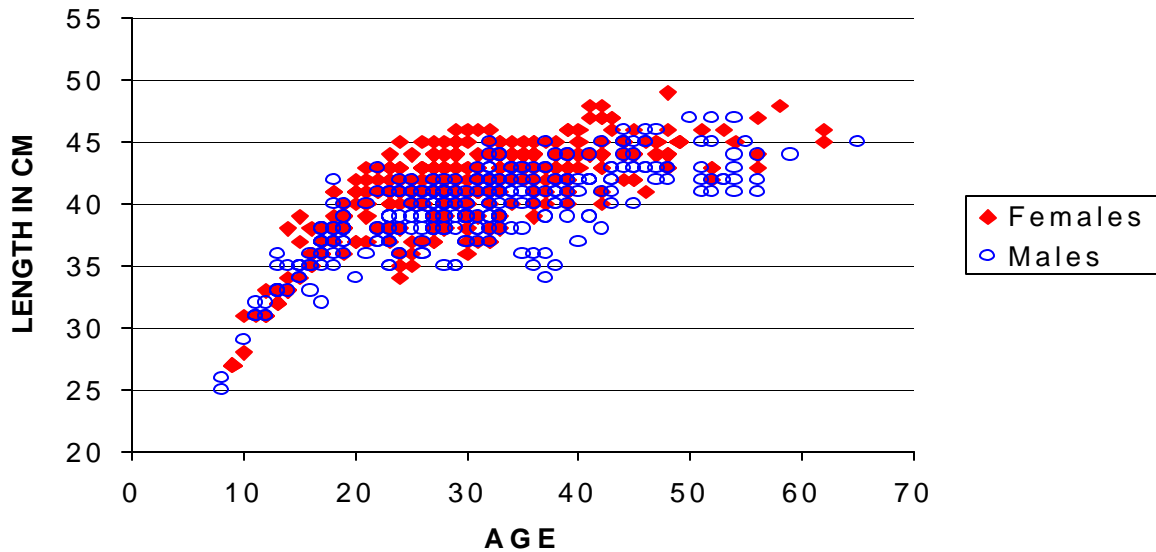


Fig. 4. Age readings for pelagic *S. mentella*, carried out by reader 3 onboard R/V *G.O. Sars* during the international survey on pelagic redfish in the Irminger Sea in June/July 2001.

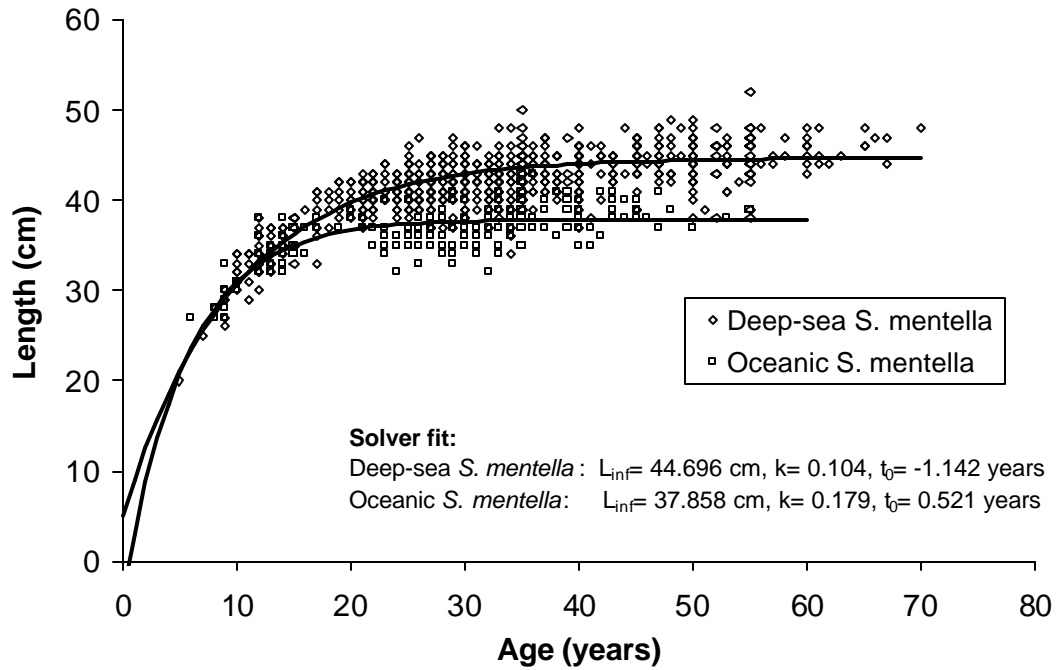


Fig. 5. Age readings for pelagic *S. mentella*, carried out by reader 2 on material collected during the international survey on pelagic redfish in the Irminger Sea in June/July 1999.